Annex 3. – For information - Aquatic Animals Commission's responses to comments considered

General Comments

Reference	Comment	Aquatic Animals Commission Response
General_1_Australia	**Category:** General **Proposed amended text:** N/A **Rationale**: Australia commends the WOAH AAHSC for its ongoing extensive work in improving the scientific rigor, clarity, consistency and accessibility of information pertinent to the international standards for aquatic animal health management. A collective awareness and consensus of understanding continues to be developed among members due to WOAH's thorough engagement and consultation with members during the revision process for these standards. The ability of members to apply appropriate biosecurity management strategies (for example, the application of disease free compartments, the application of zoning for the eradication and control of disease, and, the application of fallowing between production cycles) to facilitate the safe trade in aquatic animals and aquatic animal products continues to be greatly improved by the current revision of these standards. Australia encourages and supports the commissions ongoing workplan for 2025-2027, as documented in Annex 4. **Supporting evidence:** N/A	Noted.
General_2_EU	**Category:** General The EU supports the work programme	Noted.

Comments on the work programme:

Work_1_Brazil	**Category:** General **Proposal/Comment:**	The *Aquatic Manual* chapter on TiLV was successfully developed and is being circulated for Member comments in the

<table>
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<td>Brazil wishes to acknowledge and commend the significant efforts undertaken by the AAHSC in updating and enhancing the Aquatic Animal Health Code and Manual. Nevertheless, we respectfully propose a change in priority regarding item Ch. 2.3.X. Infection with tilapia lake virus, which is currently under priority level 3, suggesting that the AAHSC allocate additional efforts to addressing this technical gap and raise the priority to level 2.

Rationale:
Tilapia lake virus (TiLV) was listed in 2022; however, a dedicated chapter on this infection has not yet been included in the Aquatic Animal Health Manual.

While a technical card on the subject is available, it does not provide the level of detail expected in the Aquatic Manual chapters.

This gap may lead to inconsistencies in criteria related to surveillance and diagnostic methodologies.

Currently, there is no clear definition of a confirmed case nor a rating of tests against their intended purpose, which directly impacts international notification and self-declarations of disease-free countries, zones, and compartments.

Supporting evidence, if relevant: N/A</td>
<td>February 2026 report (see item 9.2.1. and Annex 25).

Note that the levels of priority are as follows:
<table><tr><td>1</td><td>- active work for the AAHSC
- to be put forward for next meeting agenda</td></tr><tr><td>2</td><td>- active work for the AAHSC
- to be included in next meeting agenda if time allows, depending on other progress</td></tr><tr><td>3</td><td>- not immediate work for the AAHSC
- needs to progress before consideration for next meeting agenda</td></tr><tr><td>4</td><td>- not active
- not to be immediately started</td></tr></table>
Items may be placed at a lower priority level due to the need for expert engagement and the time required to have appropriate engagement prior to revisions of standards.</td>
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<td>Work_2_Canada</td>
<td>Title of the item in the report or item in the work programme annex: Work program

Proposal/Comment:
Canada requests review of recent scientific evidence to evaluate Cyprinus rubrofuscus against the criteria for listing species for susceptibility to infection with KHV and SVC. Although Cyprinus carpio and C. rubrofuscus are recognised as separate species, this distinction may not have been established or used in the specific literature used to evaluate susceptibility in the 2017 ad hoc Group report. As C. carpio is currently listed as</td>
<td>The Commission noted the importance of keeping the susceptible species lists for the WOAH listed diseases up to date and thanks Members for providing recent evidence.

The Commission discussed the possibility of addressing this request with the creation of</td>
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	a susceptible species for both infection with KHV and infection with SVC, we request that *C.rubrofuscus* be evaluated as a susceptible species for both pathogens. **Rationale:** Recent morphological, genetic, and phylogeographic studies have demonstrated that the East Asian lineage was sufficiently distinct to be recognized as a separate species. Currently most taxonomic authorities, including www.fishbase.se, consider *C. rubrofuscus* a valid species in its own right, rather than a subspecies of *C. carpio*. Recent scientific evidence on detections of both KHV and SVC in koi carp reference 'Koi carp' as the common name for *C. rubrofuscus* (and 'common carp' for *C. carpio)*. However, 'Koi carp' was assessed by the *ad hoc* Group for susceptibility to KHV and SVC in 2017 as *C. carpio koi* and *C. rubrofuscus* was not evaluated. Historical challenge studies and detections of SCV and KHV in "Koi carp" are now being described as detections in *C. rubrofuscus.* This has resulted in some confusion regarding susceptibility of *C.rubrofuscus,* to infection with KHV and SVCV. Recently published articles also provide new evidence, from what was reviewed by the *ad hoc* Group in 2017, for susceptibility of *C. rubrofuscus* to infection with KHV and SVCV. Therefore, a review of the susceptibility of *C. rubrofuscus* is warranted to ensure susceptible species lists in Articles 10.7.2. and 10.9.2. are in line with current scientific evidence, support appropriate declaration of species being traded and prevent transboundry spread of disease with traded *C. rubrofuscus*. **Supporting evidence:** Adamek, M., Matras, M., Dawson, A., Piackova, V., Gela, D., Kocuour, M., Adamek, J., Kaminski, R., Rakus, K., Bergmann, S.M., Stachnik, M., Reichert, M. & Steinhagen, D. (2019). Type I interferon responses of common carp strains with different levels of resistance to koi herpesvirus disease during infection with CyHV-3 or SVCV. Fish and Shellfish Immunology, **87**, 809-819. Jeong, Y.J., Jeon, Y.G. Choi, H.J., Baek, E.J., Kim, G.H., Yang, Y.J., Kim, M.J., Min, J.G. & Kim, K.I. (2023). Genetic characterisation of alloherpesvirus (cyprinid herpesvirus-2 and koi herpesvirus) and poxvirus (carp edema virus) identified from domestic and imported cyprinids in Korea. Fish Aquatic Sci, **26(7),** 437-446.	an *ad hoc* Group to review recent evidence and allow updates of the list (see item 7.5.). Upon creation of this *ad hoc* Group submitted evidence will be provided for review.

	Kim, G.H., Jeong, Y.J., Jeon, Y.G., Yang, Y.J., Min, J.G., Kim, D-H. & Kim, K.I. (2024) Diagnostic performance of cross-priming amplification-based lateral flow assay (CPA-LFA) and real time PCR for koi herpesvirus (KHV) detection. Journal of Virological Methods, **325**, 114890.	
Work_3_Canada	**Title of the item in the report or item in the work programme annex: Work program/Code chapter 10.8. and Infection with Infection with *Megalocytivirus pagrus1*** **Proposal/Comment:** Canada wishes to request a review of the susceptibility of *Micropterus salmoides* for *Megalocytivirus pagrus1*. **Rationale:** Canada has found additional evidence in support of listing *Micropterus salmoides* for *Megalocytivirus pagrus1*. **Supporting evidence, if relevant:** Xu, Z., Liao, J., Zhang, D., Liu, S., Zhang, L., Kang, S., Xu, L., Chen, H., Peng, W., Zhou, S., Qin, Q., & Wei, J. (2023). Isolation, Characterization, and Transcriptome Analysis of an ISKNV-Like Virus from Largemouth Bass. *Viruses*, *15*(2), 398. https://doi.org/10.3390/v15020398	See response to comment Work_2.
Work_4_Canada	**Title of the item in the report or item in the work programme annex: Work program/Code chapter 10.8. and Infection with Infection with *Megalocytivirus pagrus1*** **Proposal/Comment:** Canada wishes to request a review of the susceptibility of *Sebastes schlegeli* for *Megalocytivirus pagrus1*. **Rationale:** Canada has found additional evidence in support of listing *Sebastes schlegeli* for *Megalocytivirus pagrus1*. **Supporting evidence, if relevant:** Min, J. G., Jeong, Y. J., Jeong, M. A., Kim, J.-O., Hwang, J. Y., Kwon, M.-G., & Kim, K. I. (2021). Experimental transmission of red sea bream iridovirus (RSIV) between rock bream (Oplegnathus fasciatus) and rockfish (Sebastes schlegelii). Journal of Fish Pathology, 34(1), 1–7. https://doi.org/10.7847/JFP.2021.34.1.001	See response to comment Work_2.

Work_5_Canada	**Title of the item in the report or item in the work programme annex: Work program/Code chapter 10.8. and Infection with Infection with *Megalocytivirus pagrus1*** **Proposal/Comment:** Canada wishes to request a review of the susceptibility of *Betta splendens* for *Megalocytivirus pagrus1*. **Rationale:** Canada has come to a different outcome for *Betta splendens* for *Megalocytivirus pagrus1*. WOAH AHG report had the following foot note on *Betta splendens* "Only one study was available for assessment and only one fish within that study showed clinical signs. The ad hoc Group determined that the evidence from the single fish was not sufficient for a final assessment of a '1'. As a result, the ad hoc Group assessed this species as an overall score of a '2'." The study provided internal corroboration of susceptibility with "virus-positive *B. splendens* at 13 locations during three years". **Supporting evidence, if relevant:** BAOPRASERTKUL, P. & KAENCHAN, N. (2019). Distribution and detection of Megalocytivirus in ornamental fish in Thailand. Journal of Fisheries and Environment, 43(1), 11-24.	See response to comment Work_2.
Work_6_Canada	**Title of the item in the report or item in the work programme annex: Work program Manual Chapter 2.3.10. Infection with viral haemorrhagic septicaemia virus** **Proposal/Comment:** Canada wishes to request a review of the susceptibility of *Sparus aurata* for *viral haemorrhagic septicaemia virus* for possible inclusion in section 2.2.2 of the Aquatic manual chapter 2.3.10. **Rationale:** Canada has found evidence and is requesting the review of this evidence by disease experts for *Sparus aurata* for *viral haemorrhagic septicaemia virus.* **Supporting evidence, if relevant:** Ogut H & Altuntas C, 2014. A survey of viral haemorrhagic septicaemia virus in cultured sea bass and its virulence on juveniles of sea bass, Dicentrarchus labrax (Actinopterygii:	See response to comment Work_2.

	Perciformes: Moronidae) and gilthead sea bream, Sparus aurata (Sparidae). *Acta Ichthyologica et Piscatoria* 44: 9-14. Işidan H & Kutlu İ, 2014. Viral Hemorajik Septisemi Virüs Genotip 1e Suşlarının Çipura (*Sparus aurata*) ve Levrek (*Dicentrarchus labrax*) Balıkları Üzerinde Patojenitelerinin Belirlenmesi. *Aquaculture Studies* 14: 49 - 53.	
Work_7_Canada	**Title of the item in the report or item in the work programme annex: Work program/Code Chapter 10.9. and Manual Chapter 2.3.9. Infection with spring viraemia of carp virus** **Proposal/Comment:** Canada wishes to request a review of the susceptibility of *Esox lucius* for *Spring Viremia of Carp Virus.* **Rationale:** Canada has found additional evidence and is requesting the review of this additional paper by disease experts for Esox lucis for SVCv. **Supporting evidence, if relevant:** Vicenova M, Reschova S, Pokorova D, Hulova J, Vesely T. First detection of pike fry-like rhabdovirus in barbel and spring viraemia of carp virus in sturgeon and pike in aquaculture in the Czech Republic. Dis Aquat Organ. 2011 Jun 16;95(2):87-95. doi: 10.3354/dao02340. PMID: 21848117	See response to comment Work_2.
Work_ 8_China (People's Republic of)	**Category:** general **Proposed amended text:** N/A **Rationale**: P. R. China supported the detailed list of work program of aquatic animal health standard commission for reviewing and developing WOAH aquatic animal health standard, which will give a clear and definite direction not only for Commission members, but also for the scientific network of WOAH and the members of WOAH. It fully demonstrates the principle of transparency of WOAH and is helpful to promote the Members to be active in commenting on the standard. It also reflects the hard work of the Commission and the challenge of the tendency of global diverse aquaculture styles and scales. Some suggestions are as follows: 1) Amphibian diseases are a special part of the standard. They affect not only aquaculture, but also wild populations. Sometimes they even lead to population extinction. The global	Point 1 – WOAH is in the process of recruiting amphibian experts for work on the disease-related chapters in the *Aquatic Code* and the *Aquatic Manual*. The Commission invited applications for reference laboratory experts to support this important work. Point 2 - Chapter 2.1.2. was updated to the new template and adopted in 2021. Currently needs review related to PCR methods.

	trade of amphibians and surveillance program makes it urgent to update the susceptible species information and develop the manual chapters following the new template. It should be listed above '3' for all the related work and convene experts in relevant fields. 2) Manual Chapter 2.1.2 Infection with *Batrachochytrium salamandrivorans* to be updated following the new template should be listed. 3) Emerging disease is a routine work for the Commission. It is necessary for the Members of WOAH and the collaboration center to contribute to this work, providing the information and their comments for the Commission to evaluate. 4) Reviewing the new tests recently published should be listed as a routine work for updating the Aquatic manual, since the recommended tests of WOAH are very important for the Members to adjust their surveillance program based on a more scientific rationale. 5) Aquatic animal vaccines are a crucial measure for the prevention and control of aquatic animal diseases. They also play a significant role in addressing the global issue of AMR. While they are an important component in international standards for terrestrial animals, they are not covered in international standards for aquatic animals. It is recommended to add a new relevant chapter to provide guidance for the standardized use of aquatic animal vaccines worldwide and their appropriate application within biosecurity systems. **supporting evidence:** not relevant	Point 3 – The Commission has requested to the Reference Centre on emerging diseases to undergo analysis on current emerging threats and provide evidence to the Commission for its September 2026 meeting. The Commission also *considered* the stablished network of Collaborating Centres for aquatic animal diseases to support the work on emerging diseases. Point 4 – The Commission noted the need to routine new tests and also highlighted how to validate new methods and consider gaps in research. The Commission considered how to incorporate this into questions asked in Annual Reports of Reference Laboratories. Point 5 – The Commission agreed it is important to consider inclusion of vaccination in the standards. This topic would be placed on its September 2026 agenda for further discussion.

Work_9_New Caledonia	**Proposition ou commentaire :** La Nouvelle-Calédonie est favorable à la réorganisation des chapitres suivants de la section 5 du Code aquatique, selon le même principe que le Code terrestre : 5.6 : Mesures et procédures applicables à l'exportation de marchandises 5.7 : Mesures et procédures applicables au transit de marchandises 5.8 : Mesures et procédures applicables à l'importation de marchandises 5.9 : Postes d'inspection frontaliers et centres de quarantaine **Exposé des motifs :** Cohérence entre le Code aquatique et le Code terrestre **Justification, s'il y a lieu : /**	
Work_10_New Caledonia	**Proposition ou commentaire :** La Nouvelle-Calédonie souligne l'intérêt de travailler sur les principes d'usage prudent et responsable des agents antimicrobiens chez les animaux aquatiques. **Exposé des motifs :** En effet, notamment suite à l'évolution de la réglementation européenne à ce sujet, il est important de définir s'il est possible, dans des conditions de production, de se passer totalement de certains antibiotiques, en particulier pour les stades larvaires, et quelles sont les bonnes pratiques à faire appliquer aux aquaculteurs. **Justification, s'il y a lieu : /**	

Chapter	Subject	Summary of the work	Status – September 2025		
			Stage of consideration	Remarks (Month when draft text first circulated for comment /# of rounds for comment)	Priority order *
Aquatic Code					
Ch. 1.1.	Notification of diseases, and provision of epidemiological information	Discussed with Code Commission to ensure consistency and clarity on revisions.	Revisions to be further considered at next meeting	Refer to Sept 2025 AAHSC report	**1**
Ch. 1.4.	Aquatic animal disease surveillance	Review Ch. 1.4. to consider if revision required following the update of Ch. 4.3.	Preparatory work	Refer to Sept 2025 AAHSC report	**2**
Ch. 4.2.	Application of zoning	Revision of the chapter following the update to Ch. 4.3. to focus on the application of zoning.	Preparatory work	Refer to Sept 2025 AAHSC report	**2**
Ch. 4.3.	Application of compartmentalisation	Revision of the chapter to focus on compartmentalisation. Members engaged through a questionnaire and discussion paper.	Circulated for comments	Refer to Sept 2025 AAHSC report (Feb 2025/2)	**1**
Ch. 4.7.	Fallowing in aquaculture	Review of Ch. 4.7. following the drafting of Ch. 4.X. 'Emergency disease preparedness' and Ch. 4.Y. 'Disease outbreak management'.	Circulated for comments	Refer to Sept 2025 AAHSC report (Feb 2025/2)	**1**
Section 5	Trade measures, importation/exportation procedures and health certification	Revision of chapters in Section 5 to ensure useability for trade purposes	Proposed plan for revision circulated for comments	Refer to Sept 2025 AAHSC report (Sept 2025/1)	**1**

Chapter	Subject	Summary of the work	Status – September 2025		
			Stage of consideration	Remarks (Month when draft text first circulated for comment /# of rounds for comment)	Priority order *
Ch. 5.Y.	New introductory chapter to Section 5	Taskforce between AAC and TCC convened and drafting chapter.	Preparatory work.	Refer to Sept 2025 AAHSC report	**1**
Ch. 5.1.	General obligations related to certification	Update certification procedures to align with Codex (e-certification).	Preparatory work	Refer to Feb 2025 AAHSC report	**3**
Ch. 5.2.	Certification procedures	Update certification procedures to align with Codex (e-certification).	Preparatory work	Refer to Feb 2025 AAHSC report	**3**
Ch. 5.11.	Model health certificates for international trade in live aquatic animals and aquatic animal products	Update certification procedures to align with Codex (e-certification).	Preparatory work	Refer to Feb 2025 AAHSC report	**3**
Ch. 6.2.	Principles for responsible and prudent use of antimicrobial agents in aquatic animals	*Ad hoc* Group provided Commission with recommendations and will review Commission's responses to provide a draft chapter.	Preparatory work	Refer to Sept 2025 AAHSC report	**1**
Section 7	Welfare of farmed fish	Commission review report on current science for welfare of farmed fish and proposed a consultant produce revisions of all 4 chapters.	Preparatory work	Refer to Sept 2025 AAHSC report	**2**
Ch. 8.1.	Infection with *Batrachochytrium dendrobatidis*	Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Not started	Refer to Sept 2025 AAHSC report	**3**

Chapter	Subject	Summary of the work	Status – September 2025		
			Stage of consideration	Remarks (Month when draft text first circulated for comment /# of rounds for comment)	Priority order *
Ch. 8.2.	Infection with *Batrachochytrium salamandrivorans*	Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Not started	Refer to Sept 2025 AAHSC report	**4**
Ch. 8.3.	Infection with *Ranavirus* species	Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Not started	Refer to Sept 2025 AAHSC report	**4**
Ch. 9.2.	Infection with *Aphanomyces astaci* (Crayfish plague)	Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Circulated for comment	Refer to Sept 2025 AAHSC report (Sept 2025/1)	**1**
Ch. 10.3.	Infection with *Gyrodactylus salaris*	Revise the usage of pathway 2 to declare freedom from disease, following discussion at 2025 General Session	Circulated for comments	Refer to Sept 2025 AAHSC report (Sep 2025/1)	**1**
Ch. 10.4.	Infection with salmon anaemia virus	Revise the targeted surveillance required for HPR0 for declaring a free compartment.	Circulated for comments	Refer to Sept 2025 AAHSC report (Sep 2025/1)	**1**

Chapter	Subject	Summary of the work	Status – September 2025		
			Stage of consideration	Remarks (Month when draft text first circulated for comment /# of rounds for comment)	Priority order *
Disease-specific chapters	Update viral names in X.X.1.	Update the viral names of relevant pathogenic agents based on current information in ICTV	Circulated for comments	Refer to Sept 2025 AAHSC report (Sep 2025/1)	**1**
N/A	Emerging diseases	Review emerging diseases	Standing agenda item	Refer to Sep 2025 AAHSC report	**1**
Aquatic Manual					
Ch. 1.1.2.	Validation of diagnostic assays for infectious diseases of aquatic animals	New chapter for validation of diagnostic assays for infectious diseases of aquatic animals.	Preparatory work – revision of chapter ongoing.	Refer to Sept 2025 AAHSC report	**1**
Ch. 2.1.0.	General Information	To create a general information chapter.	Not started	Refer to Sept 2025 AAHSC report	3
Ch. 2.1.1.	Infection with *Batrachochytrium dendrobatidis*	Update the chapter to the new template for disease-specific chapter.	Preparatory work	Refer to Sept 2025 AAHSC report	2
Ch. 2.1.1.	Infection with *Batrachochytrium dendrobatidis*	Sections 2.2.1. and 2.2.2. Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Not started	Refer to Sept 2025 AAHSC report	**3**

Chapter	Subject	Summary of the work	Status – September 2025		
			Stage of consideration	Remarks (Month when draft text first circulated for comment /# of rounds for comment)	Priority order *
Ch. 2.1.2.	Infection with *Batrachochytrium salamandrivorans*	Sections 2.2.1. and 2.2.2. Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Not started	Refer to Sept 2025 AAHSC report	**4**
Ch. 2.1.3.	Infection with *Ranavirus* species	Update the chapter to the new template for disease-specific chapter.	Not started	Refer to Sept 2025 AAHSC report	**4**
Ch. 2.1.3.	Infection with *Ranavirus* species	Sections 2.2.1. and 2.2.2. Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Not started	Refer to Sept 2025 AAHSC report	**4**
Ch. 2.2.2.	Infection with *Aphanomyces astaci* (Crayfish plague)	Sections 2.2.1. and 2.2.2. Update the susceptible species list in each chapter following an assessment against the criteria outlined in Ch. 1.5. 'Criteria for listing species as susceptible to infection with a specific pathogen'.	Circulated for comment	Refer to Sept 2025 AAHSC report (Sept 2025/1)	**1**
Ch. 2.2.5.	Infection with infectious hypodermal and haematopoietic necrosis virus	Expert consultation to update EVE in chapter.	Expert consultation	Refer to Sept 2025 AAHSC report	**1**

Chapter	Subject	Summary of the work	Status – September 2025		
			Stage of consideration	Remarks (Month when draft text first circulated for comment /# of rounds for comment)	Priority order *
Ch. 2.3.5.	Infection with infectious haematopoietic necrosis virus	Update the real-time PCR assays following publication of new peer-reviewed validated methods.	Preparatory work	Refer to Sept 2025 AAHSC	**1**
Ch. 2.3.7.	Infection with *Megalocytivirus pagrus1*	New chapter for infection with *Megalocytivirus pagrus1,* which was listed in the *Aquatic Code* in May 2024	Preparatory work – interlaboratory comparison	Refer to Sept 2025 AAHSC report	**2**
Ch. 2.3.9.	Infection with spring viraemia of carp virus	Update the real-time PCR assays following publication of new peer-reviewed validated methods	Preparatory work	Refer to Feb Sept AAHSC report	**1**
Ch. 2.3.X.	Infection with tilapia lake virus	New chapter for infection with tilapia lake virus, which was listed in the *Aquatic Code* in May 2022	Preparatory work	Refer to Sept 2025 AAHSC report	**3**
Ch. 2.4.2.	Infection with *Bonamia exitiosa*	Update the case definition	Circulated for comments	Refer to Sept 2025 AAHSC report (Sep 2025/1)	**1**
Ch. 2.4.3.	Infection with *Bonamia ostreae*	Update the case definition	Circulated for comments	Refer to Sept 2025 AAHSC report (Sep 2025/1)	**1**
Ch. 2.4.5.	Infection with *Perkinsus marinus*	Update the chapter to the new template for disease-specific chapter.	Circulated for comments	Refer to Sept 2025 AAHSC report (Sep 2025/1)	**1**
Ch. 2.4.6.	Infection with *Perkinsus olseni*	Update the chapter to the new template for disease-specific chapter.	Circulated for comments	Refer to Sep 2025 AAHSC report (Feb 2025/2)	**1**
Ch. 2.4.7.	Infection with *Xenohaliotis californiensis*	Update the chapter to the new template for disease-specific chapter.	Circulated for comments	Refer to Sept 2025 AAHSC report (Feb 2025/2)	**1**

* Description of priority order	
1	- active work for the AAHSC - to be put forward for next meeting agenda
2	- active work for the AAHSC - to be included in next meeting agenda if time allows, depending on other progress
3	- not immediate work for the AAHSC - needs to progress before consideration for next meeting agenda
4	- not active - not to be immediately started

List of abbreviations	
AHG	*Ad hoc* Group
Ch	Chapter
HQ	WOAH Headquarters
AAHSC	Aquatic Animal Health Standard Commission

GLOSSARY

[...]

Reference	Comment	Aquatic Animals Commission Response
Glossary-ornamental aquatic animal_1_United States	**Category:** general We thank the Commission for revisiting this definition following the discussions during the 92nd WOAH General Session. The trade of ornamental aquatic animals is of great importance to Members, and we support the proposed change which we believe more accurately reflects the intended uses.	Noted.
Glossary-ornamental aquatic animal_2_New Caledonia	**Category:** Change ANIMAL AQUATIQUE ORNEMENTAL désigne un animal aquatique destiné à faire l'objet d'une présentation au public, d'une exposition, ou d'une compétition, ou à être cédé à titre gratuit ou onéreux, ou un animal aquatique détenu ~~proposé à la vente comme vendu et détenu~~ en tant qu'animal de compagnie. Les animaux aquatiques détenus en tant qu'animal de compagnie peuvent présenter un risque de transmission de maladie lorsque les conditions de détention ne sont pas sécurisées (au-delà de la notion de transport). Par exemple, en Nouvelle-Calédonie, certains poissons détenus comme animaux de compagnie, sont maintenus dans des bassins extérieurs proches d'une rivière et insuffisamment protégés, avec débordements réguliers et possibles fuites des poissons dans le cours d'eau. Au-delà des conditions de détention, l'élimination des animaux aquatiques détenus en tant qu'animaux de compagnie après leur mort, est généralement non conforme (par exemple dans les toilettes) et présente des risques de diffusion de maladies. De même certains détenteurs relâchent leurs poissons dans la nature quand ils ne souhaitent plus s'en occuper. Ces cas de figure représentent un risque sanitaire et environnemental important pour les espèces sauvages, notamment. Il est donc nécessaire que la notion d'animal aquatique détenu en tant qu'animal de compagnie ressorte bien dans cette définition, par rapport au risque que cela représente au moment de la détention. D'autre part, la notion de vente est limitative, il est proposé de prendre en compte toute cession, qu'elle soit à titre gratuit ou onéreux.	Did not agree to expand the definition to include animals kept as pets. The purpose of this term in Chapter 5.12. 'Movement of ornamental aquatic animals' the purpose is to address the risk of pathogenic agent transmission by movements and trade. Animals kept in households as pets are not intended to be in scope as measures are intended to apply to transboundary movements. Competent authorities may wish to apply internal controls to manage the disease risks associated with end uses of ornamental aquatic animals (e.g. deliberate release or escape of pets into waterways), however this is not within the scope of the chapter.

Glossary-ornamental aquatic animal_3_Peru	**Categoría:** Adición **Texto modificado propuesto:** Animal Acuático Terapéutico Designa un animal acuático destinado a ser utilizado en fines terapéuticos, como parte del comportamiento natural del mismo. **Justificación:** Se reporta el uso de especies de peces como *Garra rufa*, para tratamiento de psoriasis en personas, dicho uso es distinto a la definición contenida como animal acuático ornamental. En ese sentido, se propone adicionar la definición. **Evidencia documentada:** Shimada, Y., Aydın, B., Kon-Nanjo, K., Handayani, K. S., Gultom, V. D. N., Simakov, O., ... & Kon, T. (2025). Potential of Garra rufa as a novel high-temperature resistant model fish: a review on current and future approaches. *Zoological Letters*, *11*(1), 3. https://doi.org/10.1186/s40851-025-00249-0	Did not agree to include the concept of a therapeutic animal. The use of the term 'ornamental aquatic animal' is applicable in Chapter 5.12. 'Movement of ornamental aquatic animals' and does not include this usage for aquatic animals in its scope.
Glossary-ornamental aquatic animal_4_EU	**Category:** General The EU supports this revised definition of the glossary.	Noted.

ORNAMENTAL AQUATIC ANIMAL

means an *aquatic animal* that is intended for display, exhibition, competition, or to be supplied for sale ~~and use~~ as a pet.

Glossary-ornamental aquatic animal_5_China (People's Republic of)	**Category:** change **Proposed amended text:** *ORNAMENTAL AQUATIC ANIMAL* means an *aquatic animal* that is intended, either directly or through its progeny after breeding and rearing, for display, exhibition, landscaping purposes, competition, or to be supplied for sale trade ~~and use~~ as a pet~~.~~, excluding those kept in households without involvement in trade circulation. **Rationale :** 1）The current definition does not fully reflect the complete industry chain and trade purposes of ornamental aquatic animals. The proposed revision by China aims to address two key deficiencies: Incomplete Purposes: The original definition omits the	Did not agree. For 'breeding/rearing' - The usage of aquatic animals for broodstock occurs in all aquatic animals and regarding ornamental species this purpose is not commonly moved or traded. For 'landscaping purposes' – This usage would be considered an aquatic animal for display, as already included in the definition.

	significant purpose of "landscaping" (e.g., for use in aquascapes, living communities, or artificial ecological parks). The addition of "landscaping purposes" provides broader coverage. Ambiguous Pathways: The original definition fails to distinguish between the direct use of an animal and its indirect use as broodstock. A substantial portion of international trade involves parent animals for breeding, whose progeny are then used for display or sale. The new phrase, "either directly or through its progeny after breeding and rearing," clearly defines both scenarios. This explicitly includes broodstock within the definition of "ornamental aquatic animal," closing a potential regulatory loophole, preventing trade disputes, and allowing for more targeted risk management measures. 2）Exclusion of Non-Circulatory Household Keeping:Household-kept aquatic animals not involved in trade circulation pose negligible disease transmission risks and should not fall under trade-related regulatory scope. The current definition does not clearly exclude this scenario, which may lead to misinterpretation and impose unnecessary regulatory burdens on low-risk situations. **supporting evidence:** not relevant	'excluding those kept in households without involvement in trade circulation' – this text is unnecessary as the current text already excludes those animals as it is only those being supplied as pets but not kept as pets.
Glossary-ornamental aquatic animal_6_Thailand	**Category:** Change **Proposed amended text:** "Means an aquatic animal that is intended for display, exhibition, competition, or <u>to be supplied</u> ~~for sale~~ ~~and used~~ as a pet" **Rationale:** To ensure rigorous and effective risk management regarding the international movement of ornamental aquatic animals. Even though these animals are imported for personal use (as pets), they could be developed into broodstock for commercial production, or may act as a source of disease spreading through their movement, especially if the sanitary measures and risk analysis outlined in Chapter 5.12 (Movement of ornamental aquatic animals) are not properly implemented. Therefore, disease risks associated with the movement of aquatic animals for "personal use" should be considered when defining the term "ornamental aquatic animals". It is proposed that the scope covers both aquatic animals supplied for sale as pets and those imported for use as a pet via personal trade. **Supporting evidence:** -	Did not agree. Trade of aquatic animals for use as pets is predominantly commercial. Non-commercial movement of aquatic animals is covered by other elements of the definition. See response to comment Glossary-ornamental aquatic animal_2.

[...]

SECTION 4

DISEASE PREVENTION AND CONTROL

CHAPTER 4.7.

FALLOWING IN AQUACULTURE

Reference	Comment	Aquatic Animals Commission Response
4.7._1_Australia	**Category:** General comment **Proposed amended text:** n/a **Rationale**: The proposed amendments to Chapter 4.7 Fallowing in Aquaculture has improved its readability, clarity and focussed content to the outcomes that fallowing aims to achieve as a disease management measure. **Supporting evidence:** n/a	Noted.
4.7._2_Norway	**Category:** General **Proposed amended text:** not relevant **Rationale:** Norway in general supports the proposed changes to this Chapter. Nevertheless, some specific comments are provided below. **Supporting evidence:** not relevant	Noted.
4.7._3_EU	**Category:** General **Proposed amended text:** not relevant **Rationale:** The EU supports this proposed chapter and thanks the Aquatic Animals Commission for addressing the previous EU comments. **Supporting evidence:** not relevant	Noted.

Article 4.7.1.

Introduction

~~Gaps in *aquaculture* production at the same location are commonly recognised to be of value in resting or restoring the local environment. As part of this strategy, *fallowing* can break re-*infection* cycles by removing loci of a *disease* from a farm. Consequently, *fallowing*~~

Fallowing is a ~~routine carried out as a regular~~ *disease* management measure in *aquaculture*, which is employed either: ~~as a best practice especially prior to the introduction of new populations of *aquatic animals* into a previously stocked used site,~~

1) voluntarily, prior to the introduction of certain new populations of *aquatic animals* into a previously stocked *aquaculture establishment* as part of a *biosecurity plan* constructed in accordance with Chapter 4.1., or

4.7.1._1_Norway	**Category**: Addition **Proposed amended text** (or precise suggested deletion): 1. voluntarily or compulsory, prior to the introduction of certain new populations of *aquatic animals* into a previously stocked *aquaculture establishment* as part of a *biosecurity plan* constructed in accordance with Chapter 4.1., or **Rationale:** While some countries may not require compulsory fallowing to ensure adequate biosecurity within the aquaculture industry, many countries need compulsory fallowing as a routine measure to reduce infection pressure. Given the importance of fallowing as a biosecurity measure, it is important that this article doesn't inadvertently restrict competent authorities' ability to impose compulsory fallowing as a legal requirement when needed, in addition to its use as an emergency measure as mentioned in point 2). We therefore suggest the addition of "or compulsory". **Supporting evidence, if relevant:** not relevant	Agreed, added to point 1 that fallowing can be voluntary or compulsory.

2) compulsorily, on the instructions of the *Competent Authority*, following an outbreak of a *disease* which is subject to emergency management measures as described in Chapter 4.11~~Y~~.

In the case of the voluntary *fallowing*, the objectives are to prevent transmission of *pathogenic agents* between successive production cycles and suppress *pathogenic agent infection* pressure.

4.7.1._2_New Caledonia	**Catégorie**: éditorial **Proposition de texte modifié :** Reformulation pour clarification et uniformisation Le *vide sanitaire* est une mesure de gestion des *maladies en aquaculture*, qui est employée soit : 1) de manière volontaire, avant l'introduction de ~~certaines~~ nouvelles populations d'*animaux aquatiques* dans un *établissement d'aquaculture* peuplé auparavant, dans le cadre d'un *plan de sécurité biologique* élaboré en conformité avec le chapitre 4.1., soit ~~ou~~	Agreed to delete certain from point 1 as fallowing where relevant should occur prior to introduction of any new population. Agreed to editorial change in French version only. Did not agree to change 'prevent' to 'limit' in first paragraph. The objective of

	2) de manière obligatoire, sur instruction de l'*Autorité compétente*, à la suite d'un foyer d'une *maladie* faisant l'objet de mesures de gestion d'urgence, comme décrit dans le chapitre 4.11. Dans le cas d'un vide sanitaire volontaire, les objectifs consistent à limiter ~~empêcher~~ la transmission d'agents pathogènes entre des cycles successifs de production et à réduire ~~supprimer~~ la pression d'infection par les agents pathogènes. **Exposé des motifs :** 1) la notion de « certaines » pour parler des nouvelles populations sous-entend que cela serait différent pour d'autres, alors que le vide sanitaire s'applique pour toute réintroduction, même s'il s'agit d'une action volontaire. Par ailleurs, si on utilise « soit » avant le 1) il convient de mettre « soit » pour introduire le 2). Enfin, il semble illusoire d'indiquer que le vide sanitaire va « empêcher » la transmission et « supprimer » la pression d'infection, il est proposé de remplacer par « limiter » et « réduire ». **Justification, s'il y a lieu :/**	fallowing is to prevent disease transmission rather than limit it, and the text is correct as written. Did not agree, to change 'suppress' to 'reduce' as there would be no material benefit to the text.
4.7.1._3_UK	**Category:** Deletion **Proposed amended text:** In the case of ~~the~~ voluntary *fallowing*, the objectives are to prevent transmission of *pathogenic agents* between successive production cycles and suppress *pathogenic agent infection* pressure. **Rationale:** Editorial	Agreed to editorial change.

In the case of compulsory *fallowing*, the objective is to eradicate a *pathogenic agent* from an *aquaculture establishment* or in the case of synchronous *fallowing*, from a group of epidemiologically connected *aquaculture establishments*.

4.7.1._4_Canada	**Comment Category:** Addition **Proposed amended text:** In the case of compulsory *fallowing*, the objective is to eradicate a *pathogenic agent* from an *aquaculture establishment* ~~or in the case of synchronous *fallowing*, from a group of epidemiologically connected *aquaculture establishments*.~~ Synchronous fallowing, when a group of epidemiologically connected aquaculture establishments are followed at the same time for the same duration, can be used as voluntary fallowing or as compulsory fallowing. **Rationale:** 'Synchronous fallowing' is used throughout the chapter without describing what it is. In the proposed sentence above, it seems that it can only be used as part of compulsory fallowing however many countries use synchronous fallowing voluntarily through bay	Agreed that the text should describe what synchronous fallowing is and clarify its use. The text was amended to take this into account.

	management plans to try to prevent transmission of pathogenic agents between successive production cycles and suppress the infection pressure in open aquaculture establishments.	
4.7.1._5_China (People's Republic of)	**Category:** change **Proposed amended text :** Article 4.7.1. ~~In the case of the~~ Voluntary *fallowing*, ~~the objectives are~~ aims to prevent transmission of *pathogenic agents* between successive production cycles and suppress *pathogenic agent infection* pressure. ~~In the case of~~ Compulsory *fallowing*, ~~he objectives are~~ aims to eradicate a *pathogenic agent* from an *aquaculture establishment* or in the case of synchronous *fallowing*, from a group of epidemiologically connected *aquaculture establishments*. **Rationale:** Syntax and logic adjustments. **supporting evidence:** not relevant	Agreed to editorial changes.

Article 4.7.2.

Considerations for fallowing

Fallowing is used to provide a temporal break in the transmission of a *pathogenic agent* ~~transmission cycles~~ between successive cohorts of *susceptible species* or where relevant vector species ~~populations~~ of *aquatic animals*. It should be implemented taking the following factors into account ~~with consideration given to~~:

4.7.2._1_Australia	**Category:** editorial **Proposed amended text:** "Fallowing is used to provide a temporal break in the transmission of a pathogenic agent between successive ~~cohorts~~ production cycles of susceptible species or …" n/a **Rationale**: The terminology for the description of fallowing should align to international aquaculture definitions. For example, the Aquaculture Stewardship Council defines fallowing by 'production cycles' (ASC Farm Standard version 1.0.1, 1 August 2025). Also, fallowing in the literature is defined by 'production or farming cycles' (Zhulay et al 2015). Production cycles may include for example successive cohorts, multiple cohorts farmed on a site over a period of time (the production cycle), or the same cohorts (production fish separated since the hatchery through different growth conditions or different grow out sites and subsequently moved to a fallowed site previously occupied by fish of the same cohort). Furthermore, the suggested edit aligns with the term 'successive production cycles' that is already used in Article 4.7.1. **Supporting evidence:** • ASC farm standard Aug 2025 • Zhulay et al. Marine Pollution Bulletin 97 (2015) 381-390	Agreed to change 'cohorts' to 'production cycles' because the term is more appropriate.
4.7.2._2_Norway	**Category:** deletion **Proposed amended text:** *Fallowing* is used to provide a ~~temporal~~ break in the transmission of a *pathogenic agent* ~~transmission cycles~~ between successive cohorts of *susceptible species* or where relevant vector species ~~populations~~ of *aquatic animals*. It should be implemented taking the following factors into account ~~with consideration given to~~: **Rationale:** Suggest deleting the word "temporal" as the word break is sufficient. **Supporting evidence:** not relevant	Did not agree to delete 'temporal' break, as the use of 'temporal' details the type of break (in time) provided by fallowing, and provides clarity.

4.7.2._3_New Caledonia	**Catégorie:** éditorial **Proposition de texte modifié :** Remplacer « effectuer une interruption dans le temps de la transmission » par « interrompre temporairement la transmission », pour clarifier le sens de la phrase « Le *vide sanitaire* est utilisé pour ~~effectuer une interruption dans le temps~~ interrompre temporairement la transmission d'*agents pathogènes* entre des cohortes successives d'*espèces sensibles* ou, le cas échéant, d'espèces de *vecteurs* pour des *animaux aquatiques*. » ***Exposé des motifs :*** *l'expression « effectuer une interruption dans le temps de la transmission » n'est pas explicite en français.* **Justification, s'il y a lieu : /**	Agreed, translation change in French version only.

~~1) the objective of *fallowing* such as preventing transmission between sequential production cycles, suppression of *pathogenic agent infection* pressure, or to eradicate a *pathogenic agent* from an *aquaculture establishment*;~~

12) the possible sources of *infection* at the *aquaculture establishment* ~~production site~~ such as farmed or wild populations of *susceptible species* ~~*aquatic animals*~~, *vectors*, fomites or *pathogenic agents* in the environment (e.g. water or sediment);

23) characteristics of the relevant *pathogenic agent*, including its survival and stability outside the host and its infective period ~~whether the *pathogenic agent* is obligate or facultative~~;

4.7.2._4_Peru	**Categoría:** Adición **Texto modificado propuesto:** *"Deberá implementarse teniendo en cuenta los siguientes factores:* *2) Las características del agente patógeno relevante, incluida su supervivencia y estabilidad fuera del hospedador y de su* ***~~periodo infeccioso~~*** *infecciosidad;".* **Justificación:** Respecto al término "periodo infeccioso" o algunas veces llamado "periodo de trasmisibilidad" se puede interpretar como el periodo de tiempo durante el cual el agente infeccioso está presente y activo en el organismo del huésped. No obstante, el término correcto sería "Infecciosidad", toda vez que el texto haciendo referencia a la capacidad de infecciosidad del agente patógeno fuera del hospedador en el medio ambiente (agua o sedimento). Por lo tanto, se sugiere eliminar el término "periodo infeccioso" y cambiarlo por "infecciosidad". **Evidencia documentada:** https://www.sciencedirect.com/topics/mathematics/infectious-period	Did not agree to change 'infective period' to 'infectiousness'. The point mentions the survival, stability, and infective period of the pathogenic agent which taken together indicates the infectiousness of the pathogenic agent.

~~4) for obligate *pathogenic agents*, the period that they may remain viable in the environment;~~

35) the need for spatial coordination to synchronously fallow epidemiologically connected *aquaculture establishments*;

4) the type of *aquaculture* production system taking into account its design, extent and application of *biosecurity* measures;

56) *aquaculture establishment(s)* should when the infective period is not known, the farm may be fallowed for a period of time, the length of which should be based on a *risk assessment*.

4.7.2._5_China (People's Republic of)	**Category:** change **Proposed amended text :** 56) *aquaculture establishment(s)* should when the infective period is not known, the farm may be fallowed for a period of time, the length of **fallowing** should be based on a *risk assessment*. **Rationale:** Context connection. **supporting evidence:** not relevant	Agreed, text revised to reflect comment.
4.7.2._6_New Zealand	**Category:** editorial **Proposed amended text:** 4) the type of *aquaculture* production system taking into account its including the system's design, extent and application of *biosecurity* measures; 56) the length of time the *aquaculture establishment(s)* should when the infective period is not known, the farm may be fallowed for a period of time, the length of which should be based on a *risk assessment*. **Rationale:** Suggested re-wording to make the sentences read better as a list, flowing from "It should be implemented taking the following factors into account:" **Supporting evidence** n/a	Agreed, editorial change.
4.7.2._7_United States	**Category:** editorial **Proposed amended texts (or precise suggested deletion):** "5) a *risk assessment* should be used to determine the fallow time period at the *aquaculture establishment*." **Rationale:** We recommend the above editorial change for clarity, enhance the flow of the sentence, and because the risk assessment is the important factor we are highlighting that should be used to determine this establishment-specific timeframe. **Supporting evidence: -**	Editorial changes made from previous comments to address the flow and clarity.

Article 4.7.3.

Voluntary fallowing

When assessing the potential benefits of recommending voluntary *fallowing*, the *Aquatic Animal Health Services* in a country should in addition to the considerations outlined in Article 4.7.2. take the following factors into account:

1) the level of *risk* a particular *pathogenic agent* poses to local *aquaculture* operations, and to other aquatic resources in the area;

2) the relevant socioeconomic conditions and ~~In order to promote improved health in *aquaculture*, the *Aquatic Animal Health Service* in a country may encourage the voluntary use of *fallowing* as a part of the *biosecurity plan* set out in Chapter 4.1. as a *biosecurity* measure for an individual *aquaculture establishment* or as a common *biosecurity* measure among all *aquaculture establishments* that are considered epidemiologically linked in a given area. a routine management strategy for many *diseases*. Account should be taken of When encouraging aquaculture operators to fallow their establishments, the *Competent Authority* should emphasise~~ the likely beneficial effects of *fallowing* in proportion to the economic costs involved.

~~The *Aquatic Animal Health Service* should also consider such factors as take into account the level of *risk* a particular *disease* poses to the local and national *aquaculture* operations, previous knowledge of the severity of a *disease(s)*, the infective period of the *disease* in question, and distribution of the *pathogenic agent(s)*, as well as the relevant socioeconomic conditions ,and when assessing the potential benefits pertaining to the general aquatic resources in the area. When the infective period is not known, the farm may be fallowed for a period, the length of which should be based on a *risk assessment*.~~

4.7.3._1_New Caledonia	**Catégorie**: éditorial ***Proposition de texte modifié*** *: Le sens de la phrase est différente en français et anglais en utilisant l'expression "d'exploitations d'aquaculture"* 1) le niveau de *risque* que représente un *agent pathogène* donné pour les activités ~~exploitations~~ d'*aquaculture* locales, et pour d'autres ressources aquatiques dans la zone ; **Exposé des motifs :** Le sens de la phrase est différent en français et anglais en utilisant l'expression « d'exploitations d'aquaculture ». La notion « d'exploitations d'aquaculture » peut être confondue avec « établissements d'aquaculture » en français alors que dans le sens de la phrase en anglais il s'agit des « activités », traduites par « opérations ». **Justification, s'il y a lieu : /**	Agreed, translation change in French version only.
4.7.3._2_New Zealand	**Category:** addition **Proposed amended text:** 2) the relevant socioeconomic conditions and ~~In order to promote improved health in *aquaculture*, the *Aquatic Animal Health Service* in a country may encourage the voluntary use of *fallowing* as a part of the *biosecurity plan* set out in Chapter 4.1. as a *biosecurity* measure for an individual *aquaculture establishment* or as a common *biosecurity* measure among all *aquaculture establishments* that are considered epidemiologically linked in a given area. a routine management strategy for many *diseases*. Account should be taken of When encouraging aquaculture operators to fallow their establishments, the *Competent Authority* should emphasise~~ the likely beneficial effects of *fallowing* in	Agreed that text is the text is describing a cost-benefit analysis, and to add text to make this explicit.

	proportion to the economic costs involved. This may be informed by a cost-benefit analysis. **Rationale:** This appears to be describing a cost-benefit analysis – if this is the case, it should be mentioned. **Supporting evidence** n/a	
4.7.3._3_UK	**Category:** Addition **Proposed amended text:** … 1) the level of *risk* a particular *pathogenic agent* poses to local *aquaculture* operations, and to other aquatic resources in the area; 2) the relevant socioeconomic conditions and the likely beneficial effects of *fallowing* in proportion to the economic costs involved. 3) any existing industry standards and codes of practice related to *fallowing* **Rationale:** CAs should be cognizant of the industry expectations within their countries and how these may impact the perceived benefits of voluntary fallowing.	Point accepted, but reference to industry standards or Codes of Practice included in Article 4.7.1. point 1, rather than in this Article.

Article 4.7.4.

Compulsory fallowing

Compulsory *fallowing* may be mandated by ~~required in accordance with the instructions of~~ the *Competent Authority* following an outbreak of an important *disease* which has been subject to the measures described in Chapters 4.10X. and 4.11Y. ~~However, where an official *stamping-out policy* is being carried out for a *disease* of concern, the *Aquatic Animal Health Service* should~~ The *Competent Authority* may require that an infected *aquaculture establishment*, and ~~all~~ other epidemiologically linked ~~relevant~~ *aquaculture establishments* in an officially declared ~~established~~ *infected zone*, are ~~be~~ subjected to a ~~required~~ period of *fallowing, if necessary synchronised*. The duration of the ~~this~~ *fallowing* period will ~~be carried out for a period of time which is~~ prescribed by the *Competent Authority*, following *risk assessment*. *Risk assessment* will also be used to determine if a A period of synchronous *fallowing* is ~~may be~~ required in epidemiologically linked ~~relevant~~ *aquaculture establishments* in the *infected zone* as well as the duration of such *fallowing* ~~should this be indicated by the *risk assessment*.~~

4.7.4._1_China (People's Republic of)	**Category:** change **Proposed amended text :** Compulsory *fallowing* may be mandated by ~~required in accordance with the instructions of~~ the *Competent Authority* following an outbreak of an important *disease* which has been subject to the measures described in Chapters 4.10~~X~~. and 4.11~~Y~~. ~~However, where an official *stamping-out policy* is being carried out for a *disease* of concern, the *Aquatic Animal Health Service* should~~ The *Competent Authority* may require that an infected *aquaculture establishment*, and ~~all~~ other epidemiologically linked ~~relevant~~ *aquaculture establishments* in an officially declared ~~established~~ *infected zone*, are ~~be~~ subjected to a ~~required~~ period of *fallowing, if necessary*	Agreed to change in text to improve readability and clarity.

	~~synchronised~~. ~~The duration of the this *fallowing* period will be carried out for a period of time which is prescribed by the *Competent Authority*, following *risk assessment*. *Risk assessment* will also be used to determine if a A period of synchronous *fallowing* is may be required in epidemiologically linked relevant *aquaculture establishments* in the *infected zone* as well as the duration of such *fallowing* should this be indicated by the *risk assessment*.~~ The duration and the necessity for fallowing of establishment (s) and synchronous fallowing in epidemiologically linked establishments within the infected zone, will be determined by the Competent Authority based on risk assessment. **Rationale**: The concepts of duration and risk assessment are repeated in relation to prescribing and determining fallowing and duration of fallowing period. **Supporting evidence:** not relevant	
4.7.4._2_New Caledonia	**Catégorie**: éditorial. **Proposition de texte modifié :** Reformulation pour éviter les redondances ~~L'*Autorité compétente* peut exiger qu'un *établissement d'aquaculture* infecté, ainsi que les autres *établissements d'aquaculture* présentant un lien épidémiologique situés dans une *zone* faisant l'objet d'une déclaration officielle de *zone infectée*, soient soumis à une période de *vide sanitaire*. La durée de la période de *vide sanitaire* sera prescrite par l'*Autorité compétente*, à la suite d'une *appréciation du risque*. L'*appréciation du risque* sera également utilisée pour déterminer si une période de *vide sanitaire synchronisé* est exigée dans les *établissements d'aquaculture* présentant un lien épidémiologique situés dans une *zone infectée*, ainsi que la durée de ce *vide sanitaire*.~~ Selon l'appréciation du risque, l'*Autorité compétente* peut exiger qu'un *établissement d'aquaculture* infecté, ainsi que, de façon synchronisée, les autres *établissements d'aquaculture* présentant un lien épidémiologique situés dans une zone faisant l'objet d'une déclaration officielle de *zone infectée*, soient soumis à une période de *vide sanitaire* dont elle définira la durée. **Exposé des motifs :** reformuler pour éviter les redondances à partir de la seconde phrase du premier paragraphe de cet article, la formulation est très redondante, **Justification, s'il y a lieu : /**	Did not agree to delete introductory text as it provides links to other relevant chapters in the *Aquatic Code*. Amendments were however, agreed to reduce redundancies, and simplify the text.
4.7.4._3_UK	**Category:** Change **Proposed amended text:** Compulsory *fallowing* may be mandated by the *Competent Authority* following ~~an outbreak of an important *disease*~~ a *disease* outbreak which has been subject to the measures described in Chapters 4.10. and 4.11.	Agreed to change important disease to disease, given the explicit reference to Chapters 4.10. and 4.11..

	Rationale: The use of the term “important disease” implies that some diseases listed by WOAH are more or less important than others, which is not the case and not in keeping with other language used throughout the Code and Manual.	

The *Competent Authority* should ensure compulsory *fallowing* is underpinned by legal provisions that set out the following details;

4.7.4._4_Canada	**Comment Category: Change** **Proposed amended text:** The *Competent Authority* should ensure the legal authority to enforce compulsory *fallowing* is underpinned by legal provisions that require the and their requirements should include the follows previsdions; **Rationale:** Current wording is overly prescriptive. The details outlined are not always explicitly stated in legislation; however, a Competent Authority (CA) may have the authority to impose any measures necessary to control an outbreak and restore disease-free status. Broad legislative provisions granting a country the authority to require fallowing should not be excluded from this chapter. We recommend revising the wording to state that the CA should have the legal authority to mandate compulsory fallowing when required by the circumstances.	Agreed to revise text to avoid being too prescriptive.

~~Article 4.7.52.~~

~~**Legal powers**~~

~~In the cases referred to in Article 4.7.1. where *fallowing* is may be a compulsory measure, prescribed by the *Competent Authority*, for instance in the establishment or restoration of a *disease free zone*, countries should establish a legal framework must be in place to: for the implementation of *fallowing* procedures in *aquaculture establishments*. Legal provisions could include:~~

1) ~~define~~ ~~defining the~~ conditions under which ~~*disease* circumstances when~~ *fallowing* or synchronised *fallowing* is required including specific implementation steps for each;

2) specific point at which *fallowing* should commence;

3) duration of the *fallowing* period;

4) conditions under which the re-introduction of *aquaculture* species will be permitted, once the *fallowing* period has been completed.

~~define defining mechanisms based on *risk assessment* where individual disease-specific measures may be determined, including when *fallowing* should commence *disinfection* and the length of the *fallowing* period prior to the re-introduction of *susceptible species*.;~~

3) ~~following permission by the *Competent Authority* to restock with *susceptible species*, defining a period of *surveillance* and *diagnostic* to verify freedom from the specified *disease*.~~

Article 4.7.5~~6~~3.

Technical parameters for the implementation of a compulsory ~~statutory~~ fallowing plan

Taking into account the categories of *aquaculture* production systems referred to in Article 4.1.5., as well as the measures described in paragraph 5 of Article 4.10.7., *fallowing* of an a*quaculture establishmen*t ~~*Fallowing* of a farm~~ should ~~take paragraph 5 of Article 4.X.7. into account and~~ start immediately after the following actions are taken:

1) removal ~~destruction of~~ and biosecure disposal of:

4.7.5._1_New Zealand	**Category:** editorial **Proposed amended text:** **Technical parameters for the implementation of a compulsory ~~statutory~~ fallowing plan** Taking into account the categories of *aquaculture* production systems referred to in Article 4.1.5., as well as the measures described in paragraph 5 of Article 4.10.7., *fallowing* period of an a*quaculture establishmen*t ~~*Fallowing* of a farm~~ should ~~take paragraph 5 of Article 4.X.7. into account and~~ start immediately after the following actions are taken: **Rationale:** Suggesting edits to make the paragraph clearer, in particular that the period of time the site is “fallowed” starts *after* these conditions are met. **Supporting evidence** n/a	Agreed, text revised for clarity that the period of fallowing occurs after conditions are met.

a) ~~removal of~~ all *susceptible species* of *aquatic animals* for the *disease* of concern; and

b2) ~~removal of~~ all species of *aquaculture* animals which are capable of acting as *vectors* of the *disease* of concern, if indicated by *risk assessment*; and

c3) ~~if appropriate, removal of~~ other species, if indicated by *risk assessment*; ~~and~~

4.7.5._2_Norway	**Category**: general **Proposed amended text**: Clarification of text needed **Rationale:** Point 1a) in this article refers to "all species of aquatic animals" while point 1b) refers to "all species of aquaculture animals". Is the use of "aquatic animal" vs "aquaculture animal" in these two sentences intentional? If yes, why should there be aquatic animals in an aquaculture establishment other than aquaculture animals (except wild species that may enter open or semi-open systems and would be difficult to control)? Furthermore, "aquaculture animal" is not defined in the glossary, so it is at present not possible to ascertain what this really refers to. Please amend the text to enhance clarity. **Supporting evidence, if relevant**: not relevant	Agreed that there should be consistency of terms and that it is "susceptible species of aquaculture animals" for the disease of concern. Text revised for clarity.

2) removal of water in which infected stocks have been held, where feasible; and

3~~5~~) appropriate *disinfection* measures have been completed on equipment and other contaminated materials in accordance with Article 4.7.4., under the oversight of the *Competent Authority* ~~supervision of the *Aquatic Animal Health Services.* equipment and other materials contaminated or otherwise capable of harbouring *infection* have either been removed or subjected to *disinfection* to standards approved by the *Aquatic Animal Health Service*.~~

4.7.5._3_Canada	**Comment Category:** Addition **Proposed amended text:** 2) removal of water in which infected stocks have been held, where feasible; ~~and~~ 3) biosecure disposal of any solid waste such as faeces, feed or mortalities from the establishment if applicable; and 4~~3~~5) appropriate *disinfection* (including dry and wet cleaning) measures have been completed on equipment and other contaminated materials in accordance with Article 4.7.4., under the oversight of the *Competent Authority* ~~supervision of the *Aquatic Animal Health Services.* equipment and other materials contaminated or otherwise capable of harbouring *infection* have either been removed or subjected to *disinfection* to standards approved by the *Aquatic Animal Health Service*.~~	Agreed to adding point 3 to include biosecure disposal of any solid wastes and agreed to add 'including cleaning' with disinfection in new point 4.

	Rationale: This section currently omits the removal of solid waste, such as feces, feed, mortalities, or other contaminated materials. Canada recommends adding a specific point to address this requirement. Furthermore, while the WOAH definition of disinfection includes cleaning, Canada recommends that Point 43 explicitly emphasize the cleaning step of removal of solids (such as biofilm, mineral deposits and solid organic material) from equipment prior to disinfection, as this step is critical to ensuring effectiveness.	
4.7.5._4_New Caledonia	**Catégorie**: modification **Proposition de texte modifié:** Supprimer l'expression "si possible" et préciser les conditions de la vidange de l'eau lors d'un vide sanitaire obligatoire 2) ~~si possible~~ Selon les caractéristiques de l'agent pathogène et sa survie dans l'eau et les fonds de bassin, la vidange, dans des conditions de sécurité biologique adéquates, de l'eau dans laquelle des stocks d'animaux infectés ont été maintenus, et… **Exposé des motifs :** supprimer l'expression « si possible » et préciser les conditions de la vidange de l'eau lors d'un vide sanitaire obligatoire. Les termes « si possible » ne semblent pas compatibles avec le principe d'un vide sanitaire obligatoire. Selon les caractéristiques de l'agent pathogène et sa survie dans l'eau et les fonds de bassin, cette vidange peut être indispensable. Dans ce cas, elle doit par ailleurs être réalisée dans des conditions de sécurité biologique adéquates. Justification, s'il y a lieu :	Did not agree. Removing all water ensures maximum efficacy of fallowing, however not all production systems allow the removal of all water. Where it is feasible, all water should be removed, but this does not preclude other production systems where full drainage is not possible, from being fallowed.

In addition to the considerations outlined in Article 4.7.2., the *Competent Authority* should consider that the duration~~The length~~ of the compulsory ~~statutory~~ *fallowing* period should be based on scientific evidence of the likelihood of ~~free~~ a *pathogenic agent* remaining infective ~~outside its aquaculture host(s)~~ in the local environment, at a level likely to cause an unacceptable risk of re-*infection* of the *aquaculture establishment*. ~~Account should be taken of~~ Factors to be considered include the extent of the *disease outbreak*, local distribution of *susceptible species* and possible *vectors* ~~availability of alternative hosts~~, the survival and infectivity characteristics of the *pathogenic agent* and the relevant ~~local~~ climatological, geographical and hydrographical conditions~~factors~~. I~~n addition, the level of *risk* to the local *aquaculture* industry and wider aquatic resources should be taken into consideration may be included. A scientifically based *risk assessment* approach should be used to determine the length of the *fallowing* period.~~

4.7.5._5_New Caledonia	**Catérogie:** éditorial. **Proposition de texte modifié :** Reformuler cet alinéa pour en clarifier le sens Outre les considérations décrites dans l'article 4.7.2., l'*Autorité compétente* doit prendre en considération ~~que la durée de la période de vide sanitaire obligatoire doit se fonder sur des~~ les preuves scientifiques ~~établissant la probabilité qu'un agent pathogène reste infectieux~~ relatives à l'infectiosité de l'agent pathogène dans le milieu aquatique environnant, pour déterminer la durée de la période de vide sanitaire obligatoire, afin de limiter au maximum ~~à un niveau suffisant pour rendre inacceptable~~ le risque de réinfection de l'établissement d'aquaculture. **Exposé des motifs :** le fait de « rendre inacceptable le risque de réinfection de l'établissement » porte à confusion par rapport au principe du vide sanitaire. **Justification, s'il y a lieu : /**	Agreed to editorial changes to increase clarity of the paragraph.

Article 4.7.6~~7~~4.

~~Instructions for disinfection~~ Disinfection prior to fallowing

Aquatic Animal Health Services ~~Competent Authorities~~ ~~Countries~~ establishing *fallowing* procedures should develop a detailed set of instructions for *disinfection* of *aquaculture establishments* prior to *fallowing* for approval by the *Competent Authority*. These instructions should be~~,~~ ~~where~~ appropriate for the type of production system and circumstances. This should be completed in accordance with Chapter 4.4. and in the case of~~f~~or compulsory *fallowing*, in accordance with Chapters 4.10~~X~~. and 4.11~~Y~~. ~~For this purpose, the instructions set out in Chapter 4.4. of the *Aquatic Code* and in Chapter 1.1.3. of the *Aquatic Manual* should be used as guidelines,~~ taking into account current scientific knowledge on the efficacy of the disinfectants~~treatments~~ for the *pathogenic agent* of concern.

4.7.6._1_New Zealand	**Category:** editorial **Proposed amended text:** **~~Instructions for disinfection~~ Disinfection prior to commencement of fallowing period** *Aquatic Animal Health Services ~~Competent Authorities~~* ~~Countries~~ establishing *fallowing* procedures should develop a detailed set of instructions for *disinfection* of *aquaculture establishments* ~~prior to *fallowing*~~ for approval by the *Competent Authority*. These instructions should be~~,~~ ~~where~~ appropriate for the type of production system and and circumstances. ~~This~~*Disinfection* should be completed prior to the commencement of the *fallowing* period and in accordance with Chapter 4.4. and in the case of~~f~~or compulsory *fallowing*, in accordance with Chapters 4.10~~X~~. and 4.11~~Y~~. ~~For this purpose, the instructions set out in Chapter 4.4. of the *Aquatic Code* and in Chapter 1.1.3. of the *Aquatic Manual* should be used as guidelines,~~ taking into account current scientific knowledge on the	Agreed to editorial changes to clarify that depopulation and disinfection must occur before the fallowing period commences.

	efficacy of the disinfectants~~treatments~~ for the *pathogenic agent* of concern. **Rationale:** Revised paragraph to make it clearer that depopulation/disinfection are all part of the fallowing process, but the fallowing *period* doesn't start until those measures are complete. **Supporting evidence**:	

Article 4.7.78~~5~~.

Restocking after fallowing

An ~~No~~ *aquaculture establishment* that has been subject to~~under~~ compulsory *fallowing* should not be restocked until the compulsory *fallowing* period has been completed and permission from the *Competent Authority* has been received.

When restocking, care should be taken not to use stocks of *aquatic animals* that could ~~would~~ compromise the objectives of the *fallowing* procedure. To increase confidence in the effectiveness of the *fallowing* procedures, all farms subjected to compulsory *fallowing* should have a period of ~~high level~~ official *surveillance* after *susceptible species* have been restocked. The duration and intensity of the *surveillance* should be appropriate for the *disease* in question ~~of concern~~ and subject to the requirements set out in Chapter 1.4., and to the relevant disease-specific chapter in cases of *listed diseases* ~~local conditions~~.

4.7.7._1_New Caledonia	**Catérogie:** éditorial. **Proposition de texte modifié :** Reformuler cet alinéa pour en clarifier le sens Un *établissement d'aquaculture* qui a été soumis à une procédure de *vide sanitaire* obligatoire ne doit pas être repeuplé ~~avant que la période de vide sanitaire obligatoire ait été achevée et~~ tant qu'il n'en a pas reçu l'autorisation par l'*Autorité compétente*. Au moment du repeuplement, il faudra s'assurer que les populations d'*animaux aquatiques* nouvellement introduites ne peuvent pas remettre en cause les bénéfices attendus du *vide sanitaire*. Afin d'accroître la confiance dans l'efficacité des procédures de *vide sanitaire*, les établissements ~~exploitations~~ ayant fait l'objet d'un *vide sanitaire* obligatoire et ayant effectué leur repeuplement avec des *espèces sensibles* devront impérativement se soumettre à une période de *surveillance* officielle. **Exposé des motifs :** le fait de préciser qu'un établissement ne doit pas être repeuplé avant la fin du vide sanitaire semble inutile car le principe même du vide sanitaire est que l'établissement soit vide. Par ailleurs, dans le 2ème alinéa, il est proposé de modifier le terme « exploitations » par « établissements » pour uniformiser avec le reste du chapitre.	Did not agree to revisions to first paragraph as current text provides flexibility to the Competent Authority to address situations as necessary. Agreed to replace 'farms' with 'aquaculture establishments' as it is a Glossary term.

	Justification, s'il y a lieu :/	

CHAPTER 9.2.

INFECTION WITH *APHANOMYCES ASTACI* (CRAYFISH PLAGUE)

Reference	Comment	Aquatic Animals Commission Response
9.2._1_Australia	**Category:** general comment **Proposed amended text:** n/a **Rationale**: Australia strongly supports the ongoing work of the WOAH ad hoc working group to identify the susceptible species of WOAH listed diseases, in this case, the crustacean species susceptible to *Aphanomyces astaci* (crayfish plague). Australia recognises this is a large body of work that is subject to limited specialised resources and appreciates the ongoing efforts of the working group to improve our collective scientific understanding of the taxa that may be potentially affected and at risk to this important disease globally. Australia agrees with the suggested amendments to the text. **Supporting evidence:** n/a	Noted.
9.2._2_Switzerland	**Category:** general comment **Proposed amended text:** n/a **Rationale**: Switzerland supports the proposed update to Article 9.2.2. We have a specific comment as regards the listing of one additional susceptible species. **Supporting evidence:** n/a	Noted.
9.2._3_EU	**Category:** general comment **Proposed amended text:** n/a **Rationale**: The EU supports the proposed update to Article 9.2.2 **Supporting evidence:** n/a	Noted.

[…]

Article 9.2.2.

Scope

The recommendations in this chapter apply to the following species that meet the criteria for listing as susceptible in accordance with Chapter 1.5.: ~~all species of crayfish in all three crayfish families (Cambaridae, Astacidae and Parastacidae). These recommendations also apply to any other *susceptible species* referred to in the *Aquatic Manual* when traded internationally.~~

Family	Scientific name	Common name
Astacidae	*Astacus astacus*	noble crayfish
	Austropotamobius pallipes	no common name
	Pacifastacus leniusculus	signal crayfish
	Pontastacus leptodactylus	Danube crayfish
Cambaridae	*Faxonius spp.* (all species)	N/A
	Procambarus spp. (all species)	N/A
Cambaroididae	*Cambaroides japonicus*	no common name
Parastacidae	*Cherax quadricarinatus*	red claw crayfish
Potamidae	*Potamon potamios*	no common name
Varunidae	*Eriocheir sinensis*	Chinese mitten crab

<table>
<tr>
<td>9.2.2._1_China (People's Republic of)</td>
<td>Category: change

Proposed amended text :
……
<table>
<tr><th>Family</th><th>Scientific name</th><th>Common name</th></tr>
<tr><td rowspan="5">Astacidae</td><td>Astacus astacus</td><td>noble crayfish</td></tr>
<tr><td>Austropotamobius torrentium</td><td>stone crayfish</td></tr>
<tr><td>Austropotamobius pallipes</td><td>no common name</td></tr>
<tr><td>Pacifastacus leniusculus</td><td>signal crayfish</td></tr>
<tr><td>Pontastacus leptodactylus</td><td>Danube crayfish</td></tr>
</table>
Rationale:
The stone crayfish (Austropotamobius torrentium) is a susceptible host for crayfish plague, with multiple studies confirming its vulnerability.
supporting evidence :
1.Aphanomyces +astaci isolate+f 2.infection-apha nomyces-astaci.p</td>
<td>Agreed, evidence was referred to subject matter expert who revised the score to ‘1’. Article 1.5.9. was applied to Astropotamobius and it was listed at the Genus level (see item 5.3. in report).</td>
</tr>
<tr>
<td>9.2.2._2_Peru</td>
<td>Categoría: Editorial</td>
<td>Agreed, that the common name</td>
</tr>
</table>

<table>
<tr>
<td></td>
<td>Texto modificado propuesto: Austropotamobius pallipes / ~~sin nombre común~~ cangrejo de patas blancas

Justificación:
Se reporta abundante literatura sobre el nombre común de la especie Austropotamobius pallipes, siendo “cangrejo de patas blancas”.

Evidencia documentada:
1. Bruno, M. C., Endrizzi, S., Basso, A., Paolini, V., & Pretto, T. (2025). Crayfish plague and microsporidiosis occurrence in wild populations of the white-clawed crayfish Austropotamobius pallipes complex in Trentino (North-East Italy). Journal of Invertebrate Pathology, 108487. https://doi.org/10.1016/j.jip.2025.108487

2. Cartwright, A., Rice, A. R., Dooley, J. S., Smyth, J., & Arnscheidt, J. (2022). Impact of dietary protein content on growth of the white-clawed crayfish Austropotamobius pallipes (Lereboullet, 1858) (Decapoda, Astacidae) in captive rearing for conservation. Crustaceana, 95(5-6), 517-532.

3. Martínez-Ríos, M., Martín-Torrijos, L., Casabella-Herrero, G., Tedesco, P., Machordom, A., & Diéguez-Uribeondo, J. (2023). On the conservation of white-clawed crayfish in the Iberian Peninsula: Unraveling its genetic diversity and structure, and origin. Plos one, 18(10), e0292679. https://doi.org/10.1371/journal.pone.0292679</td>
<td>proposed in the comment is correct. Austropotamobius was revised to be listed at the Genus level, so common name for species no longer reflected in the text.</td>
</tr>
<tr>
<td>9.2.2._3_Switzerland</td>
<td>Category: Addition

Proposed amended text: Listing as susceptible host species Austropotamobius torrentium (currently listed in the Manual as “Species with incomplete evidence for susceptibility”)

Rationale: Susceptibility has been demonstrated (see link below)

Supporting evidence: https://doi.org/10.1016/j.jip.2024.108159</td>
<td>Agreed, see response to comment 9.2.2._1.</td>
</tr>
<tr>
<td>9.2.2._4_Thailand</td>
<td>Category: Change

Proposed amended text: Propose to list susceptible species in Genus Faxonius and Procambarus at the species level, as follow;
<table>
<tr><th>Family</th><th>Scientific name</th><th>Common name</th></tr>
<tr><td rowspan="5">Cambaridae</td><td>~~Faxonius spp.~~ ~~(all species)~~
Faxonius limosus</td><td>~~N/A~~
spinycheek crayfish</td></tr>
<tr><td>Faxonius obscurus</td><td>no common name</td></tr>
<tr><td>Faxonius rusticus</td><td>no common name</td></tr>
<tr><td>~~Procambarus spp.~~ ~~(all species)~~
Procambarus alleni</td><td>~~N/A~~
no common name</td></tr>
<tr><td>Procambarus clarkii</td><td>red swamp crayfish</td></tr>
</table>
Rationale: Determining species susceptible to Aphanomyces astaci should be based on specific scientific evidences for each species by using approach in Article 1.5.4-1.5.6 of Chapter 1.5 to assess susceptibility of species to infection with a specific</td>
<td>Did not agree, Article 1.5.9. was applied and Faxonius was listed at the Genus level as described in the Commission’s September 2025 report (item 6.5.).

Review of the scientific justification for Article 1.5.9. is supplied in the Commission’s September 2025 report (item 7.3.1.).</td>
</tr>
</table>

	pathogenic agent, rather than using criteria in Article 1.5.9 for listing susceptible species at a ranking of Genus to avoid trade barrier due to insufficient of scientific evidences. **Supporting evidence:** According to the Report of the WOAH *ad hoc* Group on the Susceptibility of Crustacean Species to Infection with WOAH-Listed Diseases (April 2025), it was found that: 1. only three species of the Genus *Faxonius* meet the criteria for listing as susceptible to infection with *A. astaci*: *Faxonius limosus*, *Faxonius obscurus*, and *Faxonius rusticus*. The remaining three species, *Faxonius immunis*, *Faxonius propinquus*, and *Faxonius virilis*, currently have insufficient evidence to confirm susceptibility to infection with *A. astaci*. 2. only two species of the Genus *Procambarus* meet the criteria for listing as susceptible to infection with *A. astaci*: *Procambarus alleni* and *Procambarus clarkii*. The remaining six species, *Procambarus enoplosternum*, *Procambarus fallax*, *Procambarus llamasi*, *Procambarus raneyi*, *Procambarus vazquezae* and *Procambarus virginalis,* currently have insufficient evidence to confirm susceptibility to infection with *A. astaci*. We therefore recommend that only the species for which susceptibility has been assessed and confirmed be included in the Aquatic Code and Manual. The application of Article 1.5.9 (Listing of susceptible species at a taxonomic ranking of genus or higher) is subject to important limitations, as it may result in an inappropriate generalization of susceptibility from a limited number of species to an entire genus. Specifically, when several species within a genus have only incomplete or inconclusive evidence of susceptibility, they cannot be classified as susceptible under Article 1.5.9. Instead, such species must remain listed as “species with incomplete evidence” in the corresponding section of the WOAH Aquatic Manual.	
9.2.2._5_EU	**Category:** Addition **Proposed amended text:** including *Cherax destructor* as ’Susceptible **Rationale:** evidence of infection in farmed population of *Cherax destructor* showing increased mortality and clinical signs (loss of appendages, paralysis upside down) melanisation of the cuticle and histological evidence of hyphae branching in the cuticle and hypodermis, supported by molecular identification of *A. astaci* by end-point PCR and sequencing. **Supporting evidence:** IAA 19 Conference 2012, Innsbruck, Austria. Quaglio et al., 2012. First occurrence of *Aphanomyces astaci* epidemic infection in cultured yabby *Cherax destructor* (Clark, 1936) in Northern Italy. Book of abstract, page 88	Did not agree, the reference was referred to a subject matter expert. On review of the reference it did not provide enough information to assess all stages as described in Chapter 1.5. and *Cherax destructor* could not be scored based on the provided reference.

	https://www.astacology.org/docs/symposia/IAA19/docs/IAA19_Abstracts.pdf	
9.2.2._6_EU	**Category:** Addition ’ **Proposed amended text:** listing as susceptible host species *’Austropotamobius torrentium* (currently listed in the Manual as ‘Species with incomplete evidence for susceptibility) **Rationale:** evidence of infection in natural population of *A. torrentium* showing micromelanisation of the cuticle, supported by isolation of *A. astaci* genotype As from crayfish tissues and identification of the isolate by PCR followed by sequencing; additionally evidence of mortality induced by experimental infection by immersion with *A. astaci* genotype PsI zoospores. **Supporting evidence:** Jussila et al. 2017 *Aphanomyces astaci* isolate from latently infected stone crayfish (*Austropotamobius torrentium*) population is virulent. http://dx.doi.org/10.1016/j.jip.2017.07.003	Agreed, see response to comment 9.2.2._1.

[…]

CHAPTER 10.3.

INFECTION WITH *GYRODACTYLUS SALARIS*

[…]

Reference	Comment	Aquatic Animals Commission Response
10.3._1_Australia	**Category:** General **Proposed amended text:** n/a **Rationale**: Australia has made a joint submission with fellow QUADS alliance members (United Kingdom, New Zealand, Canada, USA) regarding the suggested changes made to articles 10.3.5. and 10.3.6. of Chapter 10.3. 'Infection with *Gyrodactylus salaris'*. **Supporting evidence:** n/a	Noted.
10.3._2_Norway	**Category**: general **Proposed amended text**: Not relevant **Rationale:** Based on our experience, not all populations of Atlantic salmon show clinical signs of infection with *G. salaris*. Suggested text additions are provided below. **Supporting evidence, if relevant:** not relevant	The Commission requested the reference laboratory expert for infection with *Gyrodactylus salaris* on the clinical expression of *G. salaris* in Atlantic salmon. The expert reported that clinical cases are not always seen in Baltic populations of Atlantic salmon, and only a few parasites would be observed and no clinical signs. In the case of Baltic salmon infection with *G. salaris* would not be detected without targeted surveillance. The following references were shared: • Bakke TA, Harris PD, Hansen H, Cable J, Hansen LP. Susceptibility of Baltic and East Atlantic salmon *Salmo salar* stocks to *Gyrodactylus salaris* (Monogenea). Dis Aquat Organ. 2004;58 2-3:171–7; doi: 10.3354/dao058171. • Kania PW, Jørgensen TR, Buchmann K. Differentiation between a pathogenic and a non-pathogenic form of *Gyrodactylus salaris* using PCR-RFLP. J Fish Dis. 2007;30 2:123–6. • Olstad K, Robertsen G, Bachmann L, Bakke TA. Variation in host preference

		within *Gyrodactylus salaris* (Monogenea): an experimental approach. Parasitology. 2007;134 4:589–97; doi: 10.1017/S0031182006001715.
10.3._3_EU	**Category**: general **Proposed amended text**: Not relevant **Rationale:** The EU would like to reiterate its previous concerns about the use of historical freedom to obtain disease freedom for infection with *G. salaris*. Please, see proposed amendment below **Supporting evidence, if relevant:** not relevant	Noted.
10.3._4_Region of the Americas	**<u>Proposal/Comment: / Propuesta/Comentario:</u>** On behalf of the 33 countries of the Region of the Americas, we appreciate the Aquatic Animal Health Standards Commission ('Commission') for their careful consideration of the comments and concerns raised during the 92nd General Session last May 2025, as well as the recommendation to the Assembly that pathway 2 (historical freedom) for *Gyrodactylus salaris* (*G. salaris*) be placed under study and discussed by the Commission at its Sepetmber 2025 meeting. The Region of the Americas urges the Commission to reconsider creating an exception for a new approach to the application of pathway 2 (historical freedom) solely for GS as we have concerns about setting a precedent that could negatively impact trade. / En nombre de los 33 países de la Región de las Américas, agradecemos a la Comisión de Normas Sanitarias para Animales Acuáticos ('la Comisión') su cuidadosa consideración de los comentarios y preocupaciones planteados durante la 92.ª Sesión General celebrada en mayo de 2025, así como la recomendación a la Asamblea de que la vía 2 (libertad histórica) para *Gyrodactylus salaris* (*G. salaris*) sea objeto de estudio y debate por parte de la Comisión en su reunión de septiembre de 2025. La Región de las Américas insta a la Comisión a que reconsidere la creación de una excepción para un nuevo enfoque de la aplicación de la vía 2 (ausencia histórica) únicamente para el GS, ya que nos preocupa que se siente un precedente que podría afectar negativamente al comercio. **<u>Rationale:/ Justificación:</u>**	See point 5.4. of this meeting report.

	The Region of the Americas recommends removing the proposal described in Annex 11 for Infection with *Gyrodactylus salaris* (*G.salaris*) which proposes that pathway 2 (historical pathway) should only be used for Atlantic salmon (in both Articles 10.3.5. and 10.3.6). Restricting the use of the historical pathway for self-declaration of freedom to only those countries/regions with Atlantic salmon in freshwater ecosystems, limits the applicability of this pathway, and implies that only Atlantic salmon monocultures are eligible for assessment via the historical pathway. If so, this effectively negates the use of this pathway for any freedom claims at zone or country levels where there is more than one of the susceptible species present. This proposed change sets a precedent which directly counters the utilization of risk-based methods which purposively focuses resources (whether observational or laboratory) on subpopulations with the highest probability of infection. As long as the target species for surveillance (e.g., Atlantic salmon) are representative of the larger (e.g., multi-species) population, risk-based approaches can dramatically enhance the quality of detection systems. This determination (and supporting rationale) should be made by individual countries within their self-declaration as surveillance designs and suitability are highly context specific. This proposed change also sets a precedent for how the Commission may approach other pathogens in the future, raising concerns about transparency and predictability that may directly impact trade for Members. We feel the Commission has not provided sufficient evidence to justify treating *G. salaris* differently from other WOAH-listed pathogens, and that Chapter 1.4. already includes provisions to address the Commission's concerns. Specifically, Chapter 1.4. already accounts for the option to provide additional data from targeted surveillance to strengthen historical freedom claims via pathway 2 if some susceptible species don't show clinical signs to a given pathogen of concern. It is unclear why this approach is being proposed for *G. salaris*, and not for any other pathogens where historical freedom is included as a pathway for self-declaration of freedom such as those as having been assessed as broad host range. In these cases, evidence for assessment of susceptibility in some species are evaluated against the criteria outlined in Chapter 1.5. and used to list all species as susceptible. In these cases, we are not aware if all species in the Genus or Family would show clinical signs, however the historical freedom pathway has remained for these pathogens. If the Commission plans to single out individual	

	species where pathway 2 applies, such as is proposed for *G. salaris*, it should instead be ideally accomplished through standardized criteria to further distinguish levels of susceptibility which is supported by specific scientific evidence (which is not currently outlined in Chapter 1.5.). / La Región de las Américas recomienda eliminar la propuesta descrita en el anexo 11 relativa a la infección por *Gyrodactylus salaris* (*G. salaris*), que propone que la vía 2 (vía histórica) solo se utilice para el salmón del Atlántico (tanto en el artículo 10.3.5 como en el 10.3.6). Restringir el uso de la vía histórica para la autodeclaración de ausencia de la enfermedad únicamente a aquellos países/regiones con salmón del Atlántico en ecosistemas de agua dulce limita la aplicabilidad de esta vía e implica que solo los monocultivos de salmón del Atlántico pueden ser evaluados mediante la vía histórica. De ser así, esto niega efectivamente el uso de esta vía para cualquier declaración de ausencia de la enfermedad a nivel de zona o de país donde haya más de una especie susceptible presente. El cambio propuesto sienta un precedente que contrarresta directamente la utilización de métodos basados en el riesgo que centran deliberadamente los recursos (ya sean observacionales o de laboratorio) en las subpoblaciones con mayor probabilidad de infección. Siempre que las especies objeto de vigilancia (por ejemplo, el salmón del Atlántico) sean representativas de la población más amplia (por ejemplo, multiespecífica), los enfoques basados en el riesgo pueden mejorar drásticamente la calidad de los sistemas de detección. Esta determinación (y la justificación que la respalda) debe ser realizada por cada país en su propia declaración, ya que los diseños y la idoneidad de la vigilancia dependen en gran medida del contexto específico. Este cambio propuesto también sienta un precedente sobre cómo la Comisión podría abordar otros patógenos en el futuro, lo que suscita inquietudes sobre la transparencia y la previsibilidad que podrían afectar directamente al comercio de los Miembros. Consideramos que la Comisión no ha aportado pruebas suficientes que justifiquen tratar a *G. salaris* de forma diferente a otros patógenos incluidos en la lista de la OMSA, y que el capítulo 1.4 ya incluye disposiciones para abordar las preocupaciones de la Comisión. En concreto, el capítulo 1.4 ya contempla la opción de proporcionar datos adicionales procedentes de la vigilancia específica para reforzar las declaraciones de ausencia histórica a través de la vía 2 si algunas especies susceptibles no muestran signos clínicos de un determinado patógeno preocupante. No está claro por qué se	

	propone este enfoque para *G. salaris* y no para otros patógenos en los que la ausencia histórica se incluye como vía para la autodeclaración de ausencia, como los que han sido evaluados como de amplio rango de hospedadores. En estos casos, las pruebas para evaluar la susceptibilidad en algunas especies se evalúan según los criterios descritos en el capítulo 1.5 y se utilizan para incluir a todas las especies como susceptibles. En estos casos, no sabemos si todas las especies del género o la familia mostrarían signos clínicos, sin embargo, la vía de la ausencia histórica se ha mantenido para estos patógenos. Si la Comisión tiene previsto seleccionar especies individuales a las que se aplica la vía 2, como se propone para *G. salaris*, lo ideal sería que se hiciera mediante criterios normalizados para distinguir mejor los niveles de susceptibilidad, respaldados por pruebas científicas específicas (que actualmente no se describen en el capítulo 1.5).	
10.3._5_QUADS Alliance	**Category**: general **Proposed amended text**: Not relevant **Rationale:** On behalf of the QUADS Alliance (Australia, Canada, New Zealand, the United Kingdom and the United States of America), we appreciate the Aquatic Animal Health Standards Commission ('Commission') for their careful consideration of the comments and concerns raised during the 92nd General Session last May 2025, as well as the recommendation to the Assembly that pathway 2 (historical freedom) for *Gyrodactylus salaris* (*G. salaris*) be placed under study and discussed by the Commission at its Sepetmber 2025 meeting. We urge the Commission to reconsider creating an exception for a new approach to the application of pathway 2 (historical freedom) solely for GS as we have concerns about setting a precedent that could negatively impact trade. Rationale: The QUADS Alliance recommends removing the proposal described in Annex 11 for Infection with *Gyrodactylus salaris* (*G.salaris*) which proposes that pathway 2 (historical pathway) should only be used for Atlantic salmon (in both Articles 10.3.5. and 10.3.6). Restricting the use of the historical pathway for self-declaration of freedom to only those countries/regions with Atlantic salmon in freshwater ecosystems, limits the applicability of this pathway, and implies that only Atlantic salmon monocultures are eligible for assessment via the historical pathway. If so, this effectively negates the	See point 5.4. of this meeting report.

	use of this pathway for any freedom claims at zone or country levels where there is more than one of the susceptible species present. This proposed change sets a precedent which directly counters the utilization of risk-based methods which purposively focuses resources (whether observational or laboratory) on subpopulations with the highest probability of infection. As long as the target species for surveillance (e.g., Atlantic salmon) are representative of the larger (e.g., multi-species) population, risk-based approaches can dramatically enhance the quality of detection systems. This determination (and supporting rationale) should be made by individual countries within their self-declaration as surveillance designs and suitability are highly context specific. This proposed change also sets a precedent for how the Commission may approach other pathogens in the future, raising concerns about transparency and predictability that may directly impact trade for Members. We feel the Commission has not provided sufficient evidence to justify treating *G. salaris* differently from other WOAH-listed pathogens, and that Chapter 1.4. already includes provisions to address the Commission's concerns. Specifically, Chapter 1.4. already accounts for the option to provide additional data from targeted surveillance to strengthen historical freedom claims via pathway 2 if some susceptible species don't show clinical signs to a given pathogen of concern. It is unclear why this approach is being proposed for *G. salaris*, and not for any other pathogens where historical freedom is included as a pathway for self-declaration of freedom such as those as having been assessed as broad host range. In these cases, evidence for assessment of susceptibility in some species are evaluated against the criteria outlined in Chapter 1.5. and used to list all species as susceptible. In these cases, we are not aware if all species in the Genus or Family would show clinical signs, however the historical freedom pathway has remained for these pathogens. If the Commission plans to single out individual species where pathway 2 applies, such as is proposed for *G. salaris*, it should instead be ideally accomplished through standardized criteria to further distinguish levels of susceptibility which is supported by specific scientific evidence (which is not currently outlined in Chapter 1.5.). **Supporting evidence, if relevant:** not relevant	

Article 10.3.5.

Country free from infection with *G. salaris*

If a country shares water bodies with other countries, it can only make a self-declaration of freedom from infection with *G. salaris* if all shared water bodies are within countries or *zones* declared free from infection with *G. salaris* (see Article 10.3.6.).

As described in Article 1.4.4., a Member Country may make a self-declaration of freedom from infection with *G. salaris* for its entire *territory* if it can demonstrate that:

1) none of the *susceptible species* referred to in Article 10.3.2. are present and *basic biosecurity conditions* have been continuously met for at least the last six months;

OR

2) ~~pathway 2 (historical freedom) is [under study]~~ there has been no occurrence of infection with *G. salaris* for at least the last 15 years, and;

10.3.5._1_United States	**Category:** change **Proposed amended texts (or precise suggested deletion):** Please see the proposed change in blue – "there has been no occurrence of infection with *G. salaris* for at least the last ~~15~~ 10 years, and;" **Rationale:** We feel this should remain at 10 years because *G. salaris* is viviparous, with rapid reproduction cycles; thus, supporting a shorter default basic biosecurity period, rather than >10 years.	Did not agree. The Commission referred to the Recommendations for periods of basic biosecurity conditions and targeted surveillance for the disease-specific chapters of the WOAH Aquatic Animal Health which provided the review of scientific evidence to provide rationale for 15 years. The default 10-year period assumes an annual detection probability of approximately 30%, leading to 95% confidence of freedom after 10 years of absence of detection.(Article 1.4.9 pt.2,b) However, for *G. salaris*, several biological, ecological, and production-related factors reduce the annual probability of detection below 30%. Consequently, a longer period (approximately 15 years) is scientifically justified to achieve ≥95% confidence of freedom. Due to: Variable clinical expression; Presence of asymptomatic carrier hosts; Temperature-dependent transmission; Wild population involvement;

		Long host lifecycle.

a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and

b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years;

10.3.5._2_Australia	**Category:** General **Proposed amended text:** n/a **Rationale**: Australia suggests that a reference citation should be included in the text to support the rationale of recommending a disease free period of **15 years** at 2 b). **Supporting evidence:** n/a	The Recommendations for periods of basic biosecurity conditions and targeted surveillance for the disease-specific chapters of the WOAH Aquatic Animal Health provided the review of scientific evidence to provide rationale for 15 years. The *Aquatic Code* does not include textual references, however the report is available on the WOAH website.
10.3.5._3_United States	**Category:** change **Proposed amended texts (or precise suggested deletion):** Please see the proposed change in blue – "b) basic biosecurity conditions as described in Chapter 1.4. have been continuously met for at least the last ~~15~~ 10 years;" **Rationale:** We feel this should remain at 10 years because *G. salaris* is viviparous, with rapid reproduction cycles; thus, supporting a shorter default basic biosecurity period, rather than >10 years.	See response 10.3.5._1.

Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other *susceptible species* that do not exhibit clinical signs then pathway 2 is not suitable.

10.3.5._4_Canada	**Comment Category:** Deletion **Proposed amended text:** a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years;	See point 5.4. of this meeting report, and the response to comments 10.3._2. and 10.3.5._5.

	~~Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*)~~ **Rationale:** Chapter 1.4. provides guidance for situations where animals may not show clinical signs. Article 1.4.3. point 2 indicates targeted surveillance may also be used in the Historical freedom pathway if appropriate and table 1.1. indicates that secondary evidence can be provided through targeted surveillance "in populations where passive surveillance is not appropriate". This directly applies to declarations of freedom for *G.salaris* for Members that have Atlantic Salmon and other susceptible species. This principal was specifically developed for cases where passive surveillance may not work, either for cases where clinical signs are not present or animals are not readily available such as wild animals. Specifically, Article 1.4.8, Point 1.d.ii states: "d. for populations of susceptible wild *aquatic animals*, they should: … ii. be epidemiologically linked to farmed populations, such that if the *disease* were to occur in wild *aquatic animal* populations it would be observed and reported in adjacent farmed populations." Article 1.4.12. point 1 states that Pathway 2 can be used if the early detection system "is sufficiently sensitive to detect the *disease* should it occur," and the follow paragraph states "The *early detection system* needs to represent all the *susceptible species* populations in the country or *zone*. If a *Competent Authority* cannot demonstrate that the required characteristics are fulfilled, due to a country"s circumstances (e.g. nature of the *early detection system*, environmental conditions, nature of the *aquaculture*), this pathway is not considered valid. Instead, an alternative pathway that utilises *targeted surveillance* data will be required, or the *passive surveillance* information will need to be supplemented with *targeted surveillance* data (see below)." Therefore, countries should be allowed to use pathway 2 for any species if the countries or zone can meet these requirements. There is no need to limit pathway 2 by species, when the limits for substantiating freedom through pathway 2 are already sufficient.Many Diseases, particularly those with broad host ranges, have susceptible species that may not show clinical signs of disease. However similar text was not added to other disease specific chapters. Singling out *Gryodactylus salaris* to apply a new approach for species that do not show clinical signs is inconsistent with the approach that was taken for all other diseases which were adopted in May 2025 Currently, Chapter 1.5. Criteria for listing species as susceptible to infection with a specific pathogen and the final assessments (score 1) of the ad hoc Group which are the basis for establishment of the list of susceptible species in	

	Articles X.X.2. does not differentiate between susceptible species that show Pathology/Clinical signs (stage 3 point C) and those that don't. In fact the criteria for Stage 3 point C for *G.salaris* was not applied for a single species assessed and for other diseases, point 3C does not have to be assessed if other criteria solely (3A) or in combination (2 of B,C,D) meet the criteria. The term 'susceptible species' and approach to establish susceptible species outlined in Chapter 1.5. is rigorous, based on scientific evidence and allows equal application of the definition and listing of species in Articles X.X.2. throughout the Code. Application of a different process to establish which species show clinical signs of disease or not for a single disease, that is not substantiated by the transparent criteria outlined in the Aquatic Code and ad hoc Group reports undermines the principals of the Aquatic Code and the significant amount of work done by the Commission since 2015.	
10.3.5._5_New Zealand	**Category: Editorial** **Proposed amended text:** 2) ~~pathway 2 (historical freedom) is [under study]~~ there has been no occurrence of infection with *G. salaris* for at least the last 15 years, and; a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years; Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other *susceptible species* that do not exhibit clinical signs then pathway 2 is not suitable. **Rationale:** Formatting – alignment - so it is clear this sentence is not a part of the bulleted list. **Supporting evidence:** n/a	Noted. To increase clarity, in point 2 a) of both Article 10.3.5. and Article 10.3.6., the Commission introduced a reference to Chapter 2.3.3 of the Aquatic Manual, highlighted the need to take into consideration relevant clinical information. As outlined in the Aquatic Manual. Furthermore, the last paragraph from point 2 of both Article 10.3.5. and Article 10.3.6., has been revised.

10.3.5._6_Norway	**Category**: addition **Proposed amended text**: Not relevant Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*) populations proven to show clinical signs of infection with *G salaris*. **Rationale:** Not all populations of Atlantic salmon show clinical signs of infection with *G. salaris*. **Supporting evidence, if relevant:** not relevant	See response to comments 10.3._2. and 10.3.5._5.
10.3.5._7_UK	**Category:** Deletion **Proposed amended text:** 2) ~~pathway 2 (historical freedom) is [under study]~~ there has been no occurrence of infection with *G. salaris* for at least the last 15 years, and; a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years; ~~Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other *susceptible species* that do not exhibit clinical signs then pathway 2 is not suitable.~~ **Rationale:** We feel that this statement sets a precedent that is not applied to any other aquatic disease listed by WOAH. There are many diseases for which susceptible species may show varying degrees of clinical expression, including no clinical expression at all, yet historical freedom is still offered as a viable means of self-declarations of disease freedom because it is expected that countries will employ appropriate surveillance and biosecurity measures to suit their susceptible populations as outlined in Chapter 1.4. We know of no justification why infection with *Gyrodactylus salaris* should be treated any differently, making this text unnecessarily restrictive to countries with multiple susceptible species beyond Atlantic salmon.	See response to comments 10.3._2. and 10.3.5._5.

10.3.5._8_United States	**Category:** deletion **Proposed amended texts (or precise suggested deletion):** Please see the proposed deletion in blue – "~~Pathway 2 (historical freedom) is only applicable to make a self-declaration of freedom from disease for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other susceptible species that do not exhibit clinical signs then pathway 2 is not suitable.~~" **Rationale:** We appreciate the Aquatic Animal Health Standards Commission ('Commission') for their careful consideration of the comments and concerns raised during the 92nd General Session last May 2025, as well as the recommendation to the Assembly that pathway 2 (historical freedom) for *Gyrodactylus salaris* (*G. salaris*) be placed under study and discussed by the Commission at its September 2025 meeting. We urge the Commission to reconsider creating an exception for a new approach to the application of pathway 2 (historical freedom) solely for GS as we have concerns about setting a precedent that could negatively impact trade. We recommend removing the proposal described in Annex 11 for Infection with *Gyrodactylus* salaris (*G.salaris*) which proposes that pathway 2 (historical pathway) should only be used for Atlantic salmon (in both Articles 10.3.5. and 10.3.6). Restricting the use of the historical pathway for self-declaration of freedom to only those countries/regions with Atlantic salmon in freshwater ecosystems, limits the applicability of this pathway, and implies that only Atlantic salmon monocultures are eligible for assessment via the historical pathway. If so, this effectively negates the use of this pathway for any freedom claims at zone or country levels where there is more than one of the susceptible species present. This proposed change sets a precedent which directly counters the utilization of risk-based methods which purposively focuses resources (whether observational or laboratory) on subpopulations with the highest probability of infection. As long as the target species for surveillance (e.g., Atlantic salmon) are representative of the larger (e.g., multi-species) population, risk-based approaches can dramatically enhance the quality of detection systems. This determination (and supporting rationale) should be made by individual countries within their self-declaration as surveillance designs and suitability are highly context specific. This proposed change also sets a precedent for how the Commission may approach other pathogens in the future, raising concerns about transparency and predictability that may directly impact trade for Members. We feel the Commission has not provided sufficient evidence to justify treating *G. salaris* differently from other WOAH-listed pathogens, and that Chapter 1.4. already includes provisions to address the Commission's concerns. Specifically, Chapter	See point 5.4. of this meeting report.

	1.4. already accounts for the option to provide additional data from targeted surveillance to strengthen historical freedom claims via pathway 2 if some susceptible species don't show clinical signs to a given pathogen of concern. It is unclear why this approach is being proposed for *G. salaris*, and not for any other pathogens where historical freedom is included as a pathway for self-declaration of freedom such as those as having been assessed as broad host range. In these cases, evidence for assessment of susceptibility in some species are evaluated against the criteria outlined in Chapter 1.5. and used to list all species as susceptible. In these cases, we are not aware if all species in the Genus or Family would show clinical signs, however the historical freedom pathway has remained for these pathogens. If the Commission plans to single out individual species where pathway 2 applies, such as is proposed for *G. salaris*, it should instead be ideally accomplished through standardized criteria to further distinguish levels of susceptibility which is supported by specific scientific evidence (which is not currently outlined in Chapter 1.5.).	
10.3.5._9_EU	**Category:** Addition **Proposed amended text:** Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for strains of Atlantic salmon (*Salmo salar*) in which it has been demonstrated that G. salaris causes clinical disease. **Rationale**: There are regional strains of Atlantic salmon where the infection is always subclinical. *G. salaris* has never been seen to cause clinical disease in the Baltic, although Baltic strains of Atlantic salmon (*Salmo salar*) are abundant and they are known to be infected with *G. salaris*. Without targeted active monitoring *G. salaris* would never have been identified in countries in the Baltic area as Finland.	See response to comments 10.3._2. and 10.3.5._5.

OR

3) *targeted surveillance*, as described in Chapter 1.4., has been in place for at least the last three years without detection of *G. salaris*, and *basic biosecurity conditions* have been continuously met and have been in place for at least two years prior to commencement of *targeted surveillance*;

OR

4) it previously made a self-declaration of freedom from infection with *G. salaris* and subsequently lost its free status due to the detection of *G. salaris* but the following conditions have been met:

   a) on detection of *G. salaris*, the affected area was declared an *infected zone* and a *protection zone* was established; and

   b) infected populations within the *infected zone* have been killed and disposed of by means that minimise the likelihood of further transmission of *G. salaris*, and the appropriate *disinfection* procedures (as described in Chapter 4.4.) have been completed followed by *fallowing* as described in Chapter 4.7.; and

c) previously existing *basic biosecurity conditions* have been reviewed and modified as necessary and have continuously been in place since eradication of infection with *G. salaris*; and

d) *targeted surveillance*, as described in Chapter 1.4., has been in place for:

i) at least the last three years in wild and farmed *susceptible species* without detection of *G. salaris*; or

ii) at least the last one year without detection of *G. salaris* if affected *aquaculture establishments* were not epidemiologically connected to wild populations of *susceptible species*.

10.3.5._10_New Zealand	**Category:** editorial **Proposed amended text:** d) *targeted surveillance*, as described in Chapter 1.4., has been in place for: i) at least the last three years in wild and farmed *susceptible species,* without detection of *G. salaris*; or ii) at least the last one year in farmed *susceptible species*, without detection of *G. salaris* if affected *aquaculture establishments* were not epidemiologically connected to wild populations of *susceptible species*. **Rationale:** Suggesting edits to make it clearer that point is referring to farmed population only. If this is not what this point is referring to, then suggesting clarifying this. **Supporting evidence:** n/a	Did not agree, as this point states it refers to *susceptible species*; therefore, would not require further clarification.

In the meantime, the part of the country outside the *infected zone* and *protection zone* may be declared a *free zone* as described in Article 1.4.4.

Article 10.3.6.

Zone free from infection with *G. salaris*

If a *zone* extends over the *territory* of more than one country, it can only be declared a *zone* free from infection with *G. salaris* if all of the relevant *Competent Authorities* confirm that all relevant conditions have been met.

As described in Article 1.4.4., a Member Country may make a self-declaration of freedom from infection with *G. salaris* for a *zone* within its *territory* if it can demonstrate that:

1) none of the *susceptible species* referred to in Article 10.3.2. are present and *basic biosecurity conditions* have been continuously met for at least the last six months;

OR

2) ~~pathway 2 (historical freedom) is [under study]~~ there has been no occurrence of infection with *G. salaris* for at least the last 15 years, and;

10.3.6._1_United States	**Category:** change **Proposed amended texts (or precise suggested deletion):** Please see the proposed change in blue – "there has been no occurrence of infection with *G. salaris* for at least the last ~~15~~ 10 years, and;" **Rationale:** We feel this should remain at 10 years because *G. salaris* is viviparous, with rapid reproduction cycles; thus, supporting a shorter default basic biosecurity period, rather than >10 years.	See response 10.3.5._1.

a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and

b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years;

10.3.6._2_Australia	**Category:** General **Proposed amended text:** n/a **Rationale**: Australia suggests that a reference citation should be included in the text to support the rationale of recommending a disease free period of **15 years** at 2 b). **Supporting evidence:** n/a	See response 10.3.5._2.
10.3.6._3_United States	**Category:** change **Proposed amended texts (or precise suggested deletion):** Please see the proposed change in blue – "there has been no occurrence of infection with *G. salaris* for at least the last ~~15~~ 10 years, and;" **Rationale:** We feel this should remain at 10 years because *G. salaris* is viviparous, with rapid reproduction cycles; thus, supporting a shorter default basic biosecurity period, rather than >10 years.	See response 10.3.5._1.

Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other *susceptible species* that do not exhibit clinical signs then pathway 2 is not suitable.

10.3.6._4_Canada	**Comment Category:** Deletion **Proposed amended text:** a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years; ~~Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*)~~ **Rationale:** Chapter 1.4. provides guidance for situations where animals may not show clinical signs. Article 1.4.3. point 2 indicates targeted surveillance may also be used in the Historical freedom pathway if appropriate and table 1.1. indicates that secondary evidence can be provided through targeted surveillance "in populations where passive surveillance is not appropriate". This directly applies to declarations of freedom for *G. salaris* for Members that have Atlantic Salmon and other susceptible species. This principal was specifically developed for cases where passive surveillance may not work, either for cases where clinical signs are not present or animals are not readily available such as wild animals. Specifically, Article 1.4.8, Point 1.d.ii states: "d. for populations of susceptible wild *aquatic animals*, they should: … ii. be epidemiologically linked to farmed populations, such that if the *disease* were to occur in wild *aquatic animal* populations it would be observed and reported in adjacent farmed populations." Article 1.4.12. point 1 states that Pathway 2 can be used if the early detection system "is sufficiently sensitive to detect the *disease* should it occur," and the follow paragraph states "The *early detection system* needs to represent all the *susceptible species* populations in the country or *zone*. If a *Competent Authority* cannot demonstrate that the required characteristics are fulfilled, due to a country"s circumstances (e.g. nature of the *early detection system*, environmental conditions, nature of the *aquaculture*), this pathway is not considered valid. Instead, an alternative pathway that utilises *targeted surveillance* data will be required, or the *passive surveillance* information will need to be supplemented with *targeted surveillance* data (see below)." Therefore, countries should be allowed to use pathway 2 for any species if the countries or zone can meet these	See point 5.4. of this meeting report, and the response to comments 10.3._2. and 10.3.5._5.

	requirements. There is no need to limit pathway 2 by species, when the limits for substantiating freedom through pathway 2 are already sufficient.Many Diseases, particularly those with broad host ranges, have susceptible species that may not show clinical signs of disease. However similar text was not added to other disease specific chapters. Singling out *G. salaris* to apply a new approach for species that do not show clinical signs is inconsistent with the approach that was taken for all other diseases which were adopted in May 2025. Currently, Chapter 1.5. Criteria for listing species as susceptible to infection with a specific pathogen and the final assessments (score 1) of the specific ad hoc Group reports which are the basis for establishment of the list of susceptible species for inclusion in the Code does not differentiate between susceptible species that show Pathology/Clinical signs (stage 3 point C) and those that don't. In fact the criteria for Stage 3 point C for *G. salaris* was not applied for a single species assessed and for other diseases, point 3C does not have to be assessed if other criteria solely (3A) or in combination (2 of B,C,D) meet the criteria. The term 'susceptible species' and approach to establish susceptible species outlined in Chapter 1.5. is rigorous, based on scientific evidence and allows equal application of the definition and listing of species in Articles X.X.2. throughout the Code. Application of a different process to establish which species show clinical signs of disease or not for a single disease, that is not substantiated by the transparent criteria outlined in the Aquatic Code and ad hoc Group reports undermines the principals of the Aquatic Code and the significant amount of work done by the Commission since 2015.	
10.3.6._5_New Zealand	**Category:** Editorial **Proposed amended text:** 2) ~~pathway 2 (historical freedom) is [under study]~~ there has been no occurrence of infection with *G. salaris* for at least the last 15 years, and; a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years; Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study	See response to comments 10.3._2. and 10.3.5._5.

	population comprises other *susceptible species* that do not exhibit clinical signs then pathway 2 is not suitable. **Rationale:** As per previous comment – edited formatting/alignment - so it is clear this sentence is not a part of the bulleted list. **Supporting evidence:** n/a	
10.3.6._6_Norway	**Category**: addition **Proposed amended text**: Not relevant Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*) populations proven to show clinical signs of infection with *G salaris*. **Rationale:** Not all populations of Atlantic salmon show clinical signs of infection with *G. salaris*. **Supporting evidence, if relevant:** not relevant	See response to comments 10.3._2. and 10.3.5._5.
10.3.6._7_UK	**Category:** Deletion **Proposed amended text:** 2) ~~pathway 2 (historical freedom) is [under study]~~ there has been no occurrence of infection with *G. salaris* for at least the last 15 years, and; a) the Member Country can demonstrate that conditions are conducive to the clinical expression of infection with *G. salaris*, as described in Article 1.4.8. of Chapter 1.4.; and b) *basic biosecurity conditions* as described in Chapter 1.4. have been continuously met for at least the last 15 years; ~~Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other *susceptible species* that do not exhibit clinical signs then pathway 2 is not suitable.~~ **Rationale:** We feel that this statement sets a precedent that is not applied to any other aquatic disease listed by WOAH. There are many diseases for which susceptible species may show varying degrees of clinical expression, including no clinical expression at all, yet historical freedom is still offered as a viable means of self-declarations of disease freedom because it is expected that countries will employ appropriate surveillance and biosecurity measures to suit their	See response to comments 10.3._2. and 10.3.5._5.

	susceptible populations as outlined in Chapter 1.4. We know of no justification why infection with *Gyrodactylus salaris* should be treated any differently, making this text unnecessarily restrictive to countries with multiple susceptible species beyond Atlantic salmon.	
10.3.6._8_United States	**Category:** deletion **Proposed amended texts (or precise suggested deletion):** Please see the proposed deletion in blue – "~~Pathway 2 (historical freedom) is only applicable to make a self-declaration of freedom from disease for infection with *G. salaris* for Atlantic salmon (*Salmo salar*). If the study population comprises other susceptible species that do not exhibit clinical signs then pathway 2 is not suitable.~~" **Rationale:** We appreciate the Aquatic Animal Health Standards Commission ('Commission') for their careful consideration of the comments and concerns raised during the 92nd General Session last May 2025, as well as the recommendation to the Assembly that pathway 2 (historical freedom) for *Gyrodactylus salaris* (*G. salaris*) be placed under study and discussed by the Commission at its September 2025 meeting. We urge the Commission to reconsider creating an exception for a new approach to the application of pathway 2 (historical freedom) solely for GS as we have concerns about setting a precedent that could negatively impact trade. We recommend removing the proposal described in Annex 11 for Infection with *Gyrodactylus* salaris (*G.salaris*) which proposes that pathway 2 (historical pathway) should only be used for Atlantic salmon (in both Articles 10.3.5. and 10.3.6). Restricting the use of the historical pathway for self-declaration of freedom to only those countries/regions with Atlantic salmon in freshwater ecosystems, limits the applicability of this pathway, and implies that only Atlantic salmon monocultures are eligible for assessment via the historical pathway. If so, this effectively negates the use of this pathway for any freedom claims at zone or country levels where there is more than one of the susceptible species present. We feel the Commission has not provided sufficient evidence to justify treating *G. salaris* differently from other WOAH-listed pathogens, and that Chapter 1.4. already includes provisions to address the Commission's concerns. Specifically, Chapter 1.4. already accounts for the option to provide additional data from targeted surveillance to strengthen historical freedom claims via pathway 2 if some susceptible species don't show clinical signs to a given pathogen of concern. It is unclear why this approach is	See point 5.4. of this meeting report, and the response to comments 10.3._2. and 10.3.5._5.

	being proposed for *G. salaris*, and not for any other pathogens where historical freedom is included as a pathway for self-declaration of freedom such as those as having been assessed as broad host range. In these cases, evidence for assessment of susceptibility in some species are evaluated against the criteria outlined in Chapter 1.5. and used to list all species as susceptible. In these cases, we are not aware if all species in the Genus or Family would show clinical signs, however the historical freedom pathway has remained for these pathogens. If the Commission plans to single out individual species where pathway 2 applies, such as is proposed for *G. salaris*, it should instead be ideally accomplished through standardized criteria to further distinguish levels of susceptibility which is supported by specific scientific evidence (which is not currently outlined in Chapter 1.5.).	
10.3.6._9_EU	**Category:** Addition **Proposed amended text:** Pathway 2 (historical freedom) is only applicable to make a *self-declaration of freedom from disease* for infection with *G. salaris* for strains of Atlantic salmon (*Salmo salar)* in which it has been demonstrated that G. salaris causes clinical disease. **Rationale:** There are regional strains of Atlantic salmon where the infection is always subclinical. *G. salaris* has never been seen to cause clinical disease in the Baltic, although Baltic strains of Atlantic salmon (*Salmo salar*) are abundant and they are known to be infected with *G.salaris*. Without targeted active monitoring *G. salaris* would never have been identified in countries in the Baltic area as Finland.	See response to comments 10.3._2. and 10.3.5._5.

OR

3) *targeted surveillance*, as described in Chapter 1.4., has been in place in the *zone* for at least the last three years without detection of *G. salaris* and *basic biosecurity conditions* have been continuously met and have been in place for at least two years prior to commencement of *targeted surveillance*;

OR

4) it previously made a self-declaration of freedom for a *zone* from infection with *G. salaris* and subsequently lost its free status due to the detection of *G. salaris* in the *zone* but the following conditions have been met:

   a) on detection of *G. salaris*, the affected area was declared an *infected zone* and a *protection zone* was established; and

   b) infected populations within the *infected zone* have been killed and disposed of by means that minimise the likelihood of further transmission of *G. salaris*, and the appropriate *disinfection* procedures (as described in Chapter 4.4.) have been completed followed by *fallowing* as described in Chapter 4.7.; and

c) previously existing *basic biosecurity conditions* have been reviewed and modified as necessary and have continuously been in place since eradication of infection with *G. salaris*; and

d) *targeted surveillance*, as described in Chapter 1.4., has been in place for:

i) at least the last three years in wild and farmed *susceptible species* without detection of *G. salaris*; or

ii) at least the last one year without detection of *G. salaris* if affected *aquaculture establishments* were not epidemiologically connected to wild populations of *susceptible species*.

In the meantime, a part of the *zone* outside the *infected zone* and *protection zone* may be declared a new *free zone* as described in Article 1.4.4.

[...]

CHAPTER 10.4.

INFECTION WITH INFECTIOUS SALMON ANAEMIA VIRUS

[…]

Reference	Comment	Aquatic Animals Commission Response
9.2._1_EU	**Category:** General **Proposed amended text:** n/a **Rationale:** The EU supports the proposed update to Article 10.4.9 **Supporting evidence:** n/a	Noted.

Article 10.4.9.

Compartment free from infection with ISAV

In this article, all statements referring to a *compartment* free from infection with ISAV are for any detectable ISAV, including HPR0 ISAV.

As described in Article 1.4.4., a Member Country may make a self-declaration of freedom from infection with ISAV for a *compartment* within its *territory* if it can demonstrate that:

1) *targeted surveillance*, as described in Chapter 1.4., has been in place in the *compartment* for at least the last two ~~[under study]~~ years without detection of ISAV, and *basic biosecurity conditions* have been continuously met and have been in place for at least one year prior to commencement of *targeted surveillance*;

OR

2) it previously made a self-declaration of freedom for a *compartment* from infection with ISAV and subsequently lost its free status due to the detection of ISAV in the *compartment* but the following conditions have been met:

   a) all *aquatic animals* within the *compartment* have been killed and disposed of by means that minimise the likelihood of further transmission of ISAV, the appropriate *disinfection* procedures (as described in Chapter 4.4.) have been completed, and the *compartment* has been fallowed as described in Chapter 4.7.; and

   b) previously existing *basic biosecurity conditions*, including the *compartment biosecurity plan*, have been reviewed and modified as necessary and have continuously been in place from the time of restocking with *aquatic animals* from an approved pathogen free source in accordance with the requirements of Articles 10.4.13. and 10.4.14. as appropriate; and

   c) one survey for infection with ISAV has been completed at least six months after restocking (as described in Article 1.4.14.) without detection of the *pathogenic agent*.

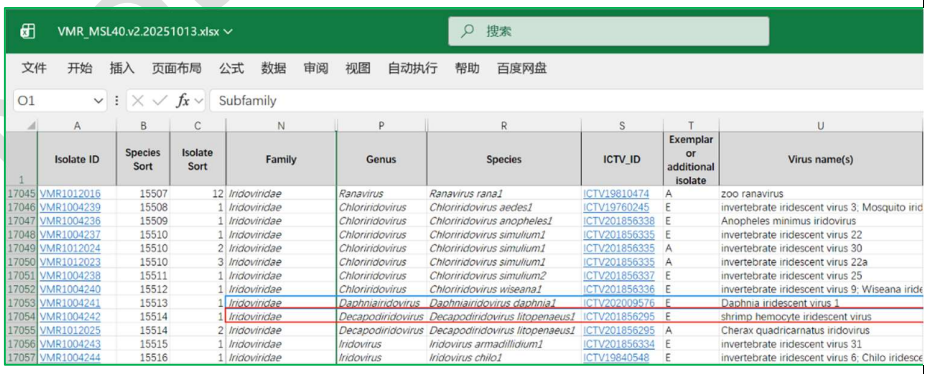
[…]

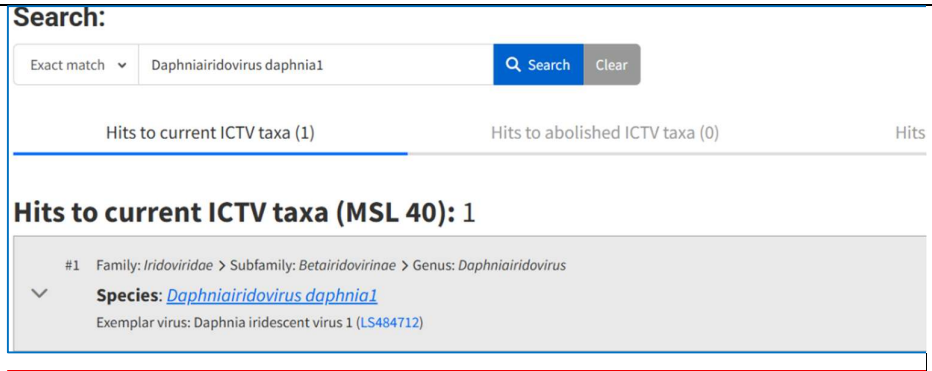
SECTION 9

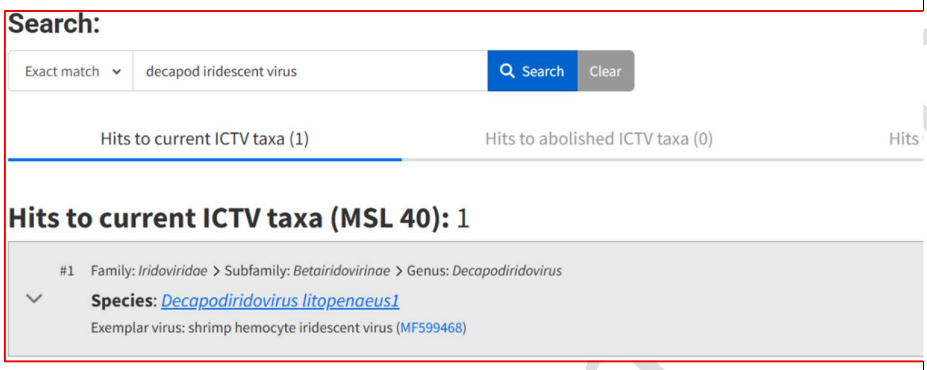
DISEASES OF CRUSTACEANS

Reference	Comment	Aquatic Animals Commission Response
9._1_Australia	**Category:** General **Proposed amended text:** n/a **Rationale**: Australia supports the proposed changes that have been made to viral pathogen names in the text (Article X.X.1. of Chapters 9.3., 9.5., 9.6., 9.8., 9.10., 10.4., 10.5., 10.6., 10.7., 10.9., 10.10., 10.11. and 11.1.) in order to align with ICTV's new binomial nomenclature for virus names. These changes improve the scientific rigour of the Aquatic code and provides clarity regarding of the taxonomy of viral pathogens listed within the code. **Supporting evidence:** n/a	Noted.
9._2_EU	**Category:** General **Proposed amended text:** n/a **Rationale**: The EU supports the revised Articles. However, the EU would kindly request further clarifications about the definition of infection with infectious salmon anaemia virus (ISAV). Please, see specific question below. **Supporting evidence:** n/a	Noted.

CHAPTER 9.3.

9.3._1_China (People's Republic of)	**Category:** general **Proposed amended text:** n/a **Rationale**: We agree with WOAH here to change decapod iridescent virus 1 (DIV1) to *Decapodiridovirus litopenaeus1*. However, it is inappropriate to use decapod iridescent virus 1 (DIV1) as the chapter title and the name of the virus in the *Aquatic Code* and *Aquatic Manual*. We suggest changing the chapter title from “infection with decapod iridescent virus 1” to “infection with *Decapodiridovirus litopenaeus1*” or “infection with shrimp hemocyte iridescent virus”. The virus name “decapod iridescent virus 1” and its abbreviation DIV1 should be discarded, and the virus name should be changed to *Decapodiridovirus litopenaeus1* or shrimp hemocyte iridescent virus (SHIV).Reasons for revision are as follows: (1) In 2023, ICTV changed the scientific name of this virus from *Decapod iridescent virus 1* to *Decapodiridovirus litopenaeus1*. The scientific name *Decapod iridescent virus 1* has been discontinued by ICTV and is no longer in use. (2) The abbreviation DIV1 has been officially used by ICTV for *Daphnia iridescent virus 1* (the exemplar isolate of species *Daphniairidovirus daphnia1*), and ICTV has never abbreviated decapod iridescent virus 1 as DIV1. Abbreviating Decapod iridescent virus 1 (a name that has been discarded by ICTV) as DIV1 can cause confusion. Therefore, DIV1 should no longer be used to refer to *Decapodiridovirus litopenaeus1*. (3) The exemplar isolate of species *Decapodiridovirus litopenaeus1* is “shrimp hemocyte iridescent virus (SHIV)” according to ICTV.  VMR_MSL40.v2.20251013.xlsx 搜索 文件 开始 插入 页面布局 公式 数据 审阅 视图 自动执行 帮助 百度网盘 O1 Subfamily A B C N P R S T U 1 Isolate ID | Species Sort | Isolate Sort | Family | Genus | Species | ICTV_ID | Exemplar or additional isolate | Virus name(s) 17045 VMR1012016 15507 12 Iridoviridae Ranavirus Ranavirus rana1 ICTV19810474 A zoo ranavirus 17046 VMR1004239 15508 1 Iridoviridae Chloriridovirus Chloriridovirus aedes1 ICTV19760245 E invertebrate iridescent virus 3; Mosquito irid 17047 VMR1004236 15509 1 Iridoviridae Chloriridovirus Chloriridovirus anopheles1 ICTV201856338 E Anopheles minimus iridovirus 17048 VMR1004237 15510 1 Iridoviridae Chloriridovirus Chloriridovirus simulium1 ICTV201856335 E invertebrate iridescent virus 22 17049 VMR1012024 15510 2 Iridoviridae Chloriridovirus Chloriridovirus simulium1 ICTV201856335 A invertebrate iridescent virus 30 17050 VMR1012023 15510 3 Iridoviridae Chloriridovirus Chloriridovirus simulium1 ICTV201856335 A invertebrate iridescent virus 22a 17051 VMR1004238 15511 1 Iridoviridae Chloriridovirus Chloriridovirus simulium2 ICTV201856337 E invertebrate iridescent virus 25 17052 VMR1004240 15512 1 Iridoviridae Chloriridovirus Chloriridovirus wiseana1 ICTV201856336 E invertebrate iridescent virus 9; Wiseana iride 17053 VMR1004241 15513 1 Iridoviridae Daphniairidovirus Daphniairidovirus daphnia1 ICTV202009576 E Daphnia iridescent virus 1 17054 VMR1004242 15514 1 Iridoviridae Decapodiridovirus Decapodiridovirus litopenaeus1 ICTV201856295 E shrimp hemocyte iridescent virus 17055 VMR1012025 15514 2 Iridoviridae Decapodiridovirus Decapodiridovirus litopenaeus1 ICTV201856295 A Cherax quadricarinatus iridovirus 17056 VMR1004243 15515 1 Iridoviridae Iridovirus Iridovirus armadillidium1 ICTV201856334 E invertebrate iridescent virus 31 17057 VMR1004244 15516 1 Iridoviridae Iridovirus Iridovirus chilo1 ICTV19840548 E invertebrate iridescent virus 6; Chilo iridesc (from ICTV database file 2025: VMR_MSL40.v2.20251013.xlsx) See also: ICTV Database Search Results	Did not agree. The name of the disease is retained to ensure stability in disease names and alignment with common use. This includes use in Chapter 1.3. ‘Diseases listed by WOAH’ and the disease-specific chapters in the *Aquatic Code* and *Aquatic Manual*. The pathogenic agent name has been updated to reflect the recent official nomenclature.





supporting evidence: not relevant

INFECTION WITH DECAPOD IRIDESCENT VIRUS 1

Article 9.3.1.

For the purposes of the *Aquatic Code*, infection with decapod iridescent virus 1 (DIV1) means *infection* with the *pathogenic agent* *Decapodiridovirus litopenaeus1*~~*Decapod iridescent virus 1* (DIV1)~~, of the ~~*Genus Decapodiridovirus*~~ and the Family *Iridoviridae*.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

9.3.1._1_Thailand	**Category:** Editorial **Proposed amended text:** "For the purposes of the *Aquatic Code*, infection with decapod iridescent virus 1 (DIV1) means infection with the *pathogenic agent* *Decapodiridovirus litopenaeus1*~~Decapod iridescent virus 1 (DIV1)~~, of the ~~Genus *Decapodiridovirus* and the~~ Family *Iridoviridae*." **Rationale:** correction **Supporting evidence:** -	Agreed to editorial change.

CHAPTER 9.5.

INFECTION WITH HYPODERMAL AND HAEMATOPOIETIC NECROSIS VIRUS

9.5._1_Canada	**Comment Category:** Addition **Proposed amended text:** INFECTION WITH INFECTIOUS HYPODERMAL AND HAEMATOPOIETIC NECROSIS VIRUS **Rationale:** Editorial to align the title of the listed disease	Agreed to editorial change.

[…]

Article 9.5.1.

For the purposes of the *Aquatic Code*, infection with infectious hypodermal and haematopoietic necrosis virus (IHHNV) means *infection* with the *pathogenic agent* *Penstylhamaparvovirus decapod1*~~*Decapod penstylhamaparvovirus 1*~~, of the ~~Genus *Penstylhamaparvovirus*~~ and Family *Parvoviridae*.

9.5.1._1_Thailand	**Category:** Editorial **Proposed amended text:** "For the purposes of the *Aquatic Code*, infection with infectious hypodermal and haematopoietic necrosis virus (IHHNV) means infection with the *pathogenic agent* *Penstylhamaparvovirus decapod1*~~*Decapod penstylhamaparvovirus 1*~~, of the ~~Genus *Penstylhamaparvovirus*~~ ~~and~~ Family *Parvoviridae*." **Rationale:** correction **Supporting evidence:** -	Agreed to editorial change.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

CHAPTER 9.6.

INFECTION WITH INFECTIOUS MYONECROSIS VIRUS

Article 9.6.1.

For the purposes of the *Aquatic Code*, infection with infectious myonecrosis virus (IMNV) means *infection* with the *pathogenic agent* infectious myonecrosis virus ~~(IMNV)~~ of the Genus *Artivirus* and of the Family Artiviridae ~~Totiviridae (tentative classification)~~.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

9.6.1._1_Canada	**Comment Category:** Change **Proposed amended text:** For the purposes of the *Aquatic Code*, infection with infectious myonecrosis virus (IMNV) means *infection* with the *pathogenic agent* *Artivirus ichi* ~~infectious myonecrosis virus (IMNV)~~ of the Genus *Artivirus* and of the Family Artiviridae ~~Totiviridae (tentative classification)~~. **Rationale:** A search for IMNv (Penaeid shrimp infectious myonecrosis virus) within ICTV returns the virus species *Artivirus ichi.* The ICTV report in family Artiviridea (Family: Artiviridae (Interim Report) | ICTV) lists Penaeid shrimp infectious myonecrosis virus as *Artivirus ichi*	Agreed to update name based on recent nomenclature.
9.6.1._2_China (People's Republic of)	**Category:** change **Proposed amended text :** For the purposes of the *Aquatic Code*, infection with infectious myonecrosis virus (IMNV) means *infection* with the *pathogenic agent* ~~infectious myonecrosis virus (IMNV) of the~~ Genus *Artivirus* and *Artivirus ichi* of the Family Artiviridae ~~Totiviridae (tentative classification)~~. **Rationale:** In 2023, ICTV updated the classification of IMNV from an unassigned genus in the family Totiviridae to a member of the genus *Artivirus* in the family Artiviridae. The scientific name of IMNV is "*Artivirus ichi*", and the exemplar isolate of species *Artivirus ichi* is Penaeid shrimp infectious myonecrosis virus (PSIMNV, AY570982). Here we recommend to use the scientific name of this virus *Artivirus ichi*. **supporting evidence:** not relevant	Agreed to update name based on recent nomenclature.

CHAPTER 9.8.

INFECTION WITH TAURA SYNDROME VIRUS

Article 9.8.1.

For the purposes of the *Aquatic Code*, infection with Taura syndrome virus (TSV) means *infection* with the *pathogenic agent* *Aparavirus tauraense*~~Taura syndrome virus (TSV)~~, of the ~~Genus *Aparavirus*,~~ Family Dicistroviridae~~ and Order Picornavirales~~.

Information on methods for *diagnosis* are provided in the *Aquatic Manual*.

[...]

CHAPTER 9.10.

INFECTION WITH YELLOW HEAD VIRUS GENOTYPE 1

Article 9.10.1.

For the purposes of the *Aquatic Code*, infection with yellow head virus genotype 1 means *infection* with the *pathogenic agent* yellow head virus genotype 1 (YHV1), of the Genus *Okavirus*~~,~~ and Family Roniviridae, ~~Order Nidovirales~~.

9.10.1._1 _Australia	**Category:** Addition **Proposed amended text:** For the purposes of the *Aquatic Code*, infection with yellow head virus genotype 1 (YHV1) means *infection* with the *pathogenic agent* ~~yellow head virus genotype 1 (YHV1)~~ *Okavirus flavicapitis*, of the Genus *Okavirus*, and Family Roniviridae, ~~Order Nidovirales~~. **Rationale**: Yellow head virus genotype 1 is listed by the ICTV as *Okavirus flavicapitis*, with email ratification February 2025 (MSL #40) **Supporting evidence:** ICTV listing (https://ictv.global/taxonomy/taxondetails?taxnode_id=202406183&taxon_name=Okavirus%20flavicapitis)	Agreed to update name based on recent nomenclature.
9.10.1._2 _Canada	**Comment Category:** Change **Proposed amended text:** For the purposes of the *Aquatic Code*, infection with yellow head virus genotype 1 means *infection* with the *pathogenic agent* *Okavirus flavicapitis* ~~yellow head virus genotype 1 (YHV1)~~, of the Genus *Okavirus*~~,~~ and Family Roniviridae, ~~Order Nidovirales~~. **Rationale:** We note that the pathogenic agent for Yellow head virus was updated in 2022. According to ICTV Yellow head disease is *Okavirus flavicapitis* Taxon Details | ICTV	Agreed to update name based on recent nomenclature.
9.10.1._3 _China (People's Republic of)	**Category:** change **Proposed amended text :** For the purposes of the Aquatic Code, infection with yellow head virus genotype 1 means infection with the pathogenic agent ~~yellow head virus genotype 1 (YHV1)~~*Okavirus flavicapitis*, of the Genus Okavirus~~,~~ and Family Roniviridae, ~~Order Nidovirales~~. **Rationale:** According to the ICTV 2025 report, the scientific name of this pathogen should be *Okavirus flavicapitis.*	Agreed

	supporting evidence:  	

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

SECTION 10

DISEASES OF FISH

CHAPTER 10.4.

INFECTION WITH INFECTIOUS SALMON ANAEMIA VIRUS

10.4._1_ EU	**Category:** General **Proposed amended text:** n/a **Rationale**: The EU in general supports this revised Article. However, we would seek clarification as regards the reasons to remove the "genus isavirus" as it is still in the ICTV description **Supporting evidence:** https://ictv.global/taxonomy/taxondetails?taxnode_id=202403964&taxon_name=Isavirus%20salaris	The reference to 'genus *Isavirus*' was removed because the genus is already included in the binomial species name. Therefore, the Commission considered its repetition unnecessary. The species name, as adopted by ICTV, provides the required taxonomic clarity.

Article 10.4.1.

For the purposes of the *Aquatic Code*, infection with infectious salmon anaemia virus (ISAV) means *infection* with the *pathogenic agent Isavirus salaris* of the Family *Orthomyxoviridae*. Infection with ISAV includes two genotypes; highly polymorphic region (HPR)-deleted ~~infectious salmon anaemia virus (~~ISAV~~)~~, or the non-pathogenic HPR0 ISAV(non-deleted highly polymorphic region ISAV) ~~ISAV, of the Genus *Isavirus* and Family *Orthomyxoviridae*~~. Both genotypes should be notified in accordance with Chapter 1.1.

10.4.1._1_Canada	**Comment Category:** Addition **Proposed amended text:** For the purposes of the *Aquatic Code*, infection with infectious salmon anaemia virus (ISAV) means *infection* with the *pathogenic agent Isavirus salaris* of the Family *Orthomyxoviridae*. Infection with ISAV includes two genotypes; highly polymorphic region (HPR)-deleted ~~infectious salmon anaemia virus (~~ISAV~~)~~ (HPR-deleted ISAV), or ~~the non-pathogenic HPR0~~ full-length highly polymorphic region-0 ISAV (~~non-deleted highly polymorphic region~~ HPR0 ISAV) ~~ISAV, of the Genus *Isavirus* and Family *Orthomyxoviridae*~~. Both genotypes should be notified in accordance with Chapter 1.1. **Rationale:** For consistency and clarity, the code should state the name in full then provide the acronym (in brackets) for both cases. • highly polymorphic region-deleted ISAV (HPR-deleted ISAV),	Agreed to editorial change for clarity.

	• full-length highly polymorphic region-0 ISAV (HPR0 ISAV) Additionally, non-pathogenic is a qualifier, not part of the name of the strain and therefore should not be included within the name of the strain within the first paragraph. It is, and is more appropriately, included within the second paragraph describing the link between HPR0 and HPR-deleted ISA).	

There is a link between non-pathogenic HPR0 ISAV and pathogenic HPR-deleted ISAV, with some *outbreaks* potentially occurring as a result of the emergence of HPR-deleted from HPR0.

The provisions in this chapter are provided in recognition of three possible levels of disease status with respect to ISAV:

1) HPR0 ISAV and HPR-deleted ISAV free;

2) HPR0 ISAV endemic (but HPR-deleted ISAV free);

3) HPR0 ISAV and HPR-deleted ISAV endemic.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

CHAPTER 10.5.

INFECTION WITH INFECTIOUS SALMONID ALPHAVIRUS

10.5.1._1_Australia	**Category:** editorial **Proposed amended text:** CHAPTER 10.5. INFECTION WITH ~~INFECTIOUS~~ SALMONID ALPHAVIRUS **Rationale**: Chapter 10.5 title 'infection with infectious salmonid alphavirus' differs to the name of the pathogen within the chapter: that is, 'infection with salmonid alphavirus (SAV)'. To be consistent with other chapters, where the title reflects the pathogen name within the chapter, it is proposed the name of the chapter be changed to 'infection with salmonid alphavirus', (SAV). This would also avoid any confusion with the acronym for infectious salmon anaemia virus (ISAV). *note the Australia's National List of Reportable Diseases of Aquatic Animals - DAFF has 'infection with salmonid alphavirus' **Supporting evidence:** n/a	Agreed to editorial change.

Article 10.5.1.

For the purposes of the *Aquatic Code*, infection with salmonid alphavirus (SAV) means *infection* with any genotype of the *pathogenic agent* *Alphavirus salmon*~~salmonid alphavirus (SAV)~~, of the ~~Genus *Alphavirus* and~~ Family *Togaviridae*.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[...]

CHAPTER 10.6.

INFECTION WITH INFECTIOUS HAEMATOPOIETIC NECROSIS VIRUS

Article 10.6.1.

For the purposes of the *Aquatic Code*, infection with infectious haematopoietic necrosis virus (IHNV) means *infection* with the *pathogenic agent* *Novirhabdovirus salmonid* ~~Salmonid novirhabdovirus (commonly known as infectious haematopoietic necrosis virus [IHNV])~~ of the ~~Genus *Novirhabdovirus* and~~ Family *Rhabdoviridae*.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

CHAPTER 10.7.

INFECTION WITH KOI HERPESVIRUS

Article 10.7.1.

For the purposes of the *Aquatic Code*, infection with koi herpesvirus (KHV) means *infection* with the *pathogenic agent* *Cyvirus cyprinidallo 3*~~koi herpesvirus (KHV)~~ of the ~~Genus *Cyprinivirus* and~~ Family *Alloherpesviridae*.

10.7.1._1_Canada	**Comment Category:** editorial **Proposed amended text:** For the purposes of the *Aquatic Code*, infection with koi herpesvirus (KHV) means *infection* with the *pathogenic agent* *Cyvirus cyprinidallo3* ~~*cyprinidallo 3*~~~~koi herpesvirus (KHV)~~ of the ~~Genus *Cyprinivirus* and~~ Family *Alloherpesviridae*. **Rationale:** R*emove space before 3 to be consistent with ICTV*	Agreed to editorial change.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

CHAPTER 10.9.

INFECTION WITH SPRING VIRAEMIA OF CARP VIRUS

Article 10.9.1.

For the purposes of the *Aquatic Code*, infection with spring viraemia of carp virus (SVCV) means *infection* with the *pathogenic agent* *Sprivivirus cyprinus* ~~spring viraemia of carp virus (SVCV)~~ of the ~~Genus *Sprivivirus* and~~ Family *Rhabdoviridae*.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[...]

CHAPTER 10.10.

INFECTION WITH VIRAL HAEMORRHAGIC SEPTICAEMIA VIRUS

Article 10.10.1.

For the purposes of the *Aquatic Code*, infection with viral haemorrhagic septicaemia virus (VHSV) means *infection* with the *pathogenic agent* *Novirhabdovirus piscine* ~~viral haemorrhagic septicaemia virus (VHSV)~~, of the ~~Genus *Novirhabdovirus*~~ and Family *Rhabdoviridae*.

10.10.1._1_Thailand	**Category:** Editorial **Proposed amended text:** "For the purposes of the *Aquatic Code*, infection with viral haemorrhagic septicaemia virus (VHSV) means infection with the *pathogenic agent* *Novirhabdovirus piscine* ~~viral haemorrhagic septicaemia virus (VHSV)~~, of the ~~Genus Novirhabdovirus~~ ~~and~~ Family *Rhabdoviridae*." **Rationale:** correction **Supporting evidence:** -	Agreed to editorial change.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

CHAPTER 10.11.

INFECTION WITH TILAPIA LAKE VIRUS

Article 10.11.1.

For the purposes of the *Aquatic Code*, infection with tilapia lake virus (TiLV) means *infection* with the *pathogenic agent* *Tilapinevirus tilapiae*~~*Tilapia tilapinevirus*~~, of the ~~Genus *Tilapinevirus* and the~~ Family *Amnoonviridae*.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

SECTION 11

DISEASES OF MOLLUSCS

CHAPTER 11.1.

INFECTION WITH ABALONE HERPESVIRUS

Article 11.1.1.

For the purposes of the *Aquatic Code*, infection with abalone herpesvirus (AbHV) means *infection* with the *pathogenic agent* *Aurivirus haliotidmalaco 1*~~Haliotid herpesvirus 1 (HaHV-1)~~, of the ~~Genus *Aurivirus* and~~ Family *Malacoherpesviridae*.

11.1.1._1_Canada	**Comment Category:** Editorial **Proposed amended text:** For the purposes of the *Aquatic Code*, infection with abalone herpesvirus (AbHV) means *infection* with the *pathogenic agent* *Aurivirus haliotidmalaco1 haliotidmalaco 1*~~Haliotid herpesvirus 1 (HaHV-1)~~, of the ~~Genus *Aurivirus* and~~ Family *Malacoherpesviridae*. **Rationale:** R*emove space before 1 to be consistent with ICTV and to align with how it is shown in Annex 20 for the Aquatic Manual.*	Agreed to editorial change.

Information on methods for *diagnosis* is provided in the *Aquatic Manual*.

[…]

SECTION 4.

DISEASE PREVENTION AND CONTROL

CHAPTER 4.3.

APPLICATION OF COMPARTMENTALISATION

Reference	Comment	Aquatic Animals Commission Response
4.3._1_Australia	**Category:** general comment **Proposed amended text:** n/a **Rationale**: Chapter 4.3 Application of compartmentalisation has been improved to provide greater clarity (e.g. risk analysis points are comprehensive) while reducing superfluous information to ensure conciseness. The chapter provides a good explanation of dependent vs independent compartments and a good description of internal and external surveillance. **Supporting evidence:** n/a	Noted.
4.3._2_Canada	**Comment Category:** General **Proposed amended text:** n/a **Rationale**: Canada recognises the work of the Commission in the significant revision completed and thanks the Commission for taking its comments into account. Canada looks forward to seeing the result of the review of Chapter 1.4. in the February 2026 meeting report and how the changes proposed within the revised Chapter 4.3. will be taken into account. Specifically, we are interested to see the changes that will be proposed for both Chapter 1.4. and the disease specific chapters to allow for self-declaration of freedom for dependent compartments using passive surveillance. Once this has been clarified, Members will be able to understand the impacts of the revisions to this chapter and the proposed changes to the disease specific chapters to incorporate provisions for self-declaration of dependent compartments. **Supporting evidence:** n/a	The Commission reviewed Chapter 1.4.'Aquatic animal disease surveillance' accounting for proposed revisions to Chapter 4.3. Information on the review and proposed revision can be found in Item 6.2. and Annex 12.

4.3._3_Canada	**Comment Category: General** **Proposed amended text:** n/a **Rationale**: We note the response of the commission to a Member's previous comment which indicated that independent compartments are limited to closed compartments and dependent compartments are limited to semi-closed. However, due to the limited knowledge of inactivation of aquatic pathogens within influent water and different requirements for inactivation between pathogens this may pose very limiting for Members. For example, in a salmonid compartment the levels of UV treatment for influent waters might differ between pathogenic agents. The revised Chapter 4.3. isn't limited to WOAH listed diseases but rather specific diseases based on the needs of Members. For example, while Infectious Pancreatic Necrosis is not a listed disease, many countries require freedom. IPN requires much higher levels of UV for inactivation in influent waters. This level of UV treatment maybe unattainable but the level required for other diseases to which the species may be susceptible (IHN or ISA etc) would be achievable. In this situation, it is unclear why the same compartment cannot be independent for IHN or ISA based on complete epi separation from the environment and dependent compartment based on the health status for surrounding waters. **Supporting evidence:** n/a	Revised Chapter 4.3. does not preclude the application of compartmentalisation as described in this comment. It is the responsibility of the Competent Authority to complete a risk analysis on the proposed situation. If the risk analysis supports this application, the Competent Authority can provide justification for approval of the compartment to trading partners through bilateral negotiations.
4.3._4_Canada	**Comment Category: General** **Proposed amended text:** n/a **Rationale:** We note that inclusion of 'approving third parties within the AAHS' is included in several locations within this revised chapter. Our review of the Aquatic and Terrestrial Codes showed a lack of guidance with respect to requirements for approval of third party auditors. Perhaps this could be included at a higher level within the Aquatic Code rather than incorporate within this chapter. **Supporting evidence:** n/a	Agreed that the current *Aquatic Code* does not provide guidance on the approval of third parties within the Aquatic Animal Health Services. The Commission will include this in the work programme for consideration in Chapter 3.1. when it is revised. The Commission added text to Article 4.3.10. 'Competent Authority responsibilities' to emphasise that third party auditors should be independent from management and ownership structures of the compartment for clarity and consistency with wording in Article 4.3.7. 'Laboratory testing'.

4.3._5_China (People's Republic of)	**Category:** general **Proposed amended text:** n/a **Rationale:** China commends the Commission's efforts in revising the chapter on the application of compartmentalisation. The new draft is better structured, and we principally support the introduction of "independent" and "dependent" compartment categories, which offers greater flexibility to accommodate diverse aquaculture systems. However, the draft contains ambiguities regarding certain key concepts and requirements, which could lead to divergent interpretations and applications among Members and cause unnecessary trade disputes. Our main concerns are as follows: 1) **Risk analysis for different compartments needs to be more detailed considerations:** Compartmentalization has been expanded from the original "closed aquaculture" mode to include both "closed aquaculture" and "semi-closed aquaculture" modes. The concept of risk analysis has been added and emphasized for the application of compartmentalization. Risk analysis should consider the category of the compartment, the specific factors of a related disease, and the environmental factors that may affect the spread of the disease, in order to determine appropriate risk management measures. 2) **The definition and risk management measures for "dependent compartments" are not sufficiently clear:** Descriptions for dependent compartments, such as "may not provide sufficient risk mitigation", weaken the credibility of compartmentalisation as a tool for international trade. It must be explicit that any officially approved compartment, regardless of its type, must provide adequate and reliable risk assurance for its **stated trade purpose**. 3) **The core role of the Competent Authority needs further strengthening:** While the draft mentions the Competent Authority multiple times, its authority in approval and oversight is insufficiently stated in some key provisions. The entire lifecycle of a compartment—including its establishment, approval, supervision, auditing, suspension, and revocation—must be under the absolute management of the Competent Authority. There should be no ambiguity on this point. Moreover, the possibility of an independent compartment to achieve disease-free status heavily depends on the transmission of risks from the surrounding environment. Therefore, additional biosecurity measures and disease surveillance need to be taken to mitigate the introduction of disease risks in the environment. However, the biosecurity	Point 1 – For an independent compartment risk is addressed through the biosecurity plan as described in Chapter 4.1. 'Biosecurity for Aquaculture Establishments'. Article 4.1.8. of this chapter describes the use of risk analysis to evaluate biosecurity threats and develop mitigation measures. Edits were made to Article 4.3.5. to emphasise this use of risk analysis regarding the biosecurity plan for independent compartments. Point 2- Agreed that both independent and dependent compartments need to provide appropriate risk management for its trade purpose. Comments addressed through the text clarify this point in the chapter. Point 3 – Regarding the steps of the lifecycle of the compartment (establishment, approval, supervision, auditing, suspension and revocation) these must be under the authority of the Competent Authority however they may be completed by third parties that are a part of the Aquatic Animal Health Services. Text was added to Article 4.3.10. to clarify that where third parties are used they must be separate from the management and ownership structures of the compartments. Agreed on the importance of the role of the Competent Authority. Noted that this is emphasised in Article 4.3.10. and was added to the definition of compartment (see Annex 14).

	measures and disease surveillance adopted in this part cannot be independently completed by the relevant establishment and require coordination and organization from the Competent Authority. The important responsibility of the Competent Authority is suggested to be reflected in this chapter. 4) **Terminology consistency needs improvement:** The terms “risk management” and “risk mitigation” are used interchangeably. We recommend unifying and standardizing these terms to ensure precision. 5) **Enhance the authenticity verification of record documents:** It is suggested adding measures to prevent record files from being modified, replaced, and forged in Article 4.3.9. 6) **More logic narrative sequence:** It’s suggested to place notification to the importing country in the second paragraph in Article 4.3.12. The second paragraph in the original text confirms the outbreak of a disease in the compartment suspending the epidemic free status. The notification to the importing country addressed in the first paragraph of the original text should follow the confirmation of the disease in the compartment. As addressed in Chapter 1.1, the diseases reported should officially be reported only after the confirmation of the disease. We suggest that the Commission clarifies and refines the aforementioned issues before the adoption of this chapter to ensure its scientific soundness, practicality, and authority. **Supporting evidence:** not relevant	Point 4 – Risk management is a defined term in the Glossary which includes taking risk mitigation measures. The chapter was reviewed to ensure that risk management is the term used throughout. Point 5 – In Article 4.3.9. it states that ‘Records should be maintained consistently by the operator of the free compartment and be accessible…’. It is implicit that they should not be modified, replaced or forged. Point 6 – Agreed, text revised in second paragraph of Article 4.3.12. to include notification to importing countries.
4.3._6_Norway	**Category**: general **Proposed amended text**: Not relevant **Rationale:** Norway would like to thank the Commission for addressing many of our previous concerns in this revised version of the chapter. However, we would like to reiterate our previous comment about automatically excluding semi-open production systems from eligibility to become dependent compartments. Please see our comment under Article 4.3.4. Some additional comments are also provided below. **Supporting evidence, if relevant:** not relevant	See report item 6.3. – Commission requests Member feedback.

4.3._7_UK	**Category:** General **Proposed amended text:** n/a **Rationale**: We thank the Commission for their work on this Chapter, including the extensive revisions that have greatly aided clarity. We do, however, continue to have questions related to the requirements for establishing and maintaining dependent compartments, which will be reflected in our feedback embedded below. Overall, we feel that there are likely to only be very limited, niche circumstances where such compartments would be possible and even in such cases, there is no guarantee that partners would be agreeable to establishing trade with them given their limitations on preventing pathogen movement from the surrounding environment. Furthermore, creating two types of compartments would create a divergence between the aquatic and terrestrial Codes, which goes against recent and ongoing work to bring them into greater alignment. **Supporting evidence:** not relevant	Revised Chapter 4.3. provides a framework for Members to apply compartmentalisation and negotiate requirements with trading partners. It is not intended to provide prescriptive requirements and instead utilises risk analysis to inform necessary requirements to mitigate risks. Text was added to Article 4.3.1. to reinforce the purpose of the revised Chapter 4.3. The concept that the disease status of a compartment or zone is dependent on the surrounding environment is not new within the standards for either terrestrial or aquatics. The basis of this is included in the *Terrestrial Code* where freedom may be required in a protection zone around compartments (e.g. Article 8.8.6. 'Compartment free from FMD where vaccination is not practiced' that states the compartment should only approved when there has been no infection or transmission of FMDV within a 10 km radius of the compartment for three months). The concept of a dependent compartment is also not excluded from the current chapter on compartmentalisation in the *Aquatic Code* although the concept may not be explicitly described. The revised draft Chapter 4.3. explicitly includes the concept of dependent compartments to provide clarity on a concept that is already utilised by some members and recognised for international trade.

4.3._8_United States	**Category:** general We appreciate the efforts of the WOAH Aquatic Commission in developing guidelines that promote the safe trade of aquatic animals and their products. The Commission's commitment to soliciting Member feedback and aligning standards with international trade principles is commendable and critical for global aquatic animal health and commerce. However, we strongly disagree with the proposed inclusion of the "dependent" type of compartment. This approach does not meet the internationally recognized definition or purpose of a compartment as outlined in the recent WOAH reports: "Use, challenges and impact of zoning and compartmentalization," Part 1 (2024) and Part 2 (2025), which emphasize that compartments are intended to: • **Facilitate Trade During Disease Outbreaks** By defining subpopulations based on biosecurity and management practices, compartments enable trade to continue even when a country or zone is affected by disease. • **Support Disease Management and Control** Compartments provide functional separation to isolate and protect animal populations, reducing the risk of disease spread. • **Provide Flexibility Beyond Geography** Unlike zoning, compartments rely on management systems and biosecurity protocols rather than geographic boundaries, making them vital in resource-limited or densely populated areas. • **Align with International Standards and Trade Agreements** Compartments must be recognized by Veterinary Authorities and comply with WOAH standards, providing a science-based framework that promotes transparency and trust among trade partners. • **Apply a Risk-Based Approach** Compartments extend risk boundaries beyond geography, incorporating epidemiological factors to support targeted surveillance and certification. The proposed "dependent compartment," does not achieve these objectives because it inappropriately incorporates core principles of regionalization/ zoning into a compartmentalization approach; it relies solely on the health status of the surrounding zone or region, and fails to provide functional separation or independent biosecurity measures, and therefore offers no trade continuity protection during a disease outbreak. This subverts the intent of compartmentalization as a risk-based, science-driven tool for safe trade. Furthermore, as noted in Annex 9, Item 6.4 also up for Member comment, the Commission's stated objectives for revising Section 5 include improving usability for	The concept of dependent compartments is not currently excluded in either of the *Aquatic Code* or *Terrestrial Code*. Although not called a dependent compartment within the FMDV chapter in the *Terrestrial Code* it contains a compartment that is dependent on the status of the external environment (See response to comment 4.3._7.). The revisions to Chapter 4.3. and inclusion of dependent compartments aims to provide clarity on a compartment already being used by Member Countries, and to provide more opportunity to facilitate trade. The Commission noted that within a compartment there is the need to show freedom from disease and that even for a dependent compartment it does not rely only on the health status of the zone or region around it. A dependent compartment is hydrologically linked to the environment and thus risk assessment is used to understand the risk and the risk management measures that need to be taken to ensure the health status of the compartment. The text was reviewed to clarify this point throughout.

	trade measures and addressing aquatic-specific trade issues while maintaining alignment with the Terrestrial Code. The dependent compartment concept does not meet these goals and risks creating confusion rather than clarity. In conclusion, we strongly recommend that the Commission removes dependent compartments from the WOAH Aquatic Code to maintain adherence to the established principles of compartmentalization that support disease control, trade resilience, and international standards, and which are already being successfully implemented by Competent Authorities for other WOAH-listed pathogens.	
4.3._9_EU	**Category**: general **Proposed amended text:** n/a **Rationale**: The EU in general supports this new chapter. The EU welcomes the confirmation that Chapter 1.4 on "Aquatic disease surveillance" will start to be reviewed in February 2026 particularly to address the surveillance in dependent compartments. Moreover, the EU considers that the higher risk of introducing a disease from the surrounding waters associated with dependent compartments must be taken into account when setting out the period of time required to gain disease-freedom, in the disease-specific chapters. As regards Article 4.3.4, the draft excludes from the concept of dependent compartments semi-open systems as net pens or cages for finfish and suspended baskets or rope systems for molluscs in natural water bodies. The EU considers that those production systems can be considered as compartments in certain circumstances. Please see a specific comment to this draft Article below. As regards Article 4.3.12 on "Notification and response measures" the EU considers that the reference to notification of suspicions to importing countries is not covered in Chapter 1.1. as it is currently proposed in this draft chapter. Please see a specific comment to this draft Article below. **Supporting evidence:** not relevant	The Commission reviewed Chapter 1.4.'Aquatic animal disease surveillance' accounting for proposed revisions to Chapter 4.3. Information on the review and proposed revision can be found in Item 6.2. and Annex 12. Regarding semi-open systems - See report item 6.3. – Commission requests Member feedback.

Article 4.3.1.

4.3.1._1_Australia	**Category:** general comment **Proposed amended text:** n/a **Rationale**: Australia suggests clarifying that a group of establishments for the purpose of a compartment must all be under the same Competent Authority. **Supporting evidence:** n/a	Did not agree that a group of establishments comprising a compartment must be under the same Competent Authority. Depending on administrative arrangements within a country there may be multiple Competent Authorities in association with the Veterinary Authority, that share responsibility for oversight of establishments that comprise the compartment.

Objective and introduction

This chapter provides recommendations for establishing and maintaining *compartments* that are free from specified *diseases* for the purpose of facilitating trade and ~~or~~ for *disease* prevention and control.

4.3.1._2_Chile	**Categoría:** Cambio **Texto modificado propuesto:** Este capítulo brinda recomendaciones para establecer y mantener *compartimentos* libres de *enfermedades* específicas, con la finalidad de facilitar el comercio ~~o~~ y la prevención y el control de *enfermedades*. **Justificación:** Este comentario fue aportado en el anexo 19 en consulta enviado en julio del 2025, y de acuerdo con la respuesta de la comisión en anexo 3 señala que este comentario fue aceptado, sin embargo, no se ve reflejado en anexo 6 en consulta. La justificación de este cambio se sustenta en que el fin del compartimento es la prevención y el control de enfermedades, que por consecuencia facilita el comercio. **Evidencia de apoyo:** No aplica	Agreed, translation revision to be made to Spanish version only.

Compartmentalisation provides a means of demonstrating that an *aquaculture establishment* is free from one or more specified *diseases* by establishing and maintaining functional epidemiological separation between the *aquatic animals* within the *compartment* and sources of *infection* outside the *compartment*. A *compartment* may comprise a single *aquaculture establishment* or a group of ~~interrelated~~ *aquaculture establishments* that operate under a common set of *risk management* measures in accordance with this chapter.

4.3.1._3_Australia	**Category:** addition **Proposed amended text:** A compartment may comprise a single aquaculture establishment or a group of ~~interrelated~~ aquaculture establishments that operate under the same Competent Authority and under a common set of risk management measures in accordance with this chapter. **Rationale**: The proposed added text provides clarity around the group of aquaculture establishments being within the same jurisdiction operating under the one competent authority. This is in line with article 4.3.10 in that the Competent Authority in one jurisdiction would not have authority over establishments in another jurisdiction. It also aligns with other text referring to the Competent Authority throughout Chapter 4.3. The need for this distinction arises from historical industry enquiries seeking this clarification in relation to their business having numerous farms across different jurisdictions and competent authorities. **Supporting evidence:** n/a	Did not agree, see response to comment 4.3.1._1.

Compartmentalisation provides an opportunity for the operator ~~private sector~~ to demonstrate *disease* freedom at the enterprise level, including in circumstances where alternatives such as *country* or *zone* freedom may not be feasible or cost-effective. Investment by the operator~~private sector~~ and oversight by the relevant *Competent Authorities* are~~is~~ essential.

4.3.1._4_Australia	**Category:** editorial **Proposed amended text:** Investment by the operator ~~private sector~~ and oversight by the relevant Competent Authority~~ies~~ are~~is~~ is essential **Rationale**: In line with the suggestion above and in line with text in paragraph 4; suggested change to text to clarify there should be oversight by one common Competent Authority **Supporting evidence:** n/a	Did not agree, see response to comment 4.3._1.
4.3.1._5_New Caledonia	**Proposition ou commentaire :** Commentaire editorial “La compartimentation offre la possibilité à l’exploitant ~~au secteur privé~~ de démontrer l'absence de *maladie* au niveau des ~~entreprises~~ établissements d’aquaculture, notamment [...]” **Exposé des motifs :** Article 4.3.1. 3ème alinéa : dans ce paragraphe, la commission a souscrit au commentaire d’un membre proposant de remplacer les termes “secteur privé” par “exploitant” car les structures n’appartiennent pas toujours au secteur privé. Dans le même esprit, il semblerait préférable de remplacer le terme “entreprises” par “établissements d’aquaculture”.	Did not agree, the term operator refers to the individual or entity managing the compartment. Regarding its use in demonstrating freedom, operator is the more appropriate term compared to aquaculture establishment.

	Justification, s'il y a lieu :	

A *Competent Authority* can make a *self-declaration of freedom from disease* for a *compartment* from specified *listed diseases* ~~can be made~~ if the requirements of this chapter to establish a *compartment* are met and the requirements for making a *self-declaration of freedom from disease* including compliance with *basic biosecurity conditions* and *surveillance* requirements as described in Chapter 1.4. and in the relevant disease-specific chapters have been met.

4.3.1._6_China (People's Republic of)	**Category:** change **Proposed amended text:** Compartmentalisation provides a means of demonstrating that an aquaculture establishment is free from one or more specified diseases by establishing and maintaining functional epidemiological separation between the aquatic animals within the compartment and sources of infection outside the compartment. A compartment may comprise a single aquaculture establishment or a group of interrelated aquaculture establishments with clear epidemiological linkage (e.g., shared water sources, movement of aquatic animals) or a unified management boundary (e.g., common operator, integrated biosecurity system) that operate under a common set of risk management measures in accordance with this chapter. … The ~~A~~ *Competent Authority* can make a *self-declaration of freedom from disease* for a *compartment* from specified *listed diseases* ~~can be made~~ if the requirements of this chapter to establish a *compartment* are met and the requirements for making a *self-declaration of freedom from disease* including compliance with *basic biosecurity conditions* and *surveillance* requirements as described in Chapter 1.4. and in the relevant disease-specific chapters have been met. **Rationale:** China agrees that the new revisions are clearer. However, the present text describes a compartment as "a group of interrelated aquaculture establishments" without defining interrelatedness, creating ambiguity in implementation. This could lead to unrelated establishments being incorrectly grouped, undermining biosecurity oversight, surveillance consistency, and disease control effectiveness. By explicitly requiring "epidemiological linkage" (e.g., shared water sources, animal movement) or "unified management boundary" (e.g., common operator, integrated biosecurity), the revision provides clear criteria for grouping establishments. This ensures logical compartmentalisation delineation, supports targeted risk management, and safeguards the compartment's disease-free status. In addition, the Competent Authority is a specific authority of a country. Here, the definite article "the" should be used instead of the indefinite article "a", which is also in line with the usage throughout the code.	Did not agree, considered clear that a group of aquaculture establishments under a common set of risk management measures in a compartment are linked. Providing examples may seem to restrict options for establishment of a compartment, which is not the intention of this article. In the final paragraph, 'A Competent Authority' was revised to 'The Veterinary Authority' as the Veterinary Authority is the responsible authority to make a self-declaration of freedom from disease.

	supporting evidence: not relevant	
4.3.1._7_Korea (Republic of)	**Category:** Addition **Proposed amended text:** A compartment may also consist of aquaculture establishments with different production system types. However, this is only acceptable provided that these establishments operate as a single integrated epidemiological unit, where all transmission pathways between establishments are controlled, and consistent, compartment-wide disease-specific risk management measures—including biosecurity, surveillance, and movement control—are applied across the system to achieve effective separation from external sources of infection. In such cases, the Competent Authority should verify that the overall system functions as a single epidemiological unit and that effective disease-specific separation from external infection sources is achieved, and may approve the integrated system as a single compartment. **Rationale:** The current draft revision categorizes compartments as independent or dependent, which generally correspond to closed or semi-closed production systems. However, in practice, aquaculture establishments with different production system types are often integrated and managed as a single epidemiological unit. For instance, in shrimp aquaculture, broodstock and larval production facilities may operate as closed systems, whereas intermediate and grow-out facilities operate as semi-closed systems. Since a compartment is defined as an epidemiological unit, it is necessary to assess the entire integrated system to determine whether it can be recognized as a single compartment. When establishments with different production systems operate as a single epidemiological unit and consistent, disease-specific risk management measures are applied across the entire compartment, the integrated system should be recognized as a single compartment. The Competent Authority should verify the adequacy of these measures, with the detailed modalities of evaluation determined at the discretion of the Competent Authority, considering the characteristics of independent and dependent compartments. Therefore, it is proposed that a new paragraph reflecting these considerations be added to the draft revision under Article 4.3.1. **Supporting evidence:** -	See response to comment 4.3._3.
4.3.1._8_New Zealand	**Category:** editorial **Proposed amended text:**	Did not agree, the Competent Authority does not establish the compartment which is completed by the

	A *Competent Authority* can ~~make a~~ establish ~~*self-declaration of freedom from disease for*~~ a *compartment* ~~from~~ for specified *listed diseases* ~~can be made~~ if the requirements of this chapter ~~to establish a *compartment*~~ are met, and the requirements for making a *self-declaration of freedom from disease* including compliance with *basic biosecurity conditions* and *surveillance* requirements as described in Chapter 1.4. and in the relevant disease-specific chapters have been met. **Rationale:** This is a very long sentence – suggested edits to reduce some of the duplication of wording and shorten it. **Supporting evidence** n/a	operator. The Competent Authority has official oversight and in the case of the Veterinary Authority responsibility to make self-declaration of freedom and notifications regarding the compartment.
4.3.1._9_Peru	**Categoría:** Adición **Texto modificado propuesto:** "…incluido el cumplimiento de las condiciones elementales de *bioseguridad*, descritos en el Capítulo 1.4." **Justificación:** Se requiere que se alinee el término de "bioseguridad" en concordancia con el glosario del código acuático. **Evidencia documentada:** Glosario del código acuático de la OMSA. Compartimento. - designa uno o varios establecimientos de acuicultura con un mismo sistema de gestión de la bioseguridad, que contienen una población de animales acuáticos con un estatus sanitario distinto respecto de una enfermedad o enfermedades determinada(s) contra la(s) cual(es) se aplican las medidas de vigilancia y control y se cumplen las condiciones elementales de **bioseguridad** requeridas para el comercio internacional. Cualquier compartimento establecido debe estar claramente documentado por la autoridad competente.	Agreed, translation change to be made to Spanish version.

Article 4.3.2.

Principles for establishing a compartment

The following principles should be applied to establish and maintain a *free compartment*. The principles should be addressed in a *self-declaration of freedom from disease* as described in Chapter 1.4.

1) A *compartment* must ensure there are effective measures to prevent the entry or spread of *pathogenic agents* from the external environments into the *compartment* (i.e. provide functional epidemiological separation);

2) The purpose and scope of a *compartment* should be clearly defined (e.g. *disease(s)* for which freedom will be claimed species produced and *aquaculture establishments* that comprise the *compartment*) as described in Article 4.3.3.;

4.3.2._1_Canada	**Comment Category:** editorial **Proposed amended text:** The purpose and scope of a *compartment* should be clearly defined (e.g. *disease(s)* for which freedom will be claimed, species produced and *aquaculture establishments* that comprise the *compartment*) as described in Article 4.3.3.; **Rationale:** editorial, a comma is required after 'claimed'.	Agreed to editorial change.

3) There are two categories of *compartments* (i.e. those with disease-free status that is dependent on the *disease* status of the surrounding environment or those with disease-free status that is independent from the *disease* status of the surrounding environment, in Article 4.3.4.) and *biosecurity* and *surveillance* measures should be appropriate for the category of *compartment*;

4.3.2._2_Thailand	**Category:** Editorial **Proposed amended text:** Propose rearranging the text to first address independent compartment, followed by dependent compartment. 3) There are two categories of compartments (i.e. those with disease-free status that is independent from ~~dependent on~~ the disease status of the surrounding environment or those with disease-free status that is ~~independent from~~ on the disease status of the surrounding environment, in Article 4.3.4.) and biosecurity and surveillance measures should be appropriate for the category of compartment; **Rationale:** To highlight that an independent compartment provides a higher level of epidemiological separation from the surrounding environment and to align with Article 4.3.4. **Supporting evidence:** -	Agreed to editorial change.
4.3.2._3_United States	**Category:** deletion **Proposed amended texts (or precise suggested deletion):** "~~3) There are two categories of *compartments* (i.e. those with disease-free status that is dependent on the *disease* status of the surrounding environment or those with disease-free status that is independent from the *disease* status of the surrounding environment, in Article 4.3.4.) and *biosecurity* and *surveillance* measures should be appropriate for the category of *compartment*;~~" **Rationale:** We strongly recommend that the Commission removes dependent compartments from the WOAH Aquatic Code to maintain adherence to the established principles of compartmentalization that support disease control, trade resilience, and international standards, and which are already being successfully implemented by Competent Authorities for other WOAH-listed pathogens. See additional rationale in the general comment provided above.	Did not agree, see response to comment 4.3._8.

	Supporting evidence: -	

4) A *biosecurity plan* must be developed and maintained in accordance with Chapter 4.1. and applied consistently across all elements of the *compartment* as described in Article 4.3.5.;

5) *Surveillance* measures to demonstrate that the *compartment* is free from specified *diseases*, and to maintain its free status, must be clearly described in accordance with Chapter 1.4., including elements of internal and external *surveillance* as appropriate, as described in Article 4.3.6.;

6) *Surveillance* testing must be supported by reliable laboratory testing services which have independence from the *compartment* operator and which are approved by *Competent Authority*, as described in Article 4.3.7.;

4.3.2._4_Australia	**Category:** editorial **Proposed amended text:** 6) Surveillance testing must be …are approved by the Competent Authority, as described in Article 4.3.7.; **Rationale**: n/a **Supporting evidence:** n/a	Agreed to editorial change.
4.3.2._5_Canada	**Comment Category:** editorial **Proposed amended text:** *Surveillance* ~~testing~~ must be supported by reliable laboratory testing services which have independence from the *compartment* operator and which are approved by *Competent Authority*, as described in Article 4.3.7.; **Rationale:** the glossary definition of surveillance includes testing, therefore inclusion of 'testing' in point 6 is unnecessary.	Agreed to remove 'testing' as included in definition of surveillance.
4.3.2._6_Chile	**Categoría**: Adición **Texto modificado propuesto:** Las pruebas de vigilancia deberán estar respaldadas por pruebas de laboratorio fiables ~~e~~ y realizadas en laboratorios independientes del operador del compartimento y que estén aprobadas por la autoridad competente, tal y como se describe en el Artículo 4.3.7.; **Justificación:** Este comentario fue aportado en el anexo 19 en consulta enviado en julio del 2025, y de acuerdo con la respuesta de la comisión en anexo 3 señala que este comentario fue aceptado, sin embargo, no se ve reflejado en anexo 6 en consulta. Las pruebas de laboratorio no son independientes sino los laboratorios donde se realizan estas pruebas. Estos laboratorios deben ser autorizados por la autoridad competente y cumplir con Capítulo: 1.1.1. Gestión de la calidad en los laboratorios de pruebas veterinarias del	Agreed, translation change in Spanish version only.

	Manual de pruebas de diagnóstico para los animales acuáticos. **Evidencia de apoyo:** N/A	
4.3.2._7_New Caledonia	**Proposition ou commentaire :** Commentaire editorial "6) les analyses réalisées dans le cadre de la *surveillance* doivent être effectuées par des services d'analyse de laboratoire fiables, ces laboratoires étant indépendants des exploitants des établissements d'aquaculture compris dans le ~~l'exploitant du~~ *compartiment* et agréés par l'*Autorité compétente*, comme décrit dans l'article 4.3.7. ;" **Exposé des motifs :** Article 4.3.2. 6) les termes "exploitant du compartiment" ne semblent pas adaptés. En effet, il peut y avoir plusieurs exploitants de plusieurs établissements d'aquaculture dans un seul compartiment. Il est donc proposé de reformuler ainsi **Justification, s'il y a lieu : /**	Did not agree, operator shouldn't be plural in point 6, as there needs to be one operator that has oversight and is responsible for the compartment. Text was added to paragraph 2 of Article 4.3.1. stating that a compartment must be under the control of a single operator.

7) Traceability systems must provide assurance of provenance of *commodities* from the *free compartment*, as described in Article 4.3.8.;

4.3.2._8_New Caledonia	**Proposition ou commentaire :** Commentaire editorial "7) les systèmes de traçabilité doivent permettre de garantir l'origine/la source~~a provenance~~ des *marchandises* ~~issues du~~ produites dans un *compartiment indemne*, comme décrit dans l'article 4.3.8. ;" **Exposé des motifs :** Article 4.3.2. 7) : il est proposé de reformuler ce point pour clarifier le sens de la phrase. En effet, les termes "provenance des marchandises issues du compartiment" peuvent prêter à confusion, car la provenance de marchandises issues d'un compartiment est justement ce compartiment. Il semble préférable de parler "d'origine", tel qu'utilisé au 4.3.8.2 ou de "source", tel qu'utilisé au 4.3.5.2., et de parler de "marchandises produites dans ce compartiment" **Justification, s'il y a lieu : /**	Did not agree, the word provenance is appropriate and indicates the origin/source of the commodity.

8) Record keeping must provide evidence of the ongoing application of all measures on which the *compartment* has been granted disease-free status, as described in Article 4.3.9.;

9) Official oversight responsibilities must be clearly documented, including approval by the *Competent Authority*, an auditing schedule, underpinning regulatory instruments and authorising third parties within the *Aquatic Animal Health Services* for important roles, as described in Articles 4.3.10. and 4.3.11.;

4.3.2._9_Canada	**Comment Category:** editorial **Proposed amended text:** Official oversight responsibilities must be clearly documented, including approval by the *Competent Authority*, an auditing schedule, underpinning regulatory instruments and ~~authorising~~ approving third parties within the *Aquatic Animal Health Services* for important roles, as described in Articles 4.3.10. and 4.3.11.; **Rationale:** To align with wording in Article 4.3.10	Agreed, editorial change to align with use of 'approving' in Articles 4.3.10. and 4.3.11.

10) *Notification* and response measures must be in place in the event of detection of the *disease* for which the *compartment* has been declared free, or for other *diseases* relevant to trade from the *compartment*, as described in Article 4.3.12.;

4.3.2._10_China (People's Republic of)	**Category:** change **Proposed amended text :** Article 4.3.2. **Principles for establishing and maintaining a compartment** The following principles should be applied to establish and maintain a ~~*free*~~ *compartment*. These ~~The~~ principles should be addressed in a *self-declaration of freedom from disease* as described in Chapter 1.4. 1) A *compartment* must ensure there are effective measures to prevent the entry or spread of *pathogenic agents* from ~~the~~ external environments into the *compartment* (i.e. provide functional epidemiological separation); 2) The purpose and scope of a *compartment* should be clearly defined (e.g. *disease(s)* for which freedom will be claimed, species produced, and *aquaculture establishments* that comprise the *compartment*) as described in Article 4.3.3.; 3) There are two categories of *compartments* (i.e. those with disease-free status that is dependent on the *disease* status of the surrounding environment or those with disease-free status that is independent from the *disease* status of the surrounding environment, in Article 4.3.4.) and *biosecurity* and *surveillance* measures should be appropriate for the category of the *compartment*; … 8) Record keeping must provide evidence of the ongoing application of all measures, on which the *compartment* has been granted disease-free status, as described in Article 4.3.9.;	Agreed to change title of article to include 'and maintaining'. In first paragraph, did not agree to remove 'free' as 'free compartment' is a Glossary term that includes that the compartment fulfills the requirements for self-declaration of freedom from disease. Agreed to editorial change in second sentence. Point 1 – deleted 'from the external environment' to simplify the point as that text was redundant. Point 3 and 9 – did not agree with editorial changes, considered clear as written. Point 10 – agreed with editorial change.

	9) Official oversight responsibilities must be clearly documented, including approval by the *Competent Authority*, an auditing schedule, underpinning regulatory instruments and authorising third parties within ~~the~~ *Aquatic Animal Health Services* for important roles, as described in Articles 4.3.10. and 4.3.11.; 10) *Notification* and response measures must be in place in the event of detection of a ~~the~~ *disease*, for which the *compartment* has been declared free, or for other *diseases* relevant to trade from the *compartment*, as described in Article 4.3.12.; **Rationale:** The title is a compartment and not a free compartment. The establishment of a compartment is based on the status of freedom for specific diseases. It's not necessary to use a free compartment. The other is some minor grammatical revisions. **Supporting evidence:** not relevant	
4.3.2._11_New Zealand	**Category:** editorial **Proposed amended text:** 1) ~~A *compartment* must ensure there are e~~Effective measures are established to prevent the entry or spread of *pathogenic agents* from the external environments into the *compartment* (i.e. provide functional epidemiological separation); 2) The purpose and scope of a *compartment* should be clearly defined (e.g. *disease(s)* for which freedom will be claimed, species produced and *aquaculture establishments* that comprise the *compartment*) as described in Article 4.3.3.; 3) There are two categories of *compartments* (i.e. those with disease-free status that is dependent on the *disease* status of the surrounding environment or those with disease-free status that is independent from the *disease* status of the surrounding environment, in Article 4.3.4.) and *biosecurity* and *surveillance* measures should be appropriate for the category of *compartment*; 4) A *biosecurity plan* must be developed and maintained in accordance with Chapter 4.1. and applied consistently across all elements of the *compartment* as described in Article 4.3.5.; 5) *Surveillance* measures to demonstrate that the *compartment* is free from specified *diseases*, and to maintain its free status, must be clearly described in accordance with Chapter 1.4., including elements of internal and external *surveillance* as appropriate, as described in Article 4.3.6.;	Point 1 – 'ensure there are' was deleted and replaced with 'have' to simplify the sentence. Point 6 – Did not agree, Article 4.3.7. addresses the need for a quality management system. Point 10 – Did not agree, Article 4.3.12. addresses the need for notification and response measures in the case of a biosecurity breach.

	6) *Surveillance* testing must be supported by reliable laboratory ~~testing~~ services with a quality management system that meets requirements of Chapter 1.1.1. of the *Aquatic Manual*, have independence from the *compartment* operator, and which are approved by *Competent Authority*, as described in Article 4.3.7.; 7) Traceability systems must provide assurance of provenance of *commodities* from the *free compartment*, as described in Article 4.3.8.; 8) Record keeping must provide evidence of the ongoing application of all measures on which the *compartment* has been granted disease-free status, as described in Article 4.3.9.; 9) Official oversight responsibilities must be clearly documented, including approval by the *Competent Authority*, an auditing schedule, underpinning regulatory instruments and authorising third parties within the *Aquatic Animal Health Services* for important roles, as described in Articles 4.3.10. and 4.3.11.; 10) *Notification* and response measures must be in place in the event of detection of the *disease* for which the *compartment* has been declared free, or for other *diseases* relevant to trade from the *compartment*, or following any event which could lead to a breach of *biosecurity* measures, as described in Article 4.3.12.; **Rationale:** - The compartment is not responsible for establishing the measures. - Specified that laboratory services should meet same requirements as detailed in 1.4 - Added that notification & response measures should also in place in for an event that might indicate a breach in biosecurity measures (which is also addressed in 4.3.12). **Supporting evidence** n/a	

Article 4.3.3~~2~~.

Purposes and scope of compartments

Compartments provide an opportunity for trade of disease-free *commodities* from a *zone* or *country* not declared free. They can also be used to provide epidemiological separation for populations of valuable *aquatic animals* within a *free country* or *free zone* to protect them in the event of a *disease outbreak*. The *disease(s)* for which freedom will be claimed and the species produced should be clearly defined.

4.3.3._1_Chile	**Categoría**: Cambio **Texto modificado propuesto**: Los compartimentos pueden establecerse con el propósito de la prevención y el control de enfermedades, a fin de facilitar el comercio. **Justificación:** El primer propósito del compartimento debe ser la prevención y control de enfermedades, ya que es lo básico para establecer el comercio. **Evidencia de apoyo**: N/A	Did not agree, the primary purpose for establishing a compartment is to facilitate trade.
4.3.3._2_China (People's Republic of)	**Category:** change **Proposed amended text :** **Purposes and scope of compartments** *Compartments* provide an opportunity for trade of disease-free *commodities* from a *zone* or *country* not declared free. They can also be used to provide epidemiological separation for populations of valuable *aquatic animals* within a *free country* or *free zone* to protect them in the event of a *disease outbreak*. The *disease(s)* for which freedom will be claimed, ~~and~~ the species produced, and *aquaculture establishments* that comprise the *compartment* should be clearly defined. **Rationale:** As described in article 4.3.2 point 2, not only the diseases and species but also the information of the aquaculture establishment should be defined. **supporting evidence:** not relevant	Agreed to include that the aquaculture establishments that comprise the compartment should be clearly defined.
4.3.3._3_New Caledonia	**Proposition ou commentaire :** Commentaire éditorial "La ou les maladies pour lesquelles le statut indemne sera demandé~~e~~ et les espèces faisant l'objet de la production doivent être définies de manière claire." **Exposé des motifs** : Article 4.3.3. dernière phrase : faute d'accord : "demandé" s'accorde avec statut et non avec "maladie". **Justification, s'il y a lieu :/**	Agreed, translation change in French version only.
4.3.3._4_New Zealand	**Category:** editorial **Proposed amended text:** *Compartments* provide an opportunity for trade of disease-free *commodities* from a *zone* or *country* not declared free. They can also be used to provide epidemiological separation for	Did not agree, defining species being produced within the compartment are the relevant susceptible species for which

	populations of valuable *aquatic animals* within a *free country* or *free zone* to protect them in the event of a *disease outbreak*. The *disease(s)* and relevant *susceptible species* for which freedom will be claimed ~~and the species produced~~ should be clearly defined. **Rationale:** Removed the word "produced" and added "susceptible species" to take into account the potential for epidemiological connections with wild susceptible species populations, that may need to be considered. **Supporting evidence** n/a	freedom will be claimed. Any connections to wild species will require risk management measures as identified through risk analysis.
4.3.3._5_Norway	**Category**: deletion **Proposed amended text**: ~~Article 4.3.32.~~ ~~**Purposes and scope of compartments**~~ ~~*Compartments* provide an opportunity for trade of disease free *commodities* from a *zone* or *country* not declared free. They can also be used to provide epidemiological separation for populations of valuable *aquatic animals* within a *free country* or *free zone* to protect them in the event of a *disease outbreak*. The *disease(s)* for which freedom will be claimed and the species produced should be clearly defined.~~ **Rationale:** The text in this article is largely a repetition from Article 4.3.1. We encourage the Commission to consider whether this additional article is needed. **Supporting evidence, if relevant:** not relevant	Did not agree to delete article. This article is intended to cover and highlight general principles which will be elaborated on through the following articles.

~~There may be a range of *commodities* produced by a *compartment* and possible end-uses. The *commodity* types (e.g. *aquatic animals*, *aquatic animal products*) and end-uses (e.g. for *aquaculture*, stocking of natural water bodies, human consumption, *ornamental aquatic animals*) have implications for *risk management* and should be defined.~~

~~Article 4.3.3.~~

~~**Principles for establishing a compartment**~~

~~The following principles should be applied to establish and maintain a *free compartment*.~~

~~1)~~ ~~A *compartment* must ensure there are effective measures to prevent the entry or spread of *pathogenic agents* between the *compartment* and external environments (i.e. provide functional epidemiological separation);~~

2) the purpose of a *compartment* should be clearly defined (e.g. *disease(s)* for which freedom will be claimed, species and *commodities* produced, intended end-uses of *commodities*) as this will have implications for the design of *risk management* measures, as described in Article 4.3.2.;

3) *biosecurity* and *surveillance* measures should be appropriate for the category of *compartment*, i.e. those with disease-free status that is dependent on the *disease* status of the surrounding environment or those with disease-free status that is independent from the *disease* status of the surrounding environment, in Article 4.3.4.;

4) a *biosecurity plan* must be developed and maintained in accordance with Chapter 4.1. and applied consistently across all elements of the *compartment* as described in Article 4.3.5.;

5) *surveillance* measures to demonstrate that the *compartment* is free from specified *diseases*, and to maintain its free status, must be clearly described in accordance with Chapter 1.4., including elements of internal and external *surveillance* as appropriate, as described in Article 4.3.6.;

6) *surveillance* testing must be supported by reliable laboratory testing services which have independence from the *compartment* operator and which are approved by *Competent Authority*, as described in Article 4.3.7.;

7) traceability systems must provide assurance of provenance of *commodities* from the *free compartment*, as described in Article 4.3.8.;

8) record keeping must provide evidence of the ongoing application of all measures on which the *compartment* has been granted disease-free status, as described in Article 4.3.9.;

9) official oversight responsibilities must be clearly documented, including approval by the *Competent Authority*, an auditing schedule, underpinning regulatory instruments and authorising third parties within the *Aquatic Animal Health Services* for important roles, as described in Articles 4.3.10. and 4.3.11.;

10) notification and response measures must be in place in the event of detection of the *disease* for which the *compartment* has been declared free, or for other *diseases* relevant to trade from the *compartment*, as described in Article 4.3.12.;

Article 4.3.4.

4.3.4._1_Chinese Taipei	**Category:** Addition **Proposed amended text:** Request the inclusion of a definition of "epidemiological separation"' in Article 4.3.4 (Independent compartments). **Rationale:** The definition of epidemiological separation is not clearly stated in the article. **Supporting evidence:** Not relevant.	The use of epidemiological separation was clarified in point 1 in Article 4.3.3.

Independent and dependent ~~Dependent and independent~~ compartments

There are two categories of *compartments* that are determined by the degree of epidemiological separation from the surrounding environment: independent and dependent *compartments*. ~~Independent *compartments* have complete epidemiological separation from the surrounding environment and are characterised by appropriate levels of physical and management measures to maintain effective *biosecurity*. Dependent *compartments* do not have complete epidemiological separation from the surrounding environment and may require the application of appropriate *risk* mitigation measures to achieve and maintain disease-free status despite epidemiological links to the surrounding environment.~~

~~If such *risk* mitigation measures cannot be applied successfully, a dependent *compartment* cannot be approved by the *Competent Authority*. The concept of dependent *compartments* enables compartmentalisation to be applied to more types of production systems and more establishments, increasing opportunities to trade in disease-free *commodities* where these *compartment* types provide an appropriate level of *risk management*.~~

Independent compartments

Independent *compartments* have complete epidemiological separation from the surrounding environment and are characterised by appropriate levels of physical and management measures to maintain effective *biosecurity*.

~~Independent and dependent *compartments* and have the following characteristics:~~

~~1)~~ Independent *compartments* have the following characteristics:

1~~a~~) are closed production system types only (as described in Chapter 4.1.);

2~~b~~) have control over all transmission pathways and complete epidemiological separation from surrounding environments;

4.3.4._2_Chile	**Categoría**: Cambio **Texto modificado propuesto**: 2) Tienen identificadas y controladas ~~el control de~~ todas las rutas de transmisión y una separación epidemiológica completa del entorno circundante; **Justificación:** Para controlar las rutas de transmisión deben previamente identificar. **Evidencia de apoyo:** N/A	Agreed that transmission routes must be identified in order allow control. Text was revised to reflect this comment.
4.3.4._3_New Caledonia	**Proposition ou commentaire :** Commentaire éditorial “~~b~~2) ont un contrôle sur toutes les voies de transmission et sont indépendants du statut sanitaire des eaux ~~environnantes présentent une séparation épidémiologique complète avec les milieux environnants~~ ;” **Exposé des motifs** : Article 4.3.4. point 2) sur les compartiments indépendants : il parait inutile de préciser à nouveau qu’ils “présentent une séparation épidémiologique complète avec les milieux environnants”, alors que c’est déjà précisé dans la définition du compartiment indépendant mentionnée juste au-dessus. Il est donc proposé de le reformuler sous la même forme que celle utilisée pour le point 2) des compartiments dépendants **Justification, s’il y a lieu : /**	Agreed to deleted ‘complete’ as while having complete epidemiological separation is the ideal it is recognized that systems can fail and this may not always be possible. Did not agree with remaining proposed text, environment is maintained in the point as there is a need to control other factors within the environment.

4.3.4._4_Norway	**Category**: addition / deletion / general **Proposed amended text**: 2) have control ~~over~~ of all transmission pathways and ~~complete~~ epidemiological separation from surrounding environments; **Rationale:** A complete epidemiological separation implies perfect systems. In practice these doesn't exist, and it would mean that no system will be able to meet the definition. The definition of a closed system in Article 4.1.5. refers to "sufficient control over water entering and exiting the system". We therefore suggest that the word "complete" is removed. We additionally suggest using "control of" rather than "control over" transmission pathways. Alternatively, the Commission could review whether point 1) might be sufficient given the definition of a closed system. If so, we suggest that this point 2) is deleted. **Supporting evidence, if relevant:** not relevant	Agreed to deleted 'complete' as while having complete epidemiological separation is the ideal it is recognized that systems can fail and this may not always be possible.

3~~c~~) have appropriate levels of *biosecurity* and *surveillance* to mitigate the *risk* of introduction of specific *pathogenic agents* into the *compartment* in accordance with Article 4.3.5.~~physical and management measures to maintain effective *biosecurity* for all pathways;~~

~~d) provide levels of *risk management* mitigation suitable for all purposes, *commodity* types and end-uses;~~

4.3.4._5_New Caledonia	**Proposition ou commentaire :** Commentaire éditorial "3) disposent de niveaux appropriés de *sécurité biologique* et de *surveillance* pour atténuer le *risque* d'introduction de ou des agents pathogènes ~~spécifiques~~ dans le *compartiment* (pour lesquels le *compartiment* a été établi), y compris depuis le milieu environnant, comme indiqué par une analyse des risques et conformément à l'article 4.3.5. ~~mesures physiques et de gestion pour maintenir une *sécurité biologique* efficace pour toutes les voies de transmission~~ ; ~~d4) satisfont aux critères supplémentaires de sécurité biologique et de surveillance visant à atténuer le risque de transmission depuis le milieu environnant, comme indiqué par une analyse des risques, aux mesures d'atténuation des risques pour la transmission à la faveur des entrées d'eaux, que l'Autorité compétente peut approuver conformément à l'article 4.3.5. ;"~~ **Exposé des motifs** : Article 4.3.4. point 3) et 4) des compartiments dépendants : pourquoi faire 2 points sur ce sujet ? En effet le point 4) est déjà compris dans le point 3), il est	Did not agree to add specific text about introduction of pathogenic agents from surrounding water bodies. This detail is inluded in the first point of 4.3.2. and does not need to be re-iterated there.

	proposé de supprimer le point 4) et de reformuler le point 3) pour insister sur le risque issu du milieu environnant. **Justification, s'il y a lieu :**	

~~e) are often preferred for high value *aquatic animals* (e.g. genetically improved lines, brood stock).~~

~~2)~~ *Dependent compartments*

4.3.4._6_New Zealand	**Category:** editorial/general comment **Proposed amended text:** n/a **Rationale:** The terms "surrounding waters", "shared water body" and "environment surrounding the compartment" have been used in reference to dependent compartments. It is unclear if these terms are interchangeable, and recommend one should be chosen for consistency and clarity, and consider if chosen term should be explicitly defined within the chapter at least once. **Supporting evidence** n/a	Agreed, the text was reviewed and the term 'surrounding water body' used consistently throughout the text.

Dependent *compartments* do not have complete epidemiological separation from the surrounding environment but conditions exist which create an effective disease-specific separation between the *compartment* and other *aquatic animal* populations that may be infected. The possibility of achieving such disease-specific separation will be determined by the *Competent Authority*, based on *risk analysis*.

4.3.4._7_Norway	**Category**: deletion / addition **Proposed amended text**: Dependent *compartments* ~~do not~~ have ~~complete~~ limited epidemiological separation from the surrounding environment but conditions exist which create an effective disease-specific separation between the *compartment* and other *aquatic animal* populations that may be infected. The possibility of achieving such disease-specific separation must be based on risk assessment and ~~will~~ be determined by the *Competent Authority*~~, based on *risk analysis*~~. **Rationale:** See comment under Article 4.3.4 regarding complete epidemiological separation. Suggest using the word «limited» instead of "complete", as the definition of a semi-closed system refers to, for example, "some control over the water entering and exiting the system" and semi-open systems to	Agreed that referencing complete epidemiological separation as the intention is to establish that there is a hydrological connection in dependent compartments. The text was revised to reflect this comment. Did not agree to add that the separation should be based on risk assessment. The Glossary term of risk analysis is the 'process of hazard

	"not possible to have control over the water entering or exiting the system". As it may or may not be the Competent Authority performing the actual risk assessment we suggest rewriting the last sentence. **Supporting evidence, if relevant:** not relevant	identification, risk assessment, risk management and risk communication' and is the more appropriate term.

The concept of dependent *compartments* broadens the application of compartmentalisation to a wider range of production systems and *aquaculture establishments*, creating additional opportunities for trade in disease-free commodities where such *compartments* ensure an appropriate level of *risk management*.

Dependent compartments have the following characteristics:

a) are semi-closed production system types only (as described in Chapter 4.1.);

4.3.4._8_Norway	**Category**: addition **Proposed amended text**: a) are semi-open or semi-closed production system types only (as described in Chapter 4.1.); **Rationale:** Norway would like to reiterate its view that semi-open production systems should not be automatically excluded from the possibility of becoming a dependent compartment. This should rather be based on epidemiological risk assessments and whether or not an establishment is able to fulfil the required criteria. An example of this approach is (EU) 2020/689 Article 73 (2b and 3). **Supporting evidence, if relevant:** not relevant	See report item 6.3. – Commission requests Member feedback.

4.3.4._9_EU	**Category:** Addition **Proposed amended text:** a) are semi-closed production system types or semi-open systems (as described in Chapter 4.1.); **Rationale**: if the Competent Authority performs a risk assessment considering the risk factors outlined in Article 4.3.5 and concludes that the compartment can be established safely, there is no reason for excluding well selected safe establishments from the possibility to become dependent compartment with the objective of obtaining disease freedom.	See report item 6.3. – Commission requests Member feedback.

b) are dependent on the health status of the surrounding waters;

c) have appropriate levels of *biosecurity* and *surveillance* to mitigate the *risk* of introduction of ~~specific~~ the *pathogenic agent(s)* (for which the *compartment* has been established) in accordance with Article 4.3.5.~~physical and management measures to maintain effective *biosecurity* for all pathways~~;

d) meet the additional *biosecurity* and *surveillance* requirements to mitigate transmission *risk* from the surrounding environment as informed by a *risk analysis* ~~via intake water criteria and *risk* mitigation measures for transmission via intake water which the *Competent Authority* may approve~~ in accordance with Article 4.3.5.;

4.3.4._10_Canada	**Comment Category:** Change **Proposed amended text:** d) meet the additional *biosecurity* and *surveillance* requirements to mitigate transmission *risk* from the surrounding water ~~environment~~ as informed by a *risk analysis* ~~via intake water criteria and *risk* mitigation measures for transmission via intake water which the *Competent Authority* may approve~~ in accordance with Article 4.3.5.; **Rationale:** point c addresses the normal biosecurity that would be required to address the risk associated with introduction from the surrounding environment (animal, air, fomites, feed). These environmental risks (animal, air, fomites, feed) should be mitigated the same as they are for an independent compartment. Additional biosecurity risks (point d) are those solely associated with dependent compartments. This should address the risks of influent water or surrounding waters	See response to comment 4.3.4_1.

~~e) may not provide sufficient *risk* mitigation for all purposes, *commodity* types and end-uses (e.g. supplying live *aquatic animals* for *aquaculture* or restocking, for high value *aquatic animals* such as genetically improved lines).~~

The suitability of a dependent *compartment* should be assessed against the minimum *risk* factors outlined in Article 4.3.5. If these measures cannot be effectively implemented, the *Competent Authority* cannot grant approval. Where effective disease-specific separation is possible, approval may be granted provided that specific *risk management* measures are applied. Both the *risk analysis* and the specified measures must be documented in the dossier of evidence referred to in Article 1.4.16.

4.3.4._11_New Caledonia	**Proposition ou commentaire :** Commentaire éditorial "Lorsqu'une séparation efficace spécifique à la maladie ~~efficace~~ est possible, celui-ci peut être approuvé, sous réserve que des mesures spécifiques de *gestion du risque* soient appliquées." **Exposé des motifs** : Article 4.3.4. dernier alinéa : il est proposé de déplacer le terme "efficace" juste après le mot "séparation" qu'il qualifie pour clarifier le sens de la phrase, car ainsi on a l'impression que c'est la "maladie" qui est "efficace" **Justification, s'il y a lieu : /**	Agreed, translation change in French version only.
4.3.4._12_United States	**Category:** change, deletion **Proposed amended texts (or precise suggested deletion):** "~~Independent and dependent~~ ~~Dependent and independent~~ ~~c~~Compartment~~s~~ characteristics" "~~There are two categories of *compartments* that are determined by the degree of epidemiological separation from the surrounding environment: independent and dependent *compartments*.~~" "~~*Independent compartments*~~" "~~Independent *c~~C~~ompartments* have complete epidemiological separation from the surrounding environment and are characterised by the following appropriate levels of physical and management measures to maintain effective *biosecurity*~~.~~:" "~~1) Independent compartments have the following characteristics~~:" "~~2) *Dependent compartments*~~ ~~Dependent *compartments* do not have complete epidemiological separation from the surrounding environment but conditions exist which create an effective disease-specific separation between the *compartment* and other *aquatic animal* populations that may be infected. The possibility of achieving such disease-specific separation will be determined by the *Competent Authority*, based on *risk analysis*.~~ ~~The concept of dependent *compartments* broadens the application of compartmentalisation to a wider range of production systems and *aquaculture establishments*, creating additional opportunities for trade in disease-free~~	Did not agree, see response to comment 4.3._8.

	~~commodities where such *compartments* ensure an appropriate level of *risk management*.~~ ~~Dependent compartments have the following characteristics:~~ ~~a) are semi-closed production system types only (as described in Chapter 4.1.);~~ ~~b) are dependent on the health status of the surrounding waters;~~ ~~c) have appropriate levels of *biosecurity* and *surveillance* to mitigate the *risk* of introduction of specific the *pathogenic agent(s)* (for which the *compartment* has been established) in accordance with Article 4.3.5.physical and management measures to maintain effective *biosecurity* for all pathways;~~ ~~d) meet the additional *biosecurity* and *surveillance* requirements to mitigate transmission *risk* from the surrounding environment as informed by a *risk analysis* via intake water criteria and *risk* mitigation measures for transmission via intake water which the *Competent Authority* may approve in accordance with Article 4.3.5.;~~ ~~e) may not provide sufficient *risk* mitigation for all purposes, *commodity* types and end uses (e.g. supplying live *aquatic animals* for *aquaculture* or restocking, for high value *aquatic animals* such as genetically improved lines).~~ ~~The suitability of a dependent *compartment* should be assessed against the minimum *risk* factors outlined in Article 4.3.5. If these measures cannot be effectively implemented, the *Competent Authority* cannot grant approval. Where effective disease-specific separation is possible, approval may be granted provided that specific *risk management* measures are applied. Both the *risk analysis* and the specified measures must be documented in the dossier of evidence referred to in Article 1.4.16.~~" **Rationale:** We strongly recommend that the Commission removes dependent compartments from the WOAH Aquatic Code to maintain adherence to the established principles of compartmentalization that support disease control, trade resilience, and international standards, and which are already being successfully implemented by Competent Authorities for other WOAH-listed pathogens. See additional rationale in the general comment provided above. **Supporting evidence:** -	

~~The suitability of a dependent *compartment* to achieve the required level of *risk* mitigation should be determined following consideration of the purpose of the *compartment* (refer to Article 4.3.2.), the *commodities* produced (e.g. *aquatic animal products* or *aquatics animals*), and their end uses (e.g. products for human consumption versus *aquatic animals* for stocking in semi-open systems).~~

~~Based on a *risk analysis*, approved by the *Competent Authority*, dependent *compartments* may require specific measures to mitigate the *risk* of *disease* transmission from the environment to the *compartment*. The *risk* mitigation measures should be developed in accordance with Article 4.1.8. and may include the application of specific *biosecurity* measures, post-production testing, auditing within the production cycle, a higher level of internal *targeted surveillance*, external *surveillance* to monitor for change in *disease risk*, and external *disease* control measures to mitigate the *risk* of *disease* transmission into the environment adjacent to the *compartment*.~~

~~Table 1. A summary of the characteristics of independent and dependent compartments~~

~~**Independent**~~	~~**Dependent**~~
~~Only closed systems are a suitable production system type~~	~~Only semi-closed systems are a suitable production system type~~
~~*Biosecurity* across all pathways in accordance with Chapter 4.1.~~	~~*Biosecurity* across most pathways in accordance with Chapter 4.1.~~
~~Disease-free status not dependent on the status of the surrounding waters~~	~~Disease-free status dependent on the status of the surrounding waters~~
~~External *surveillance* generally not required to maintain freedom (but may be useful to inform biosecurity measures)~~	~~Ongoing external *surveillance* may be required to maintain freedom in accordance with Chapter 1.4.~~
~~Suitable for all *commodities* and pathways~~	~~May not meet the required level of *risk* mitigation for all *commodities* and pathways~~

4.3.4._13_Thailand	**Category:** Change **Proposed amended text:** Propose retaining Table 1 Table 1. A summary of the characteristics of independent and dependent compartments **Independent** / **Dependent** Only closed systems are a suitable production system type / Only semi-closed systems are a suitable production system type Biosecurity across all pathways in accordance with Chapter 4.1. / Biosecurity across most pathways in accordance with Chapter 4.1. Disease-free status not dependent on the status of the surrounding waters / Disease-free status dependent on the status of the surrounding waters External surveillance generally not required to maintain freedom (but may be useful to inform biosecurity measures) / Ongoing external surveillance may be required to maintain freedom in accordance with Chapter 1.4. Suitable for all commodities and pathways / May not meet the required level of risk mitigation for all commodities and pathways **Rationale:** This table provides a clear comparison between independent and dependent compartments and will help Members consider which category is most appropriate for their application. **Supporting evidence:** -	Did not agree, the information in this table is contained in the text above. The table was removed to avoid duplication.

Article 4.3.5.

Biosecurity and other risk management~~mitigation~~ measures

4.3.5._1_New Zealand	**Category:** general comment **Proposed amended text:** It would be helpful to structure this section with sub-sections so it clear what applies to an independent compartment (or all compartments), versus any additional factors that need to be considered for a dependent compartment. **Rationale:** **Supporting evidence** n/a	Noted. The introductory text (above) discussing a biosecurity plan in accordance with Chapter 4.1. refers to compartments as a whole as it is applicable to both dependent and independent compartments. The text clarifies in paragraph 4 that a risk analysis is required for a dependent compartment and then provides factors for the risk analysis.

The integrity of a *compartment* relies on *biosecurity* to mitigate the *risk* of introduction of specific *pathogenic agents* into the *compartment* and to maintain its disease-free status. A *biosecurity plan* for the *compartment* should be developed and maintained in accordance with Chapter 4.1.

For *compartments* comprising more than one *aquaculture establishment*, the *biosecurity plan* should provide a common set of management and physical measures to provide a consistent level of *risk management*~~mitigation~~ across all elements of the *compartment*.

The *Competent Authority* should require~~Ensure~~ that all movements of disease-free *aquatic animals* into a *free compartment* originate from a *free country, free zone* or *free compartment,* and in the case of international movements are certified in accordance with Chapter 5.1.

4.3.5._2_Brazil	**Category:** addition **Proposed amended text (or precise suggested deletion):** Biosecurity and other risk management~~mitigation~~ measures The integrity of a *compartment* relies on *biosecurity* to mitigate the *risk* of introduction of specific *pathogenic agents* into the *compartment* and to maintain its disease-free status. A *biosecurity plan* for the *compartment* should be developed and maintained in accordance with Chapter 4.1. For *compartments* comprising more than one *aquaculture establishment*, the *biosecurity plan* should provide a common set of management and physical measures to provide a consistent level of *risk management*~~mitigation~~ across all elements of the *compartment*. The *Competent Authority* should require~~Ensure~~ that all movements of disease-free *aquatic animals* into a *free compartment* originate from a *free country, free zone* or *free compartment* for the diseases associated with the pathogenic agent(s) for which the compartment has been	Agreed, text revised to reflect that the status of the compartment is related to a specific pathogenic agent for which the compartment was established.

	established and in the case of international movements are certified in accordance with Chapter 5.1. **Rationale:** Although it is implicit that the status is related to the specific pathogens, the proposed inclusion makes it clear. **Supporting evidence: N/A**	
4.3.5._3_Canada	**Comment Category:** Deletion **Proposed amended text:** The *Competent Authority* should require~~Ensure~~ that all movements of ~~disease-free~~ *aquatic animals* into a *free compartment* originate from a *free country, free zone* or *free compartment,* and in the case of international movements are certified in accordance with Chapter 5.1. **Rationale:** The current wording could be interpreted as requiring only certain types of animal movements to originate from disease-free sources. However, all movements of aquatic animals should come from countries, zones, or compartments that are free of the disease—not just those explicitly labelled as “disease-free” movements.	Agreed, all movements should come from countries, zone or compartments that are free from disease.
4.3.5._4_Chile	**Categoría**: Editorial **Texto modificado propuesto**: La autoridad competente deberá exigir que todos los movimientos de animales acuáticos hacia un compartimento libre provengan de un país libre, una zona libre o de un compartimento libre de la enfermedad para la cual el compartimento es libre. En el caso de los movimientos internacionales, se certifiquen de conformidad con el Capítulo 5.1. **Justificación**: Mejorar redacción y entendimiento **Evidencia de apoyo**: N/A	Agreed, revisions made to improve clarity.

For dependent *compartments*, the *risk analysis* described in Article 4.3.4.~~4.1.8.~~ should include the assessment of *risks* within the environment surrounding the *compartment* to inform ~~and~~ the development of appropriate *risk management* and *surveillance* measures to mitigate *disease* transmission from the environment. The *risk* mitigation measures should be developed in accordance with Article 4.1.8. and may include the application of specific *biosecurity* measures, a higher level of internal *targeted surveillance* and external *surveillance* to monitor for changes in *disease risk*. ~~the identified *risks*.~~

~~The *Competent Authority* should consider in~~ At a minimum, the following factors should be addressed within the *risk analysis:*

4.3.5._5_Canada	**Comment Category: c**hange **Proposed amended text:** For dependent *compartments*, the *risk analysis* described ~~in~~ below ~~Article~~ 4.3.4.~~4.1.8.~~ should include the assessment of *risks* within the environment surrounding the *compartment* to	Agreed, reference to Article 4.3.4. was not correct, as it refers to text below.

	inform ~~and~~ the development of appropriate *risk management* and *surveillance* measures to mitigate *disease* transmission from the environment. **Rationale:** Article 4.3.4 does not describe the risk assessment. The risk assessment is described in this article, right below this paragraph	
4.3.5._6_China (People's Republic of)	**Category:** change **Proposed amended text :** For dependent *compartments*, the *risk analysis* described in Article 4.3.4.~~4.1.8.~~ should include the assessment of *risks* within the environment surrounding the *compartment*, with the geographic scope of this assessment clearly defined based on the biology of the pathogenic agent(s) (e.g., transmission distance, viability in water) and the hydrological conditions, to inform ~~and~~ the development of appropriate *risk management* and *surveillance* measures to mitigate *disease* transmission from the environment. **Rationale:** The requirement to assess risks 'within the environment' is too broad and impractical. It should be more specific and scientific by emphasizing that the geographical scope of risk assessment must be scientifically grounded and linked to pathogen characteristics and hydrological conditions. **supporting evidence:** not relevant	Agreed, that the requirement to assess risks 'within the environment' was too broad and text was revised to provide more clarity and highlight to use the factors as listed.
4.3.5._7_New Zealand	**Category:** editorial **Proposed amended text:** For dependent *compartments*, the *risk analysis* described in Article 4.3.4.~~4.1.8.~~ should ~~include the assessment of~~ also assess *risks* within the environment surrounding the *compartment* to inform ~~and~~ the development of appropriate *risk management* and *surveillance* measures. These measures are developed to mitigate *disease* transmission from the environment into the dependent *compartment*. The *risk* mitigation measures should be developed in accordance with Article 4.1.8. and may include the application of specific *biosecurity* measures, a higher level of internal *targeted surveillance* and external *surveillance* to monitor for changes in *disease risk*. ~~the identified *risks*.~~ ~~The *Competent Authority* should consider in~~ At a minimum, the following factors should be addressed ~~within~~ in the *risk analysis* for a dependent *compartment*: **Rationale:** This section contained some long sentences, so some edits have been suggested to shorten them and make the section easier to read. It is also unclear whether these are factors that need to be considered for all compartments or just dependent	Agreed to editorial changes to improve clarity in first paragraph. Did not agree editorial changes in second paragraph were needed for clarity.

	compartments – see previous comment about creating sub-sections to clarify, or suggest adding "for a dependent compartment" to end of sentence. **Supporting evidence** n/a	

1) characteristics of the *pathogenic agent(s)*;

4.3.5._8_Peru	**Categoría:** Adición **Texto modificado propuesto:** "Las características de~~l~~ las cepas o de los agentes patógenos" **Justificación:** Con respecto a un agente patógeno, puede haber más de una cepa de un agente patógeno que tenga una morbilidad y mortalidad diferente, por lo tanto, debe añadirse el término "cepa", debido a que puede haber diferentes cepas de virulencia variable en una misma zona geográfica, como por ejemplo en el caso de la infección por *Streptoccocus agalactiae*, que tiene diferentes cepas, y de ellas la cepa ST283 tiene mayor importancia, al ser zoonótica. **Evidencia documentada:** Niu, G., Khattiya, R., Zhang, T., Boonyayatra, S., & Wongsathein, D. (2020). Phenotypic and genotypic characterization of *Streptococcus* spp. isolated from tilapia (*Oreochromis* spp.) cultured in river-based cage and earthen ponds in Northern Thailand. Journal of Fish Diseases, 43(3), 391-398. https://doi.org/10.1111/jfd.13137	Did not agree, the first point is referring to the *pathogenic agent* for which the the compartment has disease freedom and including strain is not necessary at this level.

2) presence~~absence~~ of *susceptible species* and pathways of exposure~~*infection*~~ in the surrounding environment due to geographical location, environmental conditions or the application of *biosecurity* measures. Specific consideration should be given to:

4.3.5._9_New Caledonia	**Proposition ou commentaire :** ~~l'~~absence la présence d'*espèces sensibles* ~~et de voies d'exposition d'*infection*~~ dans le milieu environnant et la probabilité d'exposition à l'agent pathogène ou aux agents pathogènes, en raison de la situation géographique, des conditions environnementales ou de l'application de mesures de *sécurité biologique*. **Exposé des motifs** : Article 4.3.5. 2) : la notion de « présence de voies d'exposition » n'est pas claire, il est proposé de reformuler ainsi : **Justification, s'il y a lieu :**	Agreed to revise to highlight the presence of susceptible species and pathways of exposure.

a) the hydrological conditions in the water body;

b) the geographical location of each *aquaculture establishment* comprising the dependent *compartment* and the nature of the water supply;

c) the health status of other *aquaculture establishments* within the shared water body system;

4.3.5._10_Canada	**Comment Category:** Addition **Proposed amended text:** c) introductions of susceptible species into ~~the health status of~~ other *aquaculture establishments* or processing facilities within the shared water body system; **Rationale:** If nearby aquaculture establishments are not importing/exporting or do not introduce animals from sources outside the shared water body system, the health status should be the same as that of the surrounding waters. It is when introductions happen that the health status becomes more relevant. In addition, if premises are not importing/exporting/moving animals domestically, they may fall under the authority of a different Competent Authority and it would be difficult to know the health status of those establishments.	Agreed, revisions were made to highlight the health status of other aquaculture establishments within the surrounding water body needs to be considered.

d) the location of the *aquaculture establishments* referred to in point (c) or processing facilities and their proximity to the dependent *compartment;*

e) the method of production and the source of the *aquatic animals* used in the *aquaculture establishments* referred to in point (c);

f) the presence and abundance of wild *susceptible species* in the water body ~~and their health status~~;

4.3.5._11_Peru	**Categoría:** Adición **Texto modificado propuesto:** “la presencia y abundancia de especies silvestres ~~susceptibles~~ silvestres o asilvestradas, suceptibles en la masa de agua…” **Justificación:** Se ha hecho uso del término “especie asilvestrada” en especies que ingresan al medio acuático natural desde el medio de producción acuícola y logran establecerse con éxito. **Evidencia documentada:** Wang, J. H., Choi, H. K., Lee, H. J., & Lee, H. G. (2023). On the Species Identification of Two Non-Native Tilapia Species, Including the First Record of a Feral Population of *Oreochromis aureus* (Steindachner, 1864) in South Korea. *Animals*, 13(8), 1351. https://doi.org/10.3390/ani13081351	Did not agree, wild susceptible species would include anything not in an aquaculture establishment and would account for feral aquatic animals.

g) the details of whether the *susceptible species* referred to in point (f) are sedentary or migratory;

4.3.5._12_Australia	**Category:** addition	Agreed, text added on the natural

	Proposed amended text: 2 g) the details of whether the susceptible species referred to in point (f) are sedentary or migratory; and the extent of the populations natural distribution range or migratory movement patterns **Rationale**: Suggest addition of this detail as it may impact the extent of compartment boundaries or feasibility of implementing the compartment. **Supporting evidence:** n/a	distribution and migratory patterns of the susceptible species.

h) the exclusion of the wild *aquatic animals* referred to in point (f) from entering the *compartment*;

i) the general *biosecurity* measures applied in *aquaculture establishments* and processing facilities in the shared water body;

4.3.5._13_China (People's Republic of)	**Category:** change **Proposed amended text :** h) ~~the exclusion of the wild *aquatic animals* referred to in point (f) from entering the *compartment*~~ mitigate the risk of exposure from wild aquatic animals entering the compartment or its water supply; **Rationale:** Changing from "exclusion" to "risk reduction measures" is more consistent with the reality of reliance partitions and is consistent with other risk management measures. **supporting evidence:** not relevant	Agreed, revised 'exclusion' to 'ability to exclude' to be consistent with risk management measures.

4.3.5._14_New Caledonia	**Proposition ou commentaire :** Commentaire editorial i) les mesures générales de *sécurité biologique* appliquées dans les *établissements d'aquaculture* et les installations de transformation situé~~es~~ dans l'étendue d'eau partagée ; **Exposé des motifs** : Article 4.3.5. 2) i) : faute d'accord : "situé" s'accorde avec les "établissements d'aquaculture et les installations de transformation" **Justification, s'il y a lieu :** /	Agreed, translation change to French version only.
4.3.5._15_New Zealand	**Category:** editorial **Proposed amended text:** a) the hydrological conditions in the water body; b) the geographical location of each *aquaculture establishment* comprising the dependent *compartment* and the nature of the water supply; c) ~~the health status of other *aquaculture establishments* within the shared water body system~~ the existence of other *aquaculture establishments* containing *susceptible species* within the shared water body system and their source of *aquatic animals*, health status and method of production; d) the ~~location of the *aquaculture establishments* referred to in point (c) or~~ proximity of processing facilities ~~and their proximity~~ to the dependent *compartment;* e) ~~the method of production and the used in the *aquaculture establishments* referred to in point (c);~~ e) the presence and abundance of wild *susceptible species* in the water body ~~and their health status~~;	Agreed, revisions made to reflect comment and improve clarity.

	f) the details of whether the *susceptible species* referred to in point (f) are sedentary or migratory; g) the exclusion of the wild *aquatic animals* referred to in point (f) from entering the *compartment*; h) the general *biosecurity* measures applied in *aquaculture establishments* and processing facilities in the shared water body; **Rationale:** - Edited to summarise all points relating to "other aquaculture establishments" within the shared water body of the compartment, including specifying that it applies to those that contain *susceptible species*. - Updated bullet list **Supporting evidence** n/a	

3) presence~~absence~~ of *infection* in any nearby populations of *susceptible species* demonstrated by appropriate external *surveillance*;

4) additional internal *surveillance* (i.e. in the *aquaculture establishment(s)* that comprise the *compartment*).

For some semi-closed *aquaculture establishments*, it may not be possible to mitigate identified *risks* from the surrounding environment (e.g. presence of *disease* in adjacent wild populations of *susceptible species*) and the *aquaculture establishment* would not be eligible to be recognised as a dependent *compartment*.

4.3.5._16_Chile	**Categoría**: Eliminar **Texto modificado propuesto**: ~~En el caso de algunos *establecimientos de acuicultura* semi-cerrados, puede que no sea posible mitigar los *riesgos* identificados en el entorno circundante (por ejemplo, presencia de *enfermedad* en poblaciones silvestres de *especies susceptibles* que vivan a proximidad) y que el *establecimiento de acuicultura* pueda no ser reconocido como un *compartimento* dependiente.~~ **Justificación**: es redundante esta frase y se presta para confusiones dado que no aplicaría este capítulo **Evidencia de apoyo**: N/A	Did not agree to delete text, the paragraph on semi-closed aquaculture establishments provides clarity to understand when a dependent compartment may or may not apply.
4.3.5._17_New Zealand	**Category:** editorial **Proposed amended text:** 4) additional internal targeted *surveillance* (i.e. in the *aquaculture establishment(s)* that comprise the *compartment*). ~~For some semi-closed *aquaculture establishments,*~~ Where it ~~may~~ is not ~~be~~ possible to mitigate ~~identified~~ the *risks*	Agreed, to add 'targeted' surveillance to point 4 for consistency. Agreed to revisions of final paragraph to

	identified from the surrounding environment surrounding a semi-closed *aquaculture establishment* (e.g. presence of the specified *listed disease* in adjacent wild populations of *susceptible species*), and the *aquaculture establishment* would not be eligible to be recognised as a dependent *compartment*. **Rationale:** - Addition of "targeted' before surveillance, for consistency with other sections where internal surveillance is mentioned. - Edits suggested to improve clarity and readability. **Supporting evidence** n/a	improve clarity and readability.
4.3.5._18_United States	**Category:** deletion **Proposed amended texts (or precise suggested deletion):** "~~For dependent *compartments*, the *risk analysis* described in Article 4.3.4.4.1.8. should include the assessment of *risks* within the environment surrounding the *compartment* to inform and the development of appropriate *risk management* and *surveillance* measures to mitigate *disease* transmission from the environment. The *risk* mitigation measures should be developed in accordance with Article 4.1.8. and may include the application of specific *biosecurity* measures, a higher level of internal *targeted surveillance* and external *surveillance* to monitor for changes in *disease risk*. the identified *risks*.~~" "~~For some semi-closed *aquaculture establishments*, it may not be possible to mitigate identified *risks* from the surrounding environment (e.g. presence of *disease* in adjacent wild populations of *susceptible species*) and the *aquaculture establishment* would not be eligible to be recognised as a dependent *compartment*.~~" **Rationale:** We strongly recommend that the Commission removes dependent compartments from the WOAH Aquatic Code to maintain adherence to the established principles of compartmentalization that support disease control, trade resilience, and international standards, and which are already being successfully implemented by Competent Authorities for other WOAH-listed pathogens. See additional rationale in the general comment provided above.	Did not agree, see response to comment 4.3._8.

Article 4.3.6.

4.3.6._1_Australia	**Category:** General comment **Proposed amended text:** n/a **Rationale**: In article 4.3.6, Australia suggests more consideration be given to the qualifying requirements or suitable conditions for using passive surveillance in either internal or external surveillance. Passive surveillance may be suitable for external surveillance in some instances but not others. For example, some wild catch industry sectors that have a high degree of human interaction with- and observation of - live animals during the harvesting and trade processes (e.g. live abalone or rock lobster fisheries) present an opportunity to potentially detect disease incidents in external wild populations via passive surveillance when clinical signs are evident. However, in other instances, disease outbreaks are highly unlikely to be detected in wild susceptible species populations by passive surveillance due to: • cannibalism and scavenging of sick or dead animals • lack of clinical signs • lack of close observation (e.g. wild pelagic fisheries or susceptible species that are not subject to fishing or harvesting or that are located in remote places). Only targeted surveillance is likely to detect disease in most circumstances of external surveillance. Highlighting the specific requirements for using passive surveillance by referring to Article 1.4.8 may clarify this issue. (See specific comment below). Conversely, passive surveillance may be highly valuable for internal surveillance and has been shown to be highly sensitive in aquaculture systems with fish health monitoring programs in place (Hall et al. *In preparation*). Passive surveillance when used in conjunction with active targeted surveillance as part of internal surveillance measures can increase the overall sensitivity of the complete surveillance system. Passive surveillance is not mentioned directly for internal surveillance in article 4.3.6, but is mentioned in the referenced article 1.4.15. Australia suggests specifying passive or targeted surveillance directly in the text regarding internal surveillance for consistency (See specific comment below). **Supporting evidence:** n/a	Passive surveillance is required for a compartment as a component of the early detection system as described in Chapter 1.4. The early detection system is part of the basic biosecurity conditions which prevent the introduction and spread of a disease, and the detection of that disease should it occur. To make a self-declaration of freedom from a specific disease for a compartment the basic biosecurity conditions need to be met for all WOAH listed diseases. Targeted surveillance is required to show freedom from disease at a population level in a compartment. Targeted surveillance for the compartment is separate from external surveillance where the intention is to inform risk analysis for the compartment.

Surveillance requirements to demonstrate and maintain freedom

For recognition of a *free compartment*, a *self-declaration of freedom from disease* should be made which complies with the requirements of Article 1.4.4. The *surveillance* requirements to make a *self-declaration of freedom from disease* for a *compartment*, and to maintain a *free compartment*, should comply with Chapter 1.4.

Basic biosecurity conditions for a *compartment* must be in place and continuously met prior to the commencement of *targeted surveillance* to demonstrate freedom. The relevant disease-specific chapters provide the required periods that *basic biosecurity conditions* must be in place prior to commencement of *targeted surveillance*, and the period that *targeted surveillance* should be conducted prior to making a *self-declaration of freedom from disease* as well as the requirements to maintain that freedom in accordance with Article 1.4.15.

~~*Surveillance* requirements should be developed in accordance with factors as described in Article 4.3.5.~~

If there is an increased *risk* of exposure to the *disease* from which the *compartment* has been defined, the sensitivity of the internal and external *surveillance* system should be reviewed, documented and, where necessary, increased. At the same time, the *biosecurity plan* should be reviewed in accordance with Article 4.1.9 and revised if necessary.

4.3.6._2_New Zealand	**Category:** editorial **Proposed amended text:** If there is an increased *risk* of exposure to the *pathogenic agents* and/or *disease* ~~from~~ for which the *compartment* has been defined, the sensitivity of the internal and external *surveillance* system should be reviewed, documented and, where necessary, increased. At the same time, the *biosecurity plan* should be reviewed in accordance with Article 4.1.9 and revised if necessary. **Rationale:** - Addition of word "targeted' for consistency with other sections where internal surveillance is mentioned. - Edits suggested to improve clarity and readability. **Supporting evidence** n/a	Agreed to add 'pathogenic agents' for clarity, other revisions not required for readability.
4.3.6._3_Peru	**Categoría:** Adición **Texto modificado propuesto:** "Si existe un mayor riesgo de exposición a la enfermedad para la que se ha definido el compartimento, la ~~sensibilidad~~ **eficacia** del sistema de vigilancia interna y externa deberá revisarse, documentarse y, si es preciso, intensificarse". **Justificación:** El término "sensibilidad" es un término referido a la sensibilidad de la prueba diagnóstica. "Sensibilidad" en el glosario del código acuático de la OMSA designa un porcentaje de resultados positivos verdaderos que arroja una prueba de diagnóstico, o sea, número de resultados positivos verdaderos dividido por el número total de resultados positivos verdaderos más negativos falsos. Por	Did not agree, sensitivity is the appropriate word when referencing the internal and external surveillance system. The sensitivity of surveillance is referenced throughout Chapter 1.4. 'Aquatic Animal Disease Surveillance'.

	lo tanto, se sugiere eliminar el término “sensibilidad”. Por otro lado “eficacia” sí es un término mencionado en el código acuático de la OMSA para referirse a un sistema de vigilancia. **Evidencia documentada:** 1. “Artículo.1.4.16. del capítulo 1.4. “Vigilancia de las enfermedades de los animales acuáticos” del código acuático de la OMSA, el cual indica “Una de las principales ventajas de este método es que permite mejorar la **eficacia** de la **vigilancia** para demostrar la ausencia de **enfermedad** en comparación con los métodos de muestreo aleatorio”. 2. Glosario del código acuático de la OMSA.	

1. Internal surveillance

   Internal *surveillance* (i.e. offor populations of *susceptible species* within a *compartment*) is required to make a *self-declaration of freedom from disease* for both independent and dependent *compartments*. The *surveillance* requirements to maintain freedom are described in the relevant disease specific chapters and Article 1.4.15.

4.3.6._4_Australia	**Category:** addition **Proposed amended text:** The surveillance requirements (passive or targeted) to maintain freedom are described in the relevant disease specific chapters and Article 1.4.15. **Rationale**: The possibility of using passive surveillance for internal surveillance is not mentioned directly in Article 4.3.6, however, the possibility of using passive surveillance for external surveillance under certain circumstances is mentioned. Added the suggested text here for consistency. **Supporting evidence:** n/a	Did not agree, as per Chapter 1.4. to make a self-declaration of freedom from disease for a compartment targeted surveillance is required for internal surveillance. Passive surveillance is required as part of an early detection system contributing to the basic biosecurity conditions. Chapter 1.4. was reviewed and revised to clarify the use of surveillance regarding compartments (see item 6.2. and Annex 12).
4.3.6._5_New Zealand	**Category:** editorial **Proposed amended text:** Internal targeted *surveillance* (i.e. offor populations of *susceptible species* within a *compartment*) is required to make a *self-declaration of freedom from disease* for both independent and dependent *compartments*. The *surveillance* requirements to maintain freedom are described in the relevant disease specific chapters and Article 1.4.15. **Rationale:**	Did not agree, as described in Chapter 1.4., targeted surveillance is required to make a self-declaration of freedom from disease for a compartment and it is not necessary to include in this point.

	- Addition of word “targeted’ for consistency with other sections where internal surveillance is mentioned. **Supporting evidence** n/a	

2. External surveillance

External *surveillance* (i.e. offor populations of *susceptible species* in the environment outside a *compartment*) can be used to identify a significant change in the level of exposure for the identified pathways for *disease* introduction into the *compartment*. External *surveillance* may be passive or targeted based on the specific situation in accordance with the relevant disease-specific chapters and Chapter 1.4.

4.3.6._6_Australia	**Category:** addition **Proposed amended text:** External surveillance may be passive or targeted based depending on the specific situation in accordance with the relevant disease-specific chapters and Chapter 1.4. See Article 1.4.8. for the requirements for passive surveillance. **Rationale**: Passive surveillance is not suitable for all external surveillance situations. Added reference to Article 1.4.8 for clarification regarding the requirements for passive surveillance. **Supporting evidence:** n/a	Did not agree see response to comment 4.3.6._1.
4.3.6._7_Canada	**Comment Category: change** **Proposed amended text:**. External *surveillance* (i.e. offor populations of *susceptible species* in the environment outside a *compartment*) can be used to identify a significant change in the level of exposure for the identified pathways for *disease* introduction into the *compartment*. External *surveillance* may be passive and/or targeted based on the specific situation in accordance with the relevant disease-specific chapters and Chapter 1.4. **Rationale:** the inclusion of and/or allows flexibility dependant on the specific situation.	Agreed, although or could be interpreted to allow for both added ‘and/’ to highlight flexibility.

For dependent *compartments*, externalExternal *surveillance* is required for dependent *compartments* if populations of *susceptible species* are present in the environment surrounding the *compartment*. The *surveillance* should be developed taking into account the factors described Article 4.3.5. The area for which external *surveillance* is required must be defined and should take into account any *surveillance* which is carried out on adjacent *aquaculture establishments* keeping *susceptible species*. For a dependent *compartment* occurring within a *free zone* or *free country*, the *surveillance* to establish and maintain a *free zone* or *free country* constitutes the external *surveillance* for the *compartment*.

4.3.6._8_Brazil	**Category:** addition	Did not agree, it is implicit in the sentence that the

	Proposed amended text (or precise suggested deletion): For dependent *compartments*, external~~External~~ *surveillance* is required ~~for dependent *compartments* if~~ populations of *susceptible species* are present in the environment surrounding the *compartment*. The *surveillance* should be developed taking into account the factors described Article 4.3.5. The area for which external *surveillance* is required must be defined and should take into account any *surveillance* which is carried out on adjacent *aquaculture establishments* keeping *susceptible species*. For a dependent *compartment* occurring within a *free zone* or *free country*, for the diseases associated with the pathogenic agent(s) for which the compartment has been established, the *surveillance* to establish and maintain a *free zone* or *free country* constitutes the external *surveillance* for the *compartment*. **Rationale:** Although it is implicit that the status is related to the specific pathogens, the proposed inclusion makes it clear. **Supporting evidence: N/A**	surveillance referenced is for the diseases for which the compartment was established.
4.3.6._9_Brazil	**Category:** addition **Proposed amended text (or precise suggested deletion):** For dependent *compartments*, external~~External~~ *surveillance* is required ~~for dependent *compartments* if~~ populations of *susceptible species* are present in the environment surrounding the *compartment*. The *surveillance* should be developed taking into account the factors described Article 4.3.5. The area for which external *surveillance* is required must be defined and should take into account any *surveillance* which is carried out on adjacent *aquaculture establishments* keeping *susceptible species*. For a dependent *compartment* occurring within a *free zone* or *free country*, ,the *surveillance* to establish and maintain a *free zone* or *free country* constitutes the external *surveillance* for the *compartment*. In the event that a dependent compartment is located within a zone or country that has lost its disease-free status for the diseases associated with the pathogenic agent(s) for which the compartment was established, the operator shall assess the increased risk of disease exposure and implement measures in accordance with this chapter. The Competent Authority shall evaluate whether this situation is classified as a suspected occurrence of the disease for which the compartment was established and, if necessary, shall act in compliance with the provisions of Article 4.3.12.	Agreed, the addition provides guidance on actions to be taken when the pathogenic agent is detected in the surrounding water body for a compartment whose status is conditional on its location. Revised text was added to reflect this comment.

	Rationale: The chapter does not provide explicit guidance on the prescribed actions for dependent compartments when pathogens are detected within the same epidemiological area, considering that such compartments are epidemiologically linked to the surrounding environment. Nevertheless, these compartments are required to implement and maintain basic biosecurity and management measures to safeguard a population with a distinct health status. It is recommended that confirmation of disease within the associated environment shall result in an immediate elevation of the assessed risk level by the operator. Furthermore, the Competent Authority should consider the designation of dependent compartments as suspected of disease occurrence. This provision shall be clearly and explicitly stated within the text of the chapter to ensure regulatory clarity and consistency. **Supporting evidence: N/A**	
4.3.6._10_Chinese Taipei	**Category:** Change **Proposed amended text:** ~~The s~~Surveillance should be developed taking into account the factors described in Article 4.3.5. The area for which external *surveillance* is required must be clearly defined and should take into account ~~any surveillance which is carried out on adjacent aquaculture establishments keeping susceptible species.~~ Appropriate surveillance should be determined on the basis of risk assessment, and surveillance activities on adjacent aquaculture establishments housing susceptible species may be considered. For a dependent compartment occurring within a free zone or a free country, the surveillance used to establish and maintain the free zone or free country shall constitute the external surveillance for the compartment. **Rationale:** The scale of external surveillance should be determined based on risk assessment. **Supporting evidence:** Not relevant.	Agreed, text was added that surveillance should 'be at a level sufficient to inform the risk analysis described in Article 4.3.4.'.
4.3.6._11_United States	**Category:** deletion **Proposed amended texts (or precise suggested deletion):** "Internal *surveillance* (i.e. off~~for~~ populations of *susceptible species* within a *compartment*) is required to make a *self-declaration of freedom from disease* ~~for both independent and dependent *compartments*~~. The *surveillance* requirements to maintain freedom are described in the relevant disease specific chapters and Article 1.4.15." "~~For dependent *compartments*, external~~External ~~*surveillance* is required for dependent *compartments* if~~	Did not agree, see response to comment 4.3._8.

	populations of *susceptible species* are present in the environment surrounding the *compartment*. The *surveillance* should be developed taking into account the factors described Article 4.3.5. The area for which external *surveillance* is required must be defined and should take into account any *surveillance* which is carried out on adjacent *aquaculture establishments* keeping *susceptible species*. For a dependent *compartment* occurring within a *free zone* or *free country*, the *surveillance* to establish and maintain a *free zone* or *free country* constitutes the external *surveillance* for the *compartment*." **Rationale:** We strongly recommend that the Commission removes dependent compartments from the WOAH Aquatic Code to maintain adherence to the established principles of compartmentalization that support disease control, trade resilience, and international standards, and which are already being successfully implemented by Competent Authorities for other WOAH-listed pathogens. See additional rationale in the general comment provided above. **Supporting evidence:** -	

Article 4.3.7.

Laboratory testing

Laboratories providing testing services for a *compartment* should be approved by the relevant *Competent Authority*. In providing approval, the *Competent Authority* should ensure that the laboratory:

4.3.7._1_Chile	**Categoría**: Cambio **Texto modificado propuesto**: Los laboratorios que realicen pruebas para un *compartimento* deberán ser aprobados por la *autoridad competente* pertinente. ~~Tras~~ Para su aprobación, la *autoridad competente* deberá asegurarse de que el laboratorio: **Justificación:** La autoridad competente debe asegurarse que cumpla todos los requisitos señalados (numeral 1 al 5) antes de su aprobación. **Evidencia de apoyo:** N/A	Agreed, translation change in Spanish version only.

1) has a quality management system that meets requirements of Chapter 1.1.1. of the *Aquatic Manual*, or can demonstrate quality through another means in accordance with Chapter 3.1.;

2) ~~is required to~~ conducts testing in accordance with the recommendations of the *Aquatic Manual*;

3) can confirm or exclude cases of *infection*~~*disease*~~ as described in Article 1.4.18.;

4) is independent from management and ownership structures of the *compartment*;

5) has a legal obligation to report positive test results to the *Competent Authority* in accordance with the requirements of *basic biosecurity conditions* specified in Article 1.4.6.

Article 4.3.8.

Traceability

Traceability systems should apply throughout the supply chain and are required to reliably differentiate *commodities* that originate from a *free compartment* from those that originate from outside a *free compartment*. The traceability system should:

4.3.8._1_China (People's Republic of)	**Category:** change **Proposed amended text :** Traceability systems should apply throughout the supply chain, including simplified digital or paper-based systems suitable for small-scale operations, and are required to reliably differentiate *commodities* that originate from a *free compartment* from those that originate from outside a *free compartment*. The traceability system should: **Rationale:** There are a large number of family farms and small farms, and it is difficult to promote the complex traceability system. It is necessary to supplement the simple traceability method (digital or paper) to adapt to the practical ability of small and medium-sized operators. The FAO's Practical Guide to Traceability in Aquaculture states that “Traceability systems should be scalable, including simplified options for smallholders to ensure inclusivity.” **supporting evidence:** not relevant	Did not agree to specify that the traceability system should include simplified systems. As written the traceability system can be scaled to fit the production systems and it does not need to be specified in the text.

1) be appropriate for the nature of the supply chains of the *aquatic animal* species and for application to individual or groups of *aquatic animals* or *aquatic animal products*, as necessary;

4.3.8._2_Norway	**Category**: deletion **Proposed amended text**: ~~1)~~ be appropriate for the nature of the supply chains of the *aquatic animal* species and for application to individual or groups of *aquatic animals* ~~or *aquatic animal products*, as necessary;~~ **Rationale:** Due to the WOAH glossary definition of "aquatic animal products" and "commodity", Norway does not consider it appropriate to refer to aquatic animal products in this context. We therefore encourage the Commission to reconsider its previous decision. The WOAH glossary contains the following definitions: "**aquatic animal products** means non-viable aquatic animals, parts of aquatic animals, or manufactured goods containing any material derived from aquatic animals that are intended for sale or trade." "**commodity** means aquatic animals, aquatic animal products, biological products and pathological material." Based on these definitions, it is our understanding that both "aquatic animal products" and "commodity" is referring to aquatic animal products, including products intended for end consumers. This is because "aquatic animal products" are referring both to manufactured goods and all products intended for sale or trade. The explicit division of "trade" and "sale" indicates the following understanding: "trade" is to be understood as buying and selling at business-to-business level, because "trade" involves both buying and selling, while "sale" is to be understood as selling a product to end consumers. The last part of the sentence in point 1 should therefore be deleted. **Supporting evidence, if relevant:** not relevant	Did not agree to remove 'aquatic animal products' as the Competent Authority needs to be able to ensure the provenance of both animals and products from the compartment. The intention of the section is not to indicate that tracing is required to the consumer level. Rather tracing of products may be required until the point of export. In the first paragraph, 'should apply throughout the supply chain and' was deleted as it is inherent in the text and to avoid confusion on tracing needing to occur to consumer level.

2) record all *aquatic animal* movements into and out of the *compartment* including origin and destination. ~~ensure that all movements of disease-free *aquatic animals* into a *free compartment* originate from a *free country, free zone* or *free compartment,* and in the case of international movements are certified in accordance with Chapter 5.1.~~;

3) be reflected in the *biosecurity plan* that is developed in accordance with Article 4.3.5. and which provides appropriate *risk management*;

4) comprise record keeping requirements in accordance with Article 4.3.9.;

5) be approved by the *Competent Authority* in accordance with Article 4.3.10.

Article 4.3.9.

Record keeping

A system of record keeping by the operator of a *compartment* should provide clear evidence that the *biosecurity*, *surveillance*, traceability and management practices that form the basis of a *self-declaration of freedom from disease* are effectively and continuously applied.

Records should be maintained consistently by the operator of the *free compartment* and be accessible on request for the purposes of an audit or in response to queries from the *Competent Authority* of an *importing country*. The record keeping system should:

4.3.9._1_New Caledonia	**Proposition ou commentaire :** Commentaire editorial “Un système de tenue de registres par l'exploitant de chaque établissement d’aquaculture compris dans 'un *compartiment* indemne doit proposer des éléments démontrant clairement que les pratiques en matière de *sécurité biologique*, de *surveillance*, de traçabilité et de gestion qui constituent la base d'une *auto-déclaration d'absence de maladie* sont appliquées de manière efficace et continue. Les registres doivent être tenus de manière cohérente par l'exploitant ~~du compartiment indemne~~ et être accessibles sur demande à des fins d'audit ou pour répondre aux demandes de l'*Autorité compétente* d'un *pays importateur*.” **Exposé des motifs** : Article 4.3.9. : comme proposé précédemment, il est proposé de remplacer les termes “exploitant d’un compartiment” par “exploitant de chaque établissement d'aquaculture compris dans un compartiment”. De même, il paraît nécessaire de préciser qu’il s’agit d’un “compartiment indemne” dans le premier alinéa **Justification, s’il y a lieu : /**	Did not agree, see response to comment 4.3.2._7.

1) substantiate that the *compartment*’s *biosecurity plan* is maintained in accordance with Chapter 4.1., including the maintenance of records associated with all relevant pathways described in Article 4.1.7;

2) substantiate that the *surveillance* required to declare and maintain *free compartment* status has been conducted in accordance with Chapter 1.4. and the provisions of relevant disease-specific chapters;

3) document any changes to *biosecurity*, *surveillance*, traceability or management practices, the rationale for the changes and substantiation that they continue to meet *risk management* requirements;

4) in addition to the points above, maintain any external reports, certificates or approvals associated with the requirements of this chapter, including but not limited to audit reports, laboratory reports, health certificates, vaccination records and health investigations;

4.3.9._2_Brazil	**Category:** addition **Proposed amended text (or precise suggested deletion):**	Agreed, to add ‘veterinary treatments’ to point 4.

	4) in addition to the points above, maintain any external reports, certificates or approvals associated with the requirements of this chapter, including but not limited to audit reports, laboratory reports, health certificates, vaccination records, veterinary treatments and health investigations; **Rationale:** Since the vaccine records were included in the examples, we also propose the explicit inclusion of veterinary treatments as relevant documents to monitor the health status of animals. **Supporting evidence: N/A**	

5) maintain records for sufficient period of time to inform tracing, recall or emergency response at any point in the supply chain if a *disease* were detected within the *compartment* or in *commodities* originating from the *compartment*. The required period should be met requirements for *surveillance*, the *biosecurity plan*, auditing, and traceability. It may vary depending on the *disease, aquatic animal* species and *commodity* types produced and the duration of production cycles.

4.3.9._3_Norway	**Category**: deletion **Proposed amended text**: 5) maintain records for sufficient period of time to inform tracing, recall or emergency response ~~at any point in the supply chain~~ if a *disease* were detected within the *compartment* or in *commodities* originating from the *compartment*. The required period should be meet requirements for *surveillance*, the *biosecurity plan*, auditing, and traceability. It may vary depending on the *disease, aquatic animal* species and *commodity* types produced and the duration of production cycles. **Rationale:** As the text appears now, in combination with Article 4.3.10, the legal obligation to facilitate a traceability system for aquatic animals, parts of aquatic animals, or manufactured goods containing any material derived from aquatic animals, all the way from the compartment to the end user, is put upon the competent authority and operators of the exporting country. Our position is that traceability systems that covers the tracking of a product back from the end user to the seller of the same product, to be understood as the business-to-consumer transaction, is to be decided and implemented by the national importing authorities, and not the competent authority and operators of the exporting country. The proposed amended text in this article, in combination with the proposed amended text in Article 4.3.8 point 1), transfers the legal obligation for traceability for business-to-consumer transactions to the importing country.	Agreed, the intention is that records should allow tracing until point of sale or export, thus deleted 'at any point in the supply chain'.

	We encourage the Commission to also ensure that this principle is clearly specified in the revised section 5 'Trade measures, importation/exportation procedures and health certification'. **Supporting evidence, if relevant:** not relevant	

Article 4.3.10.

Official oversight

A *Competent Authority* must have the authority to approve the operation of the *aquaculture establishment(s)* that comprise~~compromise~~ the *compartment*. A *Competent Authority* must also have the authority to make a *self-declaration of freedom from disease* as described in Chapter 1.4., as well as grant, suspend and revoke the status of a *compartment*. It should supervise compliance with all of the requirements critical to the maintenance of the *compartment* status described in this chapter and ensure that all relevant information (as described in Article 4.3.9.) is readily accessible to *importing countries*. The *Competent Authority* should ensure appropriate auditing of the *compartment* is completed by trained officials or ~~accredited~~ third party auditors approved by *Competent Authorities*.

The *Veterinary Authority* should ensure that any changes to the health status of the *compartment* should be notified to the *Veterinary Authority* of *importing countries*.

4.3.10._1_Canada	**Comment Category: change** **Proposed amended text:** The ~~*Veterinary Authority*~~ *Competent Authority* should ensure that any changes to the health status of the *compartment* should be notified to the ~~*Veterinary Authority*~~ *Competent Authority* of *importing countries*. **Rationale:** The Competent Authority responsible for the implementation of the compartment standards should be responsible for notifying trade partners. Article 4.3.12 also uses Competent Authority as the responsible authority for notification. If there was a notification required to WOAH regarding the change in health status, then the Veterinary Authority would be involved.	Agreed, Competent Authority responsible for the oversight of the compartment will be responsible for notifying trade partners. It is noted that the Veterinary Authority as a Competent Authority may hold this responsibility.
4.3.10._2_Chile	**Categoría**: Adición **Texto modificado propuesto:** La autoridad competente debe informar cualquier modificación de los requisitos críticos identificados en el compartimento, y que permitió su aprobación inicial. Además, la autoridad competente deberá informar a los países importadores la suspensión o revocación de un compartimento. **Justificación:** La autoridad competente debe informar no solo cualquier cambio en el estatus sanitario si no también cualquier cambio en el compartimento que	Agreed, that the Competent Authority must report changes in the biosecurity and risk management measures that allowed the compartment to be formed. Text added to reflect this comment.

	eventualmente genere puntos críticos no definidos anteriormente en el análisis de riesgo, por ejemplo, una nueva infraestructura. Además, la autoridad competente deberá informar a los países importadores la suspensión o revocación de un compartimento **Evidencia de apoyo**: N/A	
4.3.10._3_Chile	**Categoría:** Eliminación **Texto modificado propuesto**: La *autoridad competente* se asegurará de que funcionarios formados o auditores externos aprobados por las *autoridades competentes* ~~acreditados~~ efectúen una auditoría ~~adecuada~~ del *compartimento*. **Justificación:** Se da por hecho que la auditoria debe ser adecuada, por lo que es redundante, además ya se entiende a que se refiere adecuado con el párrafo anterior. **Evidencia de apoyo:** N/A	Agreed, removed the word 'appropriate' as it is inherent in the text that the audit must be appropriate.
4.3.10._4_China (People's Republic of)	**Category:** change **Proposed amended text :** According to Chapter 1.1, the ~~The~~ *Veterinary Authority* should ensure that any changes to the health status of the compartment should be notified to the *Veterinary Authority* of *importing countries*. **Rationale:** The notification responsibility is stipulated in Chapter 1.1, and since the original text does not refer to Chapter 1.1, the statement here may seem to lack a basis. **supporting evidence:** not relevant	Did not agree, this detail is included in Article 4.3.12.
4.3.10._5_New Zealand	**Category:** editorial **Proposed amended text:** The *Competent Authority* should ensure that any changes to the health status of the *compartment* are notified to the *Veterinary Authority*, so that the ~~The~~ *Veterinary Authority* ~~should ensure that any changes to the health status of the *compartment* should be notified to~~ can notify the *Veterinary Authority* of *importing countries*.	Text changed to say 'Competent Authority' rather than 'Veterinary Authority' to simplify text on how changes in status should be communicated.

	Rationale: To ensure there is clear communication between the Competent Authority and the Veterinary Authority if there are changes to the health status of the compartment. **Supporting evidence** n/a	

Article 4.3.11.

Quality of aquatic animal health services

The quality of *Aquatic Animal Health Services* relevant to the self-declaration of *compartment* freedom should be documented by the *Competent Authority*, including how they meet the requirements of Chapter 3.1.

Article 4.3.12.

Notification and response measures

In the event of suspicion ~~of occurrence~~ of the *disease* for which the *compartment* was defined, the operator of the *compartment* should immediately notify the *Competent Authority*. The *Competent Authority* should then determine whether the disease-free status of the *compartment* should be immediately suspended and *importing countries* should be notified following the provisions of Chapter 1.1., while the occurrence of the disease is confirmed or ruled-out.

In the event of confirmation of the *disease* for which the *compartment* was defined, the free status should immediately be suspended.

4.3.12._1_Canada	**Comment Category: change** **Proposed amended text:** In the event of confirmation of the *disease* for which the *compartment* was defined within a compartment, the free status should immediately be suspended. **Rationale:** the location of the disease incursion should be included for clarity. If there is a detection within a compartment, independent or dependent, the free status should always be suspended. However, Article 4.3.4 indicates "The possibility of achieving such disease-specific separation will be determined by the *Competent Authority*, based on *risk analysis*." Based on the specific situation and risk assessment completed by the Competent Authority, there may be a functional disease separation between the dependent compartment and the surrounding waters. Decisions regarding suspension of status for dependent compartments, as a result of detections outside of the Compartment, should be left to the Competent Authority, based on the specific disease investigation and response activities.	Text was revised to reflect where that the disease for which the compartment was established is confirmed in the compartment.

	If the dependent compartment was free based on a country freedom declaration, the detection was on one side of the country and the dependent compartment on the other (potentially 1000s of kms away), the detection should not automatically result in suspension of the dependent compartment status. The competent Authority should be able to rezone (if necessary) and the dependent compartment should be unaffected.	
4.3.12._2_Chile	**Categoría:** adición **Texto modificado propuesto:** En caso de sospecha ~~de aparición~~ de la *enfermedad* para la que se definió el *compartimento*, el operador del *compartimento o el laboratorio que realice el diagnóstico* deberá notificar inmediatamente a la *autoridad competente*. **Justificación:** Cualquier operador o laboratorio que sospeche o diagnostique debe informar inmediatamente a la autoridad competente. **Evidencia de apoyo:** N/A	Did not agree, in the case of suspicion of disease in a compartment the operator has the responsibility to notify the Competent Authority.
4.3.12._3_New Caledonia	**Proposition ou commentaire : Commentaire éditorial** “En cas de suspicion ~~d'apparition~~ de la *maladie* pour laquelle le *compartiment* a été déclaré indemne ~~défini~~, l'exploitant de l’établissement d’aquaculture concerné compris dans ce ~~du~~ *compartiment* doit adresser immédiatement une notification à l'*Autorité compétente*.” **Exposé des motifs** : Article 4.3.12. premier alinéa : il semble préférable de remplacer “défini” par “déclaré indemne” dans la phrase ci-dessous pour clarifier le sens de la phrase et comme précédemment, il est proposé de remplacer “exploitant du compartiment” par “exploitant de l’établissement d’aquaculture concerné compris dans ce compartiment” **Justification, s’il y a lieu :**	Did not agree, ‘defined’ is used throughout the text and links to principle 2 in Article 4.3.2. where it provides examples for what factors need to be considered when defining a compartment.
4.3.12._4_New Zealand	**Category:** editorial **Proposed amended text:** In the event of suspicion ~~of occurrence~~ of the *disease* for which the *compartment* was defined, the operator of the *compartment* should immediately notify the *Competent Authority*. The *Competent Authority* should then determine whether the disease-free status of the *compartment* should be immediately suspended and notify the *Veterinary Authority*, and the Veterinary	Did not agree, the Veterinary Authority is a Competent Authority and as there may be differences between countries this notification is covered by Competent Authority.

	Authority of *importing countries* ~~should be~~ notified following the provisions of Chapter 1.1., while the occurrence of the disease is confirmed or ruled-out. In the event of confirmation of the *disease* for which the *compartment* was defined, the free status should immediately be suspended. **Rationale:** To ensure there is clear communication between the Competent Authority and the Veterinary Authority if there are changes to the health status of the compartment, and clarify the roles of the Veterinary and Competent Authorities. **Supporting evidence** n/a	

The operator of a *compartment* should report any event which could lead to a breach of *biosecurity* measures to the *Competent Authority*. ~~In the event of detection of any *disease* which may indicate a breach of *biosecurity* measures, the management of the *compartment* should notify the *Competent Authority*.~~ A review should be initiated by the *Competent Authority* to determine whether a breach of *biosecurity* measures has occurred which could impact the health status of the compartment.

4.3.12._5_Canada	**Comment Category: Addition** **Proposed amended text:** *In the event of suspicion of occurrence of disease within the surrounding water for dependent compartments, the Competent Authority is responsible for notifying the operator and for determining if an investigation and/or additional surveillance is warranted to ensure the disease status of the dependent compartment remains unchanged or if the dependent compartment should lose its free status.* **Rationale:** If the detection of disease is at another aquaculture establishment or processing plant within the surrounding waters, a dependent compartment would not be aware of the potential change in health status and consequently impacts on their health status and biosecurity. In these cases, there may or may not be an impact for the compartment or a breach in biosecurity. However there does need to be communication between the Competent Authority and the operator as the health status upon which the compartment is dependent may have changed and as a result the risk analysis and associated risk management measures creating the functional separation would need to be reviewed.	Text was added to Article 4.3.6. on if the pathogenic agent is detected in the surrounding water body of a dependent compartment, the Competent Authority should notify the operator and asses the risk of disease exposure.
4.3.12._6_New Caledonia	**Proposition ou commentaire :** Commentaire editorial "Tout exploitant d'un établissement d'aquaculture compris dans un ~~du~~ *compartiment indemne* doit déclarer à l'*Autorité compétente* tout événement susceptible d'entraîner une faille dans les mesures de *sécurité biologique."*	Did not agree, see response to comment 4.3.2._7.

	Exposé des motifs : Article 4.3.12. second alinéa : il est proposé de remplacer “exploitant du compartiment” par “tout exploitant d’un établissement d’aquaculture compris dans un compartiment indemne” **Justification, s’il y a lieu :**	

If a significant breach in *biosecurity* is identified, even in the absence of the *disease(s)* for which the *compartment* was declared free, the *compartment*’s free status should be suspended. There should be an immediate suspension of trade to disease-free areas if a *disease* for which the *compartment* has been declared disease-free, is suspected or confirmed, and trading partners should be notified in accordance with Article 5.1.4.

Disease-free status of the *compartment* may only be reinstated by the *Competent Authority* after the depopulation, decontamination and *fallowing* have been completed, previously existing *basic biosecurity conditions* have been reviewed and modified as necessary, and *surveillance* in accordance with Chapter 1.4. and the relevant disease-specific chapter(s) has been completed. ~~*compartment* has adopted the necessary measures to re-establish the original *biosecurity* level and the *Competent Authority* re-approves the status of the *compartment*. If the health status of the *compartment* is at *risk*, the *Competent Authority* should immediately re-evaluate the status of the *compartment* and consider whether any additional *biosecurity* measures are needed to ensure that the integrity of the *compartment* is maintained.~~

4.3.12._7_China (People's Republic of)	**Category:** change **Proposed amended text :** Disease-free status of the compartment may only be re-approved and self-declared ~~reinstated~~ by the *Competent Authority* after the depopulation, decontamination and fallowing have been completed, previously existing *basic biosecurity conditions* have been reviewed and modified as necessary, and *surveillance* in accordance with Chapter 1.4. and the relevant disease-specific chapter(s) has been completed **Rationale:** The original text correctly lists the technical steps required to reinstate disease-free status (depopulation, decontamination, fallowing, surveillance, etc.). However, it omits the most critical step: the re-approval by the Competent Authority. The reinstatement of a compartment's status cannot be an automatic process upon the operator's completion of a series of technical actions. It must be verified, assessed, and formally declared reinstated by the Competent Authority. The absence of this "official re-approval" step implies that the operator could self-determine when its free status is restored, which completely contradicts the core principles of compartment management. Our proposed revision adds this necessary legal procedure after all technical steps are completed, ensuring the process is closed-loop and authoritative. **supporting evidence:** not relevant	Agreed that following loss of status, to self-declare return to freedom the compartment and its risk management measures would need to be approved again. Text revised to reflect this comment.

4.3.12._8_New Caledonia	**Proposition ou commentaire :** Commentaire editorial "Le statut sanitaire indemne du *compartiment* ne peut être ~~recouvré~~ rétabli par l'*Autorité compétente* qu'une fois que le dépeuplement, la décontamination et le *vide sanitaire* ont été achevés, que les *conditions élémentaires de sécurité biologique* précédemment existantes ont été réexaminées et modifiées si nécessaire, et qu'une *surveillance* en conformité avec les dispositions figurant dans le chapitre 1.4. et le ou les chapitres spécifiques à des maladies pertinents a été mise en place ~~achevée~~." **Exposé des motifs :** Article 4.3.12. dernier alinéa : il semble préférable de parler de "surveillance mise en place" plutôt que de "surveillance achevée" à la fin de la phrase suivante **Justification, s'il y a lieu :** /	Did not agree, self-declaration of freedom of disease can only be declared after the period of surveillance in the relevant disease-specific chapters has been completed.
4.3.12._9_United States	**Category:** general **Proposed amended text: n/a** **Rationale:** We thank the Commission for their edits to Article 4.3.12. based on previous Member comments, and we support the proposed changes to this article. **supporting evidence: -**	Noted.
4.3.12._10_EU	**Category:** Deletion and Addition **Proposed amended text:** The words "and importing countries should be notified following the provisions of Chapter 1.1" should be deleted from the second sentence of paragraph 1 ("The Competent Authority should then determine whether the disease-free status of the compartment should be immediately suspended ~~and importing countries should be notified following the provisions of Chapter 1.1.,~~ while the occurrence of the disease is confirmed or ruled-out."). These words could be added to the following sentence: "In the event of confirmation of the disease for which the compartment was defined, the free status should immediately be suspended and importing countries should be notified following the provisions of Chapter 1.1." **Rationale:** There are no such provisions for notifying import countries about suspicions in Chapter 1.1. **supporting evidence: -**	Agreed, that on suspicion of disease the Competent Authority would suspend the status of the compartment for investigation. However, notification to trading partners would occur only if confirmation of disease. Text modified to reflect comment.

GLOSSARY

[…]

Reference	Comment	Aquatic Animals Commission Response
Glossary-Compartment_1_**EU**	**Category:** General The EU supports this revised definition of the glossary.	Noted.

COMPARTMENT

means one or more *aquaculture establishments* each containing a population of *aquatic animals* with a distinct health status for a specific *disease* or *diseases* ~~that is~~ established and maintained through ~~the application of~~ a common *biosecurity* system, appropriate *surveillance* and *disease* control measures.

~~means one or more *aquaculture establishments* under a common *biosecurity* management system containing an *aquatic animal* population with a distinct health status with respect to a specific *disease* or *diseases* for which required *surveillance* and control measures are applied and *basic biosecurity conditions* are met for the purpose of *international trade*. Such must be clearly documented by the *Competent Authority(ies)*.~~

Glossary-Compartment_2_**Australia**	**Category:** addition **Proposed amended text:** "means one or more aquaculture establishments each containing a population of aquatic animals with a distinct health status for a specific disease or diseases ~~that is~~ established and maintained through ~~the application of~~ a common biosecurity system, appropriate surveillance and disease control measures, as approved by the Competent Authority." **Rationale**: Suggest additional text to clarify approval and administration and by the one competent authority, in line with comments above about article 4.3.1 **Supporting evidence:** n/a	Agreed to add 'as approved by the Competent Authority' to clarify the approval of a compartment by a Competent Authority should occur.
Glossary-Compartment_3_**China (People's Republic of)**	**Category:** change **Proposed amended text :** means one or more *aquaculture establishments* each containing a subpopulation of *aquatic animals* with a distinct health status for a specific *disease* or *diseases* that is established and maintained through ~~the application of~~ a common *biosecurity* system, appropriate *surveillance* and *disease* control measures. The establishment, approval and auditing of such a compartment must be under the management	Did not agree to change population to subpopulation. A compartment has a functional separation from other populations of aquatic animals, and is considered a distinct population. The Commission agreed to add that the compartment was

	of the Competent Authority and be clearly documented. **Rationale :** In the terrestrial animal code, the term "subpopulation" is used in the definition of compartment. And both population and subpopulation have been defined in the glossary of the code. The compartment for aquatic animals is also applicable. Suggest adding definitions for populations and subpopulations in the glossary of the Aquatic Code, being aligned with the terrestrial animal code. In principle, China agrees that the newly proposed definition (the underlined text) is more concise and clear. However, the new draft critically omits a fundamental core element: the role of the Competent Authority (CA). Necessity of Official Recognition: The essential purpose of establishing and maintaining a compartment is to gain official recognition from the national authority and trust from trading partners. Without the approval, documentation, and oversight of a Competent Authority, any private enterprise could self-declare a "compartment," rendering the concept void of credibility and legal standing in international trade. Prevention of Trade Disputes: The involvement of the CA is the prerequisite for ensuring that a compartment's status is objective, transparent, and reliable. Omitting this requirement would create significant uncertainty in international trade and could easily lead to mistrust and disputes among trading partners. Requirement for Integrity: A valid compartment requires not only establishment but also official approval and ongoing auditing to ensure its biosecurity system remains effective over time. Therefore, we strongly recommend reintroducing and strengthening the role of the Competent Authority while retaining the conciseness of the new definition. Our proposed alternative text clarifies that the establishment, approval, and auditing of a compartment must be under the management of the Competent Authority and be clearly documented. This ensures the definition is both rigorous and practical, maintaining its authority in real-world application. **supporting evidence:** not relevant	'approved by the Competent Authority' as in response to Glossary-comaprtment_2. The Commission considered that additional text suggested was unnecessary because it is well described in Chapter 4.3..

Glossary-Compartment_4_**New Zealand**	**Category:** editorial **Proposed amended text:** means one or more epidemiologically-linked *aquaculture establishments* ~~each~~ containing ~~a~~ an *aquatic animal subpopulation* ~~population~~ of *aquatic animals* with a distinct health status for a specific *disease* or *diseases* ~~that is~~ established and maintained separately from other susceptible populations through ~~the application of~~ a common *biosecurity* system, appropriate *surveillance* and *disease* control measures. **Rationale:** - Suggest making it clear that if there is more than one establishment in a compartment, that they are epidemiologically linked. - Other edits to align definition with that in the Terrestrial Code **Supporting evidence:** n/a	Did not agree to added text as this detail is addressed in Chapter 4.3. and is not considered necessary here to understand the definition. Regarding suggested edit from 'population' to 'subpopulation' see response to Glossary-compartment_3.
Glossary-Compartment_5_**United States**	**Category:** change **Proposed amended texts (or precise suggested deletion):** "means an animal subpopulation contained in one or more *aquaculture establishments*, separated from other susceptible~~each containing a~~ populations of *aquatic animals* by a common biosecurity management system, and with a specific animal ~~distinct~~health status with respect to one or more infections or infestations for which the necessary~~for a specific *disease* or *diseases* that is established and maintained through the application of a common *biosecurity* system, appropriate~~ *surveillance*, *biosecurity* and ~~*disease*~~ control measures have been applied for the purposes of international trade or disease prevention and control in a country or zone." **Rationale:** We strongly recommend closer alignment with the Terrestrial Code definition to improve usability for trade measures while addressing aquatic-specific components.	Did not agree to extensive edits. The intention of the Glossary is to provide a definition of what a compartment is while the related Chapter 4.3. provides the details on how to apply the principles for implementation of a compartment. Additional details in the Glossary create unneeded complexity in the definition.

	These proposed changes meet several of the Commission's objectives for revising Section 5 of the Aquatic Code including "Address any gaps in standards for trade", "Improve useability for developing trade measures", and "Address aquatic specific trade issues while also maintaining alignment with Terrestrial Code" (see Annex 9 also up for Member comment). **Supporting evidence: -**	

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SECTION 5

TRADE MEASURES, IMPORTATION/EXPORTATION PROCEDURES AND HEALTH CERTIFICATION

Reference	Comment	Aquatic Animals Commission Response
5._1_Australia	**Category:** general **Proposed amended text:** n/a **Rationale**: Australia strongly supports the proposed approach to the review of this section in support of safe trade of aquatic animals, noting there are numerous points for review and clarification. For example, a couple of suggestions below: Chapter 5.7 article 5.7.2 (point 2) may benefit from more detail regarding the conditions into which aquatic animals that are in transit to another importing country may be permitted to be unloaded if an emergency situation arises (eg aquatic animals may be unloaded into quarantine facilities approved by both the Competent Authority of the transit country and the Competent Authority of the importing country). In addition, guidelines as to what may constitute as an emergency warranting unloading during transit may be beneficial (eg significantly delayed freight transit period?) Chapter 5.8 article 5.8.1 may benefit from further clarification and more specific mention of: • Appropriately trained and qualified personnel at frontier posts • *'quarantine holding facilities'* to house aquatic animal populations during the importation health assessment process period at frontier posts in importing countries. • the potential application of quarantine holding periods while aquatic animal import health assessments are thoroughly carried out to completion, as deemed appropriate by the competent authority of the importing country. **Supporting evidence:** n/a	Noted, when Chapters 5.7. and 5.8. are reviewed and revised the *Terrestrial Code* revisions will be considered along with Member comments raised.
5._2_Canada	**Comment Category:** General **Proposed amended text:** n/a **Rationale**: Canada supports the comprehensive approach of the Commission to review the section and notify Member of the anticipated approach. Canada also supports the proposed approach as it works to ensure alignment between the Aquatic and Terrestrial Code which makes the standards more accessible to Members.	Noted.

	Supporting evidence: n/a	
5._3_China (People's Republic of)	**Category:** general **Proposed amended text:** n/a **Rationale**: China supports the revision of the fifth article by the Aquatic Animal Health Standards Committee. The fifth part is an important content in the Code that guides the international trade of aquatic animal safety, fully reflecting the spirit of the WTO/SPS agreement. There are the following suggestions for the revision plan of section 5: 1. There needs to be clearer guidelines for the safe trade of wild aquatic animals, which mainly involves the development of biosecurity measures for packaging establishments in exporting countries and the surveillance of diseases; 2. We noticed that the revision of the fifth article was almost simultaneously carried out with the revision of the fifth article of the Terrestrial Animal Code. Attention should be paid to the characteristics of aquatic animals and the features of international trade in aquatic animals; 3. When formulating, it is necessary to prevent the risk of excessive amplification in the equivalence assessment of aquatic products and fresh edible aquatic animals by trading countries; 4. After entry, the importing country can adopt compliance sampling surveillance to evaluate whether the biosecurity measures of the exporting country are effectively operating; 5. For items carrying pathogens, refine the requirements for accompanying documents during international transportation. For example, a certificate or statement issued by the officials in the local competent authorities is required; 6.If actively reporting the epidemic brings obstacles to trade to member countries, then the transparency of WOAH reporting will face significant challenges. Can we refer to the practice of the Authorized Economic Operator (AEO) certification of the World Customs Organization in this regard, and use notification and transparency as the recognition basis for the integrity of the quarantine system of trading countries, as well as the basis for animal and its product trade. Suggest reflecting this aspect in Section 5. **supporting evidence:** not relevant	Noted support for the approach, specific details on the revision of the chapters will be taken into consideration when revision of Section 5 commences. At its September 2026 meeting, the Commission will consider further the plan for revisions.
5._4_Ecuador	**Categoría:** General **Propuesta/Comentario:** ítem: Objetivos de la revisión de la Sección 5. Se solicita a la Comisión para los Animales Acuáticos homologar el criterio 1 del Artículo 5.4.1 del Código Acuático al criterio 1 del 2.2.2 del Código Terrestre, para que ambos consideren como mercancía segura aquella en la que “existen pruebas convincentes de que el agente patógeno no está presente en los tejidos de los que se deriva el producto de animal en una cantidad capaz de producir infección en personas o animales”. **Justificación:**	Noted. Specific details on the revision of the chapters will be taken into consideration when revision of Section 5 commences. At its September 2026 meeting, the Commission will consider further the plan for revisions.

El Código Acuático reconoce una mercancía como segura por ausencia del patógeno, o si el tratamiento o métodos de procedimiento lo inactivan. En cambio, el Código Terrestre considera la presencia del patógeno en niveles que no representen riesgo de infección en animales susceptibles.

Los ambientes acuáticos favorecen la diseminación de microorganismos en comparación a los ambientes terrestres, siendo muchas enfermedades de distribución global e imposibles de erradicar, como son IHHNV[1] y WSSV[2] (1,2). Existen factores que permiten un comercio seguro de mercancías aún con presencia de patógenos, tales como:

- Bajas cargas de patógeno en animales resistentes, por ejemplo IHHNV (6-9).

- Bajo impacto de enfermedades que no generan signología clínica, tales como ciertas cepas de IHHNV (6,8,10,16,17).

- Zonas de escasa prevalencia: reconocidas en el Artículo 6 de los Acuerdos MSF de la OMC[3] (3-5).

- Congelamiento del camarón como forma de preservación (11), que pese a su comercio masivo no ha mostrado evidencia de transmisión de enfermedades (12).

- Menor riesgo asociado al camarón destinado al consumo humano respecto al comercio de animales vivos, además de los procesos previos al consumo (12-15).

- Múltiples eventos que ocurren desde la salida del producto desde el país de origen hasta su consumo en el país de destino (18).

Evidencia documentada, si corresponde:

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15. Soto, M. A., Shervette, V. R., & Lotz, J. M. (2001). Transmission of white spot syndrome virus (WSSV) to *Litopenaeus vannamei* from infected cephalothorax, abdomen, or whole shrimp cadaver. *Diseases of aquatic organisms, 45*, 81-87.
16. Dhar, A. K., Cruz-Flores, R., Warg, J., Killian, M. L., Orry, A., Ramos, J., ... & Lyons, G. (2022). Genetic relatedness of infectious hypodermal and hematopoietic necrosis virus isolates, United States, 2019. *Emerging infectious diseases, 28*(2), 373.

	17. Ofori-Amponsah, E., Martinez, M. A., Meehan, M. T., & Condon, K. The Phylogenetics and Pathogen Population Dynamics of Infectious Hypodermal and Hematopoietic Necrosis Virus (IHHNV) in Global Shrimp Populations. *Available at SSRN 5702023*. 18. Ejercicio de modelación para estimar la probabilidad de transmisión de enfermedades a partir de la exportación de camarón ecuatoriano. CERES BCA.	
5._5_Ecuador	**Category**: general **Propuesta/Comentario:** ítem "Otras consideraciones relevantes para los intercambios comerciales". Se solicita a la Comisión para los Animales Acuáticos agregar en los capítulos específicos de cada enfermedad (Artículos X.X.3 y X.X.14[4]) para el comercio independientemente del estatus sanitario del origen, un punto que reconozca la evaluación de riesgo como herramienta para definir en que condiciones un producto es seguro, facilitando el comercio al no restringir los productos únicamente a los listados. **Justificación:** Los Capítulos específicos de cada enfermedad de animales acuáticos definen listados de productos seguros que pueden limitar el comercio y no consideran la evaluación de riesgo para definir otras medidas equivalentes, independiente del estatus sanitario del país de origen o de la presencia del patógeno en el producto. Tal como se explica la propuesta/comentario anterior, existen factores que permiten un comercio seguro de mercancías aún con la presencia de patógenos, lo cual es considerado por la evaluación de riesgo. Esta metodología también recoge otros factores de relevancia, tales como la sensibilidad y especificidad de la técnica diagnóstica, donde como ejemplo para IHHNV se ha descrito hasta un 82% de falsos positivos con las técnicas de PCR[5] recomendadas por la OMSA[6] (19). La evaluación de riesgo es parte del espíritu del Código Acuático, siendo reconocida en el Capítulo 5.3 como base para determinar equivalencia de medidas sanitarias entre países. Un ejemplo en animales terrestres es el comercio de carne positiva a *Campylobacter*, la que puede exportarse cumpliendo principios basados en Buenas Prácticas de Higiene y una adecuada gestión del sistema productivo, entendiendo que *Campylobacter* es una bacteria ubicua, presente en todos los países y por tanto imposible de erradicar (20). **Evidencia documentada, si corresponde:** Bibliografía de la 3 a la 18 de la propuesta/comentario anterior.	Noted. Specific details on the revision of the chapters will be taken into consideration when revision of Section 5 commences. At its September 2026 meeting, the Commission will consider further the plan for revisions.

	19. Intriago, P., Del Barco, M., Vásquez, M. M., Montiel, B., & Villamar, R. (2025). Insights on Minimizing False Positives in IHHNV Detection: Experiences from Ecuador's *Penaeus vannamei* Aquaculture. *International Journal of Molecular Sciences, 26*(23), 11484. 20. Codex Alimentarius. (2011). Directrices para el control de Campylobacter y Salmonella en la carne de pollo. CAC/GL 78-2011.	
5._6_Ecuador	ítem: Objetivos de la revisión de la Sección 5. Se solicita a la Comisión para los Animales Acuáticos incorporar explícitamente al Capítulo 5.3 el reconocimiento de la implementación progresiva de medidas sanitarias y la asistencia técnica entre las partes interesadas, descritos en los Artículos 9 y 10 de los Acuerdos MSF, para alcanzar el estándar recomendado por la OMSA en función del NAP[7] acordado con el o los países importadores. Justificación: El Artículo 10 de los Acuerdos MSF de la OMC, señala que "cuando el NAP permita el establecimiento gradual de nuevas medidas sanitarias o fitosanitarias, deberán concederse plazos más largos para su cumplimiento con respecto a los productos de interés para los países en desarrollo Miembros, con el fin de mantener sus oportunidades de exportación". Esto no queda explícito en el contenido del Capítulo 5.3 del Código Acuático de la misma forma que se trata lo relativo a la equivalencia de medidas sanitarias, dando a entender que la implementación de los estándares recomendados por la OMSA se deben aplicar desde el inicio de la relación comercial entre países. Incorporarlo puede facilitar el comercio, fortaleciendo progresivamente la gestión del riesgo. Lo mismo sucede con el Artículo 9 de los Acuerdos MSF, el cual promueve la asistencia técnica entre países, incluyendo tecnologías de elaboración, investigación e infraestructura, mediante asesoramiento, financiamiento, capacitación y provisión de equipos, para facilitar el cumplimiento de las medidas sanitarias requeridas por los mercados de destino. Item: Objectives of the review of Section 5.	Noted. During the next September meeting, section 5 will be reviewed, taking into consideration the work with the terrestrial code
5._7_Japan	**Category:** General Comment **Proposed amended text**: Not relevant **Rationale:** In Japan, Health certificates for aquatic animals are issued by officers in fisheries departments of local governments which are responsible for aquatic animal disease prevention and control.	Noted. Specific details on the revision of the chapters will be taken into consideration when revision of Section 5 commences.

	We acknowledge that the proposal of Terrestrial Code Annex 8 Article 5.4.2 encourages to limit export procedures only through Veterinary Authority. In addition, the proposal in Terrestrial Code Annex10 Article 5.6.3 mentions that international veterinary certificates may only be issued by the Official Veterinarian. However, fisheries departments of Japanese local governments which are responsible for aquatic animal disease prevention and control also can confirm the sanitary conditions of aquatic animals through officers who are experts in aquatic animal health. In addition, the health certificates issued by Japanese local government can be used for safe and sound import/export. Taking into account this situation, health certificate in Aquatic code should be issued by not only veterinarians but also officers who are experts in aquatic animal health in fisheries departments of local governments which are responsible for aquatic animal disease prevention and control. Furthermore, we request that the name of the health certificate be changed from “international veterinary certificate” to “international aquatic animal health certificate” as per the Aquatic code. **Supporting evidence, if relevant:** not relevant	
5._8_Norway	**Category**: general **Proposed amended text**: Not relevant **Rationale:** Norway wishes to express its support for the revision of Section 5 in the Aquatic Code. We have a few comments for the Commission to consider when revising the section: <u>Certificates:</u> Norway considers this an important opportunity to promote a common direction for safe, efficient, and paperless exchange of certificates. We regard the digitalisation of certificate exchange as a necessity, both for reasons of security, cost-effectiveness, and sustainability. At the same time, it is important that WOAH encourages secure electronic exchange without specifying any particular technological format. Technological developments are moving quickly, and it is essential that the standards are forward-looking and technology-neutral, so they do not bind Member Countries to solutions that may become outdated. Norway wishes to highlight that many countries are already in a position to implement e-cert solutions, where certificates are exchanged directly between national systems. This represents the most secure and cost-effective model. At the same time, we recognise that many countries, for various reasons, need a gradual transition. It is therefore important that	Noted. There is a planned *ad hoc* Group on e-certification in both the *Aquatic Code* and the *Terrestrial Code* that will address this (see item 8.1.2. in the Commission’s February 2025 report). Regarding traceability, see response to comment 4.3.8._2. In relation to SPS agreement and other trade issues, the Commission will work into specific details on the chapters when revision of Section 5 commences.

	the standards also accommodate temporary, yet secure and user-friendly digital solutions that enable the phase-out of paper in the short term. In this regard, Norway considers electronically signed and encrypted original certificates in PDF format to be a practical and secure step for countries that are not yet ready for full e-cert integration. This approach ensures security, control, and accessibility, while also supporting further digital maturity and harmonisation over time. Norway therefore recommends that WOAH's revised standards enable: 1. **Technology neutrality**, allowing solutions to evolve in line with technological progress. 2. **Secure and verifiable digital exchange** as an overarching principle. 3. **Flexible implementation pathways**, enabling Member Countries to adopt digital solutions at a level suited to their technological maturity – for example electronically signed PDF certificates as an interim step – and gradually move toward full electronic certificate exchange when ready. <u>Additional comments</u>: • Continue to ensure alignment with the SPS Agreement. • Traceability systems covering tracking of a product back from the end user to the seller of the same product (business-to-consumer transaction) is to be decided and implemented by the national importing authorities and not the competent authority and operators of the exporting country. • Should countries be able to impose possibly unreasonable trade restrictions, e.g. impose restrictions based on disease presence in the exporting country while not having susceptible species for the disease in question on their territory? **Supporting evidence, if relevant:** not relevant	
5._9_United States	Category: general We thank the Commission for this important work to review, and revise when necessary, Section 5 of the Aquatic Code. We recommend the following be taken into consideration throughout this revision process: **1) Keep certificates outcome-based, animal health focused, and commodity-specific.** Revisions should reinforce that model health certificates address only the relevant disease risks for the specific commodity, avoiding conditions not	Noted. The Commission will work into specific details on the chapters when revision of Section 5 commences.

	transmitted by that commodity. This maintains precision and reduces arbitrary barriers, consistent with existing Section 5 certification principles. (https://www.woah.org/fileadmin/Home/eng/Health_standards/aahc/current/chapitre_certification_general.pdf) **2) Prioritize usability via digital-by-default design.** Modernize Section 5 chapters and model certificates for harmonized electronic certification (e-cert), standard data fields, and clearer instructions—so Competent Authorities and industry can reduce errors, speed clearance, and improve consistency in border processing. (Aligns with Annex 9's stated focus on improving usability.) **3) Align with the Terrestrial Code only where aquatic realities match.** Support alignment for coherence, but ensure aquatic-specific differences (multi-species consignments, ornamental pathways, live aquatic animal production/management logistics) are preserved. Avoid copy-over constructs from Terrestrial that don't fit aquatic supply chains. **4) Clarify interfaces with Section 4 (compartmentalization, fallowing, etc.).** As Section 5 is revised, WOAH should explicitly reference how certification and eligibility link to updated Chapter 4.3 (compartments) and Chapter 4.7 (fallowing), minimizing administrative ambiguity for exporters/importers and Competent Authorities. **5) Facilitate legitimate movement of diagnostic/pathological materials.** Preserve risk-managed pathways in Chapter 5.9 for international transport of disease agents/pathological material needed for surveillance, proficiency testing, and R&D—avoiding unnecessary impediments while maintaining biosafety.	
5._10_EU	**Category**: general **Proposed amended text**: Not relevant **Rationale:** The EU supports the proposed approach to revision of Section 5 **Supporting evidence, if relevant:** not relevant	Noted.

Objectives of Section 5 revision:

- Revise older standards and improve quality as necessary
- Address any gaps in standards for trade
- Improve useability for developing trade measures
- Address aquatic specific trade issues while also maintaining alignment with *Terrestrial Code*
- Improve alignment of chapters within Section 5 and between Section 5 and other sections of the *Aquatic Code*

Aquatic Code **Section 5 structure**	***Terrestrial Code*** **Section 5 structure** (for comparison only)	***Aquatic Code*** **Proposed approach**

N/A	N/A	**New chapter.** Development of a new introductory chapter to Section 5 in cooperation with the Code Commission (see item 7.1.2 in this report)
5.1. General obligations related to certification	5.1. General obligations related to certification	**Review**. Consider the need for revision in collaboration with the Code Commission. Proposed certification a*d hoc* Group to review chapter in both Codes and provide recommendations (see item 8.1.2. Feb 2025 report).
5.2. Certification procedures	5.2. Certification procedures	**Review**. Consider need for revision in collaboration with the Code Commission. Proposed certification a*d hoc* Group to review chapter in both Codes and provide recommendations (see item 8.1.2. Feb 2025 report).
5.3. WOAH procedures relevant to the Agreement on the Application of Sanitary and Phytosanitary Measures of the World Trade Organization	5.3. WOAH procedures relevant to the Agreement on the Application of Sanitary and Phytosanitary Measures of the World Trade Organization	**Revise**. Consider need for revision in collaboration with the Code Commission
5.4. Criteria to assess the safety of aquatic animal commodities	N/A	**Relocate.** Move to section 2, risk analysis (as for TC)
5.5. Control of aquatic animal health risks associated with transport of aquatic animals	N/A	**Revise.** Last adopted in 2010. Consider whether more appropriate in Section 4 or 5. If remains in Section 5 consider renumbering.
5.6. Aquatic animal health measures applicable before and at departure	5.4. Animal health measures applicable before and at departure	**Revise.** Consider revisions proposed to equivalent *Terrestrial Code* chapter.
5.7. Aquatic animal health measures applicable during transit from the place of departure in the exporting country to the place of arrival in the importing country	5.5. Animal health measures applicable during transit from the place of departure in the exporting country to the place of arrival in the importing country	**Revise.** Consider revisions proposed to equivalent *Terrestrial Code* chapter.
5.8. Frontier posts in the importing country	5.6. Border posts and quarantine stations in the importing country	**Revise.** Consider revisions proposed to equivalent *Terrestrial Code* chapter.
5.9. Aquatic animal health measures applicable on arrival	5.7. Animal health measures applicable on arrival	**Revise.** Consider revisions proposed to equivalent *Terrestrial Code* chapter.
5.10. Measures concerning international transport of aquatic animal pathogens and pathological material	5.8. International transfer and laboratory containment of animal pathogenic agents	**Revise.** Last adopted 2010.

N/A	5.9. Quarantine measures applicable to non-human primates	Not relevant to Aquatic Code
5.11. Model health certificates for international trade in live aquatic animals and products of aquatic animal origin	5.10. Model veterinary certificates for international trade in live animals, hatching eggs and products of animal origin	**Review.** Consider alignment between the three model certificates and with other provisions of the *Aquatic Code.* Proposed certification a*d hoc* Group to review chapter in both Codes and provide recommendations (see item 8.1.2. Feb 2025 report)
5.12. Movement of ornamental aquatic animals	N/A	**No action.** Adopted 2025.
N/A	5.11. Model veterinary certificate for international movement of dogs, cats and ferrets originating from countries considered infected with rabies	Not relevant to Aquatic Code
NA	5.12. Model passport for international movement of competition horses	Not relevant to Aquatic Code
N/A	5.13. Model veterinary certificate for international trade in laboratory animals	Consider need for a model certificate in *Aquatic Code* or whether Ch 5.11 of *Aquatic Code* is sufficient.

5._11_Chinese Taipei	**Category:** Addition **Proposed amended text:** Agree to adding a model certificate in the Aquatic Code. **Rationale:** None. **Supporting evidence:** Not relevant.	Noted.

Other trade relevant considerations:

- Revision of trade articles in disease specific chapters (X.X.9 to X.X14; 10.4.13. to 10.4.20.); including reordering of articles and revision of commodity and pathway (end-use) combination; including addressing commodity gaps (e.g. ornamental aquatic animals)
- Consider diagrammatic guidance on appropriate application of provisions of disease-specific chapters, possibly in the User's Guide
- Consider general guidance on trade in live aquatic animal for aquaculture similar in approach to new ornamentals chapter
- Define "official control" considering approach in terrestrial code, and IPCC definition

CHAPTER 2.2.9.

INFECTION WITH *APHANOMYCES ASTACI* (CRAYFISH PLAGUE)

Reference	Member	Comment	Aquatic Animals Commission response
2.4.9_1	**EU**	**Category:** General The EU supports in general the proposed update to these sections. Nevertheless, specific comments are provided below.	Noted.

[…]

2.2. Host factors

2.2.1. Susceptible host species

Species that fulfil the criteria for listing as susceptible to infection with *A. astaci* according to Chapter 1.5. of the *Aquatic Animal Health Code* (*Aquatic Code*) are:

Family	Scientific name	Common name
Astacidae	*Astacus astacus*	noble crayfish
	Austropotamobius pallipes	no common name
	Pacifastacus leniusculus	signal crayfish
	Pontastacus leptodactylus	Danube crayfish
Cambaridae	*Faxonius spp.* (all species)	N/A
	Procambarus spp. (all species)	N/A
Cambaroididae	*Cambaroides japonicus*	no common name
Parastacidae	*Cherax quadricarinatus*	red claw crayfish
Potamidae	*Potamon potamios*	no common name
Varunidae	*Eriocheir sinensis*	Chinese mitten crab

2.4.9_2.2.1_1	**Thailand**	**Category:** change **Proposed amended text:** Propose to list susceptible species in Genus *Faxonius* and *Procambarus* at the species level, as follows: Family: Cambaridae Scientific name: ~~*Faxonius spp.* (all species)~~ *Faxonius limosus*; *Faxonius obscurus*; *Faxonius rusticus*; ~~*Procambarus* spp. (all species)~~ *Procambarus alleni*; *Procambarus clarkii* Common name: ~~N/A~~ spinycheek crayfish; no common name; no common name; ~~N/A~~ no common name; red swamp crayfish **Rationale/ Supporting evidence:** Please see comment for Annex 10.	Did not agree, Article 1.5.9. was applied and *Faxonius* and *Procambarus* were listed at the Genus level as described in the Commission's September 2025 report (item 6.5.). Review of the scientific justification for Article 1.5.9. is supplied in the Commission's September 2025 report (item 7.3.1.).

2.4.9_2.2.1_2	**Perú**	**Categoría:** Editorial **Texto modificado propuesto:** *Austropotamobius pallipes* / ~~sin nombre común~~ cangrejo de patas blancas **Justificación:** Se reporta abundante literatura sobre el nombre común de la especie *Austropotamobius pallipes*, siendo "cangrejo de patas blancas". **Evidencia documentada:** 1. Bruno M.C., Endrizzi S., Basso A., Paolini V. & Pretto T. (2025). Crayfish plague and microsporidiosis occurrence in wild populations of the white-clawed crayfish Austropotamobius pallipes complex in Trentino (North-East Italy). J. Invertebr. Pathol., 108487. https://doi.org/10.1016/j.jip.2025.108487 2. Cartwright A., Rice A.R., Dooley J.S., Smyth J. & Arnscheidt J. (2022). Impact of dietary protein content on growth of the white-clawed crayfish Austropotamobius pallipes (Lereboullet, 1858) (Decapoda, Astacidae) in captive rearing for conservation. Crustaceana, 95(5-6), 517-532. 3. Martínez-Ríos M., Martín-Torrijos L., Casabella-Herrero G., Tedesco P., Machordom A. & Diéguez-Uribeondo J. (2023). On the conservation of white-clawed crayfish in the Iberian Peninsula: Unraveling its genetic diversity and structure, and origin. Plos One, 18(10), e0292679. https://doi.org/10.1371/journal.pone.0292679	Agreed, that the common name proposed in the comment is correct. *Austropotamobius* was revised to be listed at the Genus level, so common name for species no longer reflected in the text.
2.4.9_2.2.1_3	**EU**	**Category**: Addition **Proposed amended text:** including *Cherax destructor* to 2.2.1. 'Susceptible host species' **Rationale**: evidence of infection in farmed population of *Cherax destructor* showing increased mortality and clinical signs (loss of appendages, paralysis upside down) melanisation of the cuticle and histological evidence of hyphae branching in the cuticle and hypodermis, supported by molecular identification of *A. astaci* by end-point PCR and sequencing. **Supporting evidence:** IAA 19 Conference 2012, Innsbruck, Austria. Quaglio et al., 2012. First occurrence of *Aphanomyces astaci* epidemic infection in cultured yabby *Cherax destructor* (Clark, 1936) in Northern Italy. Book of abstract, page 88 https://www.astacology.org/docs/symposia/IAA19/docs/IAA19_Abstracts.pdf	Did not agree, reference was referred to subject matter expert. On review of the reference it did not provide enough information to assess all stages as described in Chapter 1.5. and *Cherax destructor* could not be scored based on the provided reference.

<table>
<tr>
<td>2.4.9_2.2.1_4</td>
<td>China (People's Rep. of)</td>
<td>
Category: change

Proposed amended text:
<table>
<tr><th>Family</th><th>Scientific name</th><th>Common name</th></tr>
<tr><td rowspan="5">Astacidae</td><td>Astacus astacus</td><td>noble crayfish</td></tr>
<tr><td>Austropotamobius pallipes</td><td>no common name</td></tr>
<tr><td>Austropotamobius torrentium</td><td>stone crayfish</td></tr>
<tr><td>Pacifastacus leniusculus</td><td>signal crayfish</td></tr>
<tr><td>Pontastacus leptodactylus</td><td>Danube crayfish</td></tr>
</table>
Rationale: The stone crayfish (Austropotamobius torrentium) is a susceptible host for crayfish plague, with multiple studies confirming its vulnerability. It is recommended to adjust from 2.2.2 to 2.2.1.

supporting evidence:
1.Aphanomyces +astaci isolate+fnomyces-astaci.p 2.infection-apha
</td>
<td>Agreed, evidence was referred to subject matter expert who revised the score to '1'. Article 1.5.9. was applied to Astropotamobius and it was listed at the Genus level (see item 8.1.1. in report).</td>
</tr>
</table>

~~The recommendations in this chapter apply to all species of crayfish in all three crayfish families (Cambaridae, Astacidae and Parastacidae).~~

~~[**Note**: an assessment of species that meet the criteria for listing as susceptible to infection with *A. astaci* in accordance with Chapter 1.5. has not yet been completed]~~

2.2.2. Species with incomplete evidence for susceptibility

Species for which there is incomplete evidence to fulfil the criteria for listing as susceptible to infection with *A. astaci* according to Chapter 1.5. of the *Aquatic Code* are:

Family	Scientific name	Common name
Astacidae	*Austropotamobius torrentium*	no common name
Cambaridae	*Cambarellus (Cambarellus) montezumae*	no common name
	Cambarellus patzcuarensis	no common name
	Cambarellus shufeldtii	Cajun dwarf crayfish
	Cambarus bartonii	Appalachian brook crayfish

<table>
<tr>
<td>2.4.9_2.2.2_1</td>
<td>Thailand</td>
<td>Category: change

Proposed amended text: Propose to add the list of species with incomplete evidence for susceptibility from Genus Faxonius and Procambarus, as follows:

<table>
<tr><th>Family</th><th>Scientific name</th><th>Common name</th></tr>
<tr><td rowspan="10">Cambaridae</td><td>Faxonius immunis</td><td>calico crayfish</td></tr>
<tr><td>Faxonius propinquus</td><td>no common name</td></tr>
<tr><td>Faxonius virilis</td><td>no common name</td></tr>
<tr><td>Procambarus enoplosternum</td><td>no common name</td></tr>
<tr><td>Procambarus fallax</td><td>no common name</td></tr>
<tr><td>Procambarus llamasi</td><td>no common name</td></tr>
<tr><td>Procambarus raneyi</td><td>no common name</td></tr>
<tr><td>Procambarus vazquezae</td><td>no common name</td></tr>
<tr><td>Procambarus virginalis</td><td>no common name</td></tr>
</table>

Rationale/ Supporting evidence: Please see comment for Annex</td>
<td>Did not agree, see response to comment 2.4.9_2.2.1_1.</td>
</tr>
<tr>
<td>2.4.9_2.2.2_2</td>
<td>Mexico</td>
<td>Categoría cambio.

Texto modificado propuesto (o supresión sugerida):

<table>
<tr><td>Astacidae</td><td>Austropotamobius torrentium</td><td>~~no common name~~
Stone crayfish</td></tr>
</table>

Justificación: El nombre común es cangrejo de río y en inglés se ha publicado como stone cryfish. Las referencias encontradas son en libros y listados.

Evidencia documentada, si corresponde: Bulletin français de la pêche et de la pisciculture. Editorial: Conseil supérieur de la pêche. Tomos 378-381. 2006. Pag.1019 y 1030.</td>
<td>Agreed, that the common name proposed in the comment is correct. Austropotamobius was revised to be listed at the Genus level, so common name for species no longer reflected in the text.</td>
</tr>
<tr>
<td>2.4.9_2.2.2_3</td>
<td>EU</td>
<td>Category: Addition

Proposed amended text: moving Austropotamobius torrentium from 2.2.2 ‘Species with incomplete evidence for susceptibility’ to 2.2.1. ‘Susceptible host species’

Rationale: evidence of infection in natural population of A. torrentium showing micromelanisation of the cuticle, supported by isolation of A. astaci genotype As from crayfish tissues and identification of the isolate by PCR followed by sequencing; additionally, evidence of mortality induced by experimental infection by immersion with A. astaci genotype PsI zoospores.

Supporting evidence: Jussila et al. 2017 Aphanomyces astaci isolate from latently infected stone crayfish (Austropotamobius torrentium) population is virulent. http://dx.doi.org/10.1016/j.jip.2017.07.003</td>
<td>Agreed, see response to comment 2.4.9_2.2.1_4.</td>
</tr>
</table>

<table>
<tr>
<td>2.4.9_2.2.2_4</td>
<td>China (People's Rep. of)</td>
<td>
Category: change

Proposed amended text:
<table>
<tr><th>Family</th><th>Scientific name</th><th>Common name</th></tr>
<tr><td>~~Astacidae~~</td><td>~~Austropotamobius torrentium~~</td><td>~~no common name~~</td></tr>
<tr><td rowspan="4">Cambaridae</td><td>Cambarellus (Cambarellus) montezumae</td><td>no common name</td></tr>
<tr><td>Cambarellus patzcuarensis</td><td>no common name</td></tr>
<tr><td>Cambarellus shufeldtii</td><td>Cajun dwarf crayfish</td></tr>
<tr><td>Cambarus bartonii</td><td>Appalachian brook crayfish</td></tr>
</table>
Rationale: The stone crayfish (Austropotamobius torrentium) is a susceptible host for crayfish plague, with multiple studies confirming its vulnerability. It is recommended to adjust from 2.2.2 to 2.2.1.

supporting evidence:
1.Aphanomyces +astaci isolate+fnomyces-astaci.p 2.infection-apha
</td>
<td>Agreed, see response to comment 2.4.9_2.2.1_4.</td>
</tr>
</table>

In addition, pathogen-specific positive polymerase chain reaction (PCR) results have been reported in the following species, but no active infection has been demonstrated:

Family	Scientific name	Common name
Cambaridae	*Cambarus latimanus*	no common name
	Cambarus striatus	Hay crayfish
	Creaserinus fodiens	no common name
	Creaserinus oryktes	no common name
Palaemonidae	*Palaemon kadiakensis*	no common name

~~[Under study]~~

<table>
<tr>
<td>2.4.9_2.2.2_2</td>
<td>Thailand</td>
<td>Category: change

Proposed amended text: Propose to add the list of species in with pathogen-specific PCR results have been reported but no active infection has been demonstrated from Genus Faxonius and Procambarus, as follows:

<table>
<tr><th>Family</th><th>Scientific name</th><th>Common name</th></tr>
<tr><td rowspan="11">Cambaridae</td><td>Faxonius etnieri</td><td>no common name</td></tr>
<tr><td>Faxonius tricuspis</td><td>no common name</td></tr>
<tr><td>Faxonius wrighti</td><td>no common name</td></tr>
<tr><td>Procambarus ablusus</td><td>no common name</td></tr>
<tr><td>Procambarus acutus</td><td>white river crayfish</td></tr>
<tr><td>Procambarus hayi</td><td>no common name</td></tr>
<tr><td>Procambarus hybus</td><td>no common name</td></tr>
<tr><td>Procambarus pubescens</td><td>no common name</td></tr>
<tr><td>Procambarus troglodytes</td><td>no common name</td></tr>
<tr><td>Procambarus viaeviridis</td><td>vernal crayfish</td></tr>
<tr><td>Procambarus vioscai</td><td>no common name</td></tr>
</table>

Rationale/ Supporting evidence: To be in line with Table 8 from the Report of the WOAH ad hoc Group on the Susceptibility of Crustacean Species to Infection with WOAH-Listed Diseases (April, 2025).</td>
<td>Did not agree, see response to comment 2.4.9_2.2.1_1.</td>
</tr>
</table>

[…]

CHAPTER 2.4.5.

INFECTION WITH *PERKINSUS MARINUS*

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_1	**EU**	**Category:** General The EU supports the proposed amendments. For the sake of harmonisation, the EU would encourage the Aquatic Animals Commission to provide the same level of detail notably for the case definitions as it is the case for *Bonamia exitiosa, Bonamia ostreae* or *Marteilia refringens*. Please see below comments to the case definitions In addition, the EU proposes to reintroduce a reference to a real-time-PCR in point 4.4.1, that it is widely used.	Agreed: the Commission agreed to stay with the standard approach to retain two confirmatory diagnostic tests as presented in Table 4.1. for the case definitions. .
2.4.5_2	**Australia**	**Category:** general **Proposed amended text:** n/a **Rationale**: For most of the mollusc chapters, in Table 4.1 assays are validated to Stage 2. This includes estimates of DSe and DSp which need to be published in peer reviewed literature and added to Table 6.3.1 and/or Table 6.3.2. If there is no data in these tables, there is no peer-reviewed evidence to support claims of validation to Stage 2 (or greater). Tables 6.3.1 and 6.3.2. were added to provide the reader with some evidence of the level of validation of recommended tests – if data exists but is unpublished there is the validation template that can be used. If the AAHSC do not require published estimates of validation to support claims of Stage 2 in Table 4.1 then everyone will just add Stage 2 which defeats the purpose of what the ad hoc group and AAHSC was trying to achieve. **Supporting evidence:** n/a	The Commission rechecked the literature and found that no assays have been validated to level 2; Table 4.1 has been amended accordingly. Tables 6.3.1 and 6.3.2. remain unpopulated.

1. Scope

Infection with *Perkinsus marinus* means infection with the pathogenic agent *Perkinsus marinus,* of the Family Perkinsidae.

2. Disease information

2.1. Agent factors

2.1.1. Aetiological agent

Perkinsus marinus is a Perkinsid parasite initially described infecting eastern oysters, *Crassostrea virginica* and causing disease and mortality (Mackin *et al.,* 1950).

Current phylogenomic studies place *P. marinus* within the class Perkinsea of the phylum Perkinsozoa, under the Kingdom Alveolata (Reece *et al.,* 1997).

The parasite occurs as motile zoospores, trophozoites, and schizonts. Following infection, trophozoites proliferate within oyster tissues, particularly in connective tissue and haemolymph. As trophozoites grow, they undergo schizogony: cleavage furrows form, leading to the production of multiple merozoites within a morula-like stage. Fully developed schizonts rupture to release large numbers of merozoites, which disseminate through host tissues or into the environment (Andrews, 1988; Villalba *et al.,* 2004).

As in *P. olseni*, the ribosomal RNA transcription units in *P. marinus* are highly duplicated and variable, complicating the use of rDNA as a reliable quantitative marker for infection intensity (Bogema *et al.,* 2021).

Alternative genomic regions, including internal transcribed spacer (ITS) sequences and non-ribosomal single-copy genes, have been explored for molecular diagnostics (Audemard *et al.,* 2004; De Faveri *et al.,* 2009; Gauthier *et al.,* 2006; Reece *et al.,* 1997; Robledo *et al.,* 1999).

Cytogenetic and molecular studies suggest that *P. marinus* may also display variable ploidy. Early work using microsatellite loci and karyotyping suggested diploidy (Reece *et al.,* 1997; Robledo *et al.,* 1999), but subsequent genomic sequencing has raised the possibility of polyploidy or aneuploidy, with variation across life stages and isolates (Bogema *et al.,* 2021; Thompson *et al.,* 2011). Further investigation is needed to resolve the extent and biological implications of ploidy variation in this species.

2.1.2. Survival and stability in processed or stored samples

The survival and stability of *P. marinus* varies according to the conditions when processing and storing samples. Refrigeration (2–6°C) and low salinity (7 ppt) significantly decreases the viability and metabolic activity of *P. marinus* but could not eliminate *P. marinus* from oysters (La Peyre *et al.,* 2008b; 2010). It is likely that *P. marinus* can survive in dead or frozen oysters for an unknown, potentially long period (Andrews & Hewatt, 1957).

2.1.3. Survival and stability outside the host

Perkinsus marinus meront cells can survive for at least 14 days in the water column, with their infectivity potentially persisting beyond 7days (Chu & Lund, 2006).

For inactivation methods, see Section 2.4.5.

2.2. Host factors

2.2.1. Susceptible host species

Species that fulfil the criteria for listing as susceptible to infection with *P. marinus* according to Chapter 1.5. of the *Aquatic Animal Health Code* (*Aquatic Code*) are:

Family	**Scientific name**	**Common name**
Ostreidae	*Crassostrea corteziensis*	Cortez oyster
	Crassostrea virginica	American cupped oyster
	Magallana [syn. *Crassostrea*] *ariakensis*	Ariake cupped oyster
	Saccostrea palmula	palmate oyster

2.2.2. Species with incomplete evidence for susceptibility

Species for which there is incomplete evidence to fulfil the criteria for listing as susceptible to infection with *P. marinus* according to Chapter 1.5. of the *Aquatic Code* are:

Family	Scientific name	Common name
Ostreidae	*Crassostrea tulipa*	Gasar cupped oyster
	Crassostrea rhizophorae	mangrove cupped oyster
	Magallana [syn. *Crassostrea*] *gigas*	Pacific cupped oyster

In addition, pathogen-specific positive polymerase chain reaction (PCR) results have been reported in the following species, but no active infection has been demonstrated:

Family	Scientific name	Common name
Myidae	*Mya arenaria*	soft shell clam
Ostreidae	*Crassostrea columbiensis*	Columbia black oyster

	Striostrea prismatica	stone oyster

2.2.3. Likelihood of infection by species, host life stage, population or sub-populations

Perkinsus marinus usually infects and kills oysters between 1 and 3 years of age. Differences in clinical expression to the infection with *Perkinsus marinus* has been shown between *Crassostrea virginica* populations (e.g. Bushek & Allen, 1996; Green-Beach *et al.,* 2011).

Compared with *C. virginica, Crassostrea corteziensis, Saccostrea palmula* and triploid *M. ariakensis* were found to exhibit partial resistance to *P. marinus* infection and associated disease (Cáceres-Martínez *et al.,* 2012; 2016; Calvo *et al.* 2000; 2001).

2.2.4. Distribution of the pathogen in the host

While *P. marinus* can be found in various tissues and organs, it mostly infects intestine, vesicular connective tissue, haemocytes and digestive gland (Remacha-Triviño *et al.,* 2008). The proliferation of the parasite causes systemic disruption of connective tissue and epithelial cells.

2.2.5. Aquatic animal reservoirs of infection

None known

2.2.6. Vectors

None known

2.3. Disease pattern

2.3.1. Mortality, morbidity and prevalence

Infection is often lethal for *C. virginica*. Death usually occurs 1 or 2 years after infection, during or shortly after the warmest annual water temperatures (Burreson & Ragone Calvo, 1996). Prevalence is expected to be higher in individuals with more than 1 year of exposure to the pathogen (Andrews, 1996; Burreson & Ragone Calvo, 1996).

Prevalence is highly variable depending on host populations, pathogen strain and environmental factors (Kim *et al.,* 2025; Proestou *et al.,* 2023). Along the US east coast, *C. virginica* populations consistently exhibit high *P. marinus* infection rates, typically exceeding 80% and sometimes reaching 100% prevalence (Smolowitz, 2013). While populations from the Mexico Gulf typically exhibit low to moderate prevalence, with infection intensities generally remaining light (Ammons *et al.,* 2023; Cáceres-Martínez *et al.,* 2016; Gullian-Klanian *et al.,* 2008).

Infection prevalence and intensity are consistently low in *M. ariakensis*, *C. corteziensis* and *S. palmula* populations (Cáceres-Martínez *et al.,* 2012; 2016; Calvo *et al.,* 2000; 2001). Notably, no significant *P. marinus*-associated mortality has been observed in natural populations of *C. corteziensis* and *S. palmula* (Cáceres-Martínez *et al.,* 2012; 2016). Furthermore, *M. ariakensis* demonstrates superior survival, faster growth, and reduced disease susceptibility compared to *C. virginica* across varying salinity regimes (low, medium, and high) (Calvo *et al.,* 2001).

2.3.2. Clinical signs, including behavioural changes

Perkinsus marinus causes a chronic wasting disease.

Infected oysters may have a pale appearance to the digestive gland, reductions in condition index, severe emaciation, gaping, shrinkage of the mantle away from the outer edge of the shell, reduced gonadal development and/or retardation of growth (Ford & Tripp, 1996). Occasionally, pus-like pockets may occur in the soft tissues. However, these signs are not pathognomonic of perkinsosis.

2.3.3 Gross pathology

Gross signs are thin, watery tissue, eventually presence of pus-like pockets, but these gross signs are not specific to infection with *P. marinus*.

2.3.4. Modes of transmission and life cycle

The life cycle is direct from host to host (Andrews, 1996; Villalba *et al.,* 2004). *Perkinsus marinus* is transmitted directly from host to host through the water column following release of parasites in faeces or the decay or scavenging of moribund tissues (Bushek *et al.,* 2002). The parasite enters the paleal cavity of the oyster during filter-feeding. Labial palps and particularly the pseudo-faeces discharge area of the mantle are important sites of infection (Allam *et al.,* 2013; Winnicki *et al.,* 2008). Parasites infect and multiply within haemocytes. The infected circulating haemocytes migrate throughout the host tissues.

Experimental infection in controlled conditions is possible by exposing animals to *in vitro* culture of *P. marinus* (Dungan *et al.,* 2007).

2.3.5. Environmental factors

The prevalence and intensity of *P. marinus* are primarily driven by salinity and temperature. On the east coast of US, infections are acquired during the warm months of the year, when the water temperatures reach above 18°C (Smolowitz, 2013). Proliferation of the parasite and associated lesions are correlated with temperature. Temperature controls the annual cycle of *P. marinus*, with maximum prevalence and intensity lagging 1–2 months behind maximum summer water temperatures and minimum prevalence and intensity lagging 1–2 months behind minimum winter temperatures. Thus, *P. marinus* infections are most intense in autumn and least intense in early spring (Burreson & Ragone Calvo, 1996). Parasite infection intensity peaked at about 35°C (Malek *et al*., 2018). In tropical regions such as the Gulf of Mexico, however, *P. marinus* proliferates at significantly higher temperatures, suggesting an adaptation of host-parasite dynamics to these conditions (Cáceres-Martínez *et al.,* 2016).

Perkinsus marinus cells were released on a diurnal cycle, with most cells released during the hottest and brightest period of the day (12:00–18:00). Temperature had a strong and immediate effect on the number of cells released, but salinity did not, only influencing the intensity of infection over the course of several months (Gignoux-Wolfsohn *et al*., 2020). Prevalence and intensity of *P. marinus* infections are greatest at salinities greater than 12 practical salinity units (psu). Transmission can occur between 9 and 12 psu, but infections remain low in intensity. *Perkinsus marinus* can persist for long periods in hosts at salinities less than 9 psu, but replication is low and no host mortality occurs.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_2.3.5_1	**New Zealand**	**Category:** editorial **Proposed amended text:** …Parasite infection intensity peaked at about 35°C (Malek *et al*., 2018). In tropical regions such as the Gulf of Mexico, however, *P. marinus* can proliferates even at significantly higher temperatures, suggesting an adaptation of host-parasite dynamics to these conditions (Cáceres-Martínez *et al.,* 2016). *Perkinsus marinus* cells ~~were~~ are released on a diurnal cycle, with most cells released during the hottest and brightest period of the day (12:00–18:00). Temperature has ~~had~~ a strong and immediate effect on the number of cells released, but salinity does ~~did~~ not, only influencing the intensity of infection over the course of several months (Gignoux-Wolfsohn *et al*., 2020). **Rationale:** Changed to present tense so it is consistent with rest of section. **Supporting evidence:** n/a	Agreed. . .

2.3.6. Geographical distribution

Infection with *P. marinus* has been reported from the Atlantic coast of North and South America (da Silva *et al.,* 2013). It was introduced to the Pacific coast of North America (Cáceres-Martínez *et al.,* 2008).

Reference	Member	Comment	Aquatic Animals Commission response

2.4.5_2.3.6_1	**Perú**	**Categoría:** Adición **Texto modificado propuesto:** "…Also, it has been reported from the east coast of Korea (Kim *et al.*, 2024)" **Justificación:** Se ha reportado la presencia de *P. marinus* en la costa este de Corea **Evidencia documentada:** Kim S.H., Bathige S.D.N.K., Jeon H.B., Lee D., Choi K.S., Kim, H.J. & Park K.I. (2024). First report of *Perkinsus marinus* occurrence associated with wild Pacific oysters *Crassostrea gigas* from the west coast of Korea. Journal of Invertebrate Pathology, **204**, 108119. https://doi.org/10.1016/j.jip.2024.108119	Agreed . .

See WAHIS (https://wahis.woah.org/#/home) for recent information on distribution at the country level.

2.4. Biosecurity and disease control strategies

2.4.1. Vaccination

None.

2.4.2. Chemotherapy including blocking agents

N-Halamine disinfectant compounds killed cultured *P. marinus* cells without affecting oyster larvae (Delaney *et al.,* 2003). Bacitracin, cycloheximide and freshwater have been shown to reduce, but not eliminate *P. marinus* in infected oyster hosts (Calvo & Burreson, 1994; Faisal *et al.,* 1999; La Peyre *et al.,* 2003). Moreover, the antimicrobial drug triclosan (5-chloro-2-[2,4 dichlorophenoxy]) phenol, may be effective in treating *P. marinus*-infected oysters (Chu *et al.,* 2008; Lund *et al.,* 2005). Finally copper (as $CuCl_2$) was shown to greatly reduce infection levels of *P. marinus* in haemocytes *in vivo* (Foster *et al.,* 2011) These treatments may be relevant for aquaculture, but are not practical in the natural environment.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_2.4.2_1	**New Zealand**	**Category:** editorial **Proposed amended text:** N-Halamine disinfectant compounds ~~killed cultured~~ can kill *P. marinus* cells without affecting oyster larvae (Delaney *et al.,* 2003). ….Finally copper (as $CuCl_2$) ~~was shown to~~ can greatly reduce infection levels of *P. marinus* in haemocytes *in vivo* (Foster *et al.,* 2011) **Rationale:** Changed to present tense so it is consistent with rest of section. **Supporting evidence:** n/a	Agreed. . .

2.4.3. Immunostimulation

None.

2.4.4. Breeding resistant strains

Selective breeding of surviving oysters from epizootics has demonstrated effectiveness for reducing mortality caused by *P. marinus* (Ragone Calvo *et al.,* 2003).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_2.4.4_1	**Perú**	**Categoría:** Adición	Agreed.

		Texto modificado propuesto: "It has been reported that survival in genomically selected individuals is 20% higher than survival in individuals selected through phenotypic selection"." **Justificación:** Se ha reportado que, la supervivencia en ejemplares seleccionados genómicamente es superior a la supervivencia en ejemplares seleccionados mediante selección fenotípica en un 20%. **Evidencia documentada:** Wang Z., Casas S., La Peyre J., Rikard S., Williams M.L., Tarnecki A., ... & Guo X. (2025). Genomic Selection for Dermo Resistance in the Eastern Oyster Crassostrea virginica: Production and Laboratory Testing of F1 Generation. J. Shellfish Res., 44, 75-87. https://doi.org/10.2983/035.044.0108	

2.4.5. Inactivation methods

Desiccation, chlorination (>0.3 mg ml^{-1} = 300 ppm [parts per million]), UV light (>28,000 µWs cm^{-2}) and freshwater have all been shown to inactivate *P. marinus* cells (Bushek *et al.,* 1997; Bushek & Howell 2000). UV irradiation from 4000 to 14,000 µWs cm^{-2} will inhibit proliferation of *P. marinus* (Bushek & Howell, 2000).

2.4.6. Disinfection of eggs and larvae

Perkinsus marinus is not known to infect eggs or larvae, but cells could occur extracellulary. Disinfection of eggs or larvae may be possible using N-halamine disinfectant compounds (Delaney *et al.,* 2003).

2.4.7. General husbandry

Farming in areas where salinity is less than 12 psu and use of fast-growing, disease-tolerant strains has shown some benefit (Andrews, 1996; Burreson & Ragone Calvo, 1996).

Water filtration to 1 µm followed by UV exposure should allow protecting hatchery produced seed against infection (Ford *et al.,* 2001). Inversely, treating water effluents should minimise the transmission of *P. marinus* to local oyster populations from infected oysters maintained in facilities such as processing plants, depuration facilities or hatcheries (Bushek & Howell, 2000).

Cultured off-bottom and air daily exposed oysters from infected area showed increased survival and higher condition indexes (La Peyre *et al.,* 2008).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_2.4.7_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Farming in areas where salinity is less than 12 psu and use of fast-growing, disease-resistant ~~tolerant~~ strains has shown some benefit (Andrews, 1996; Burreson & Ragone Calvo, 1996). **Rationale:** Change "resistant" to "tolerant" to be consistent with terminology with rest of chapter. **Supporting evidence:** n/a	Disagreed, the published paper used the term "tolerant'. . .

3. Specimen selection, sample collection, transportation and handling

This section draws on information in Sections 2.2, 2.3 and 2.4 to identify populations, individuals and samples that are most likely to be infected.

3.1. Selection of populations and individual specimens

Gaping or freshly dead individuals (2 or more years old) should be sampled by priority, to increase the chances of finding infected animals. For histology, only live (including moribund) animals should be sampled.

Sampling of bivalves should be organised once a year when prevalence is known to be at a maximum. When such data is not available in a particular ecosystem, sampling should preferably be carried out during periods of oyster spawning which also corresponded with warmer temperatures (Burrell *et al.,* 1984).

3.2. Selection of organs or tissues

For Ray's fluid thioglycollate culture method (RFTM), pieces of gill, mantle and rectum are typically used. For histology, a 5-mm thick section through the visceral mass that includes digestive gland, gill and mantle is used. For PCR, a pool of organs including digestive gland, gill and mantle is best.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_3.2_1	**Canada**	**Category:** addition Canada requests the reference(s) for this tissue type selection be included for Member information.	Disagreed: the information is a summary from multiple published sources and Reference Laboratory experience.

3.3. Samples or tissues not suitable for pathogen detection

Tissues other than digestive gland, mantle and gills are less suitable.

3.4. Non-lethal sampling

A real-time PCR assay targeting the multicopy internal transcribed spacer region of the genome was developed to detect *P. marinus* DNA in environmental waters allowing detecting equivalent of $3.3 \times 10^{(-2)}$ cell per 10-microl reaction mixture (Audemard *et al.,* 2004).

Perkinsus cells can be detected from haemolymph, faeces or water samples using RFTM methods (Bushek *et al.,* 2002; Ellin & Bushek, 2006; Gauthier & Fisher, 1990).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_3.4_1	**Canada**	**Category:** addition **Proposed amended text:** At this time, there are no non-lethal tissue samples that can support detection of *P.marinus.* A real-time PCR assay targeting the multicopy internal transcribed spacer region of the genome was developed to detect *P. marinus* DNA in environmental waters allowing detecting equivalent of $3.3 \times 10^{(-2)}$ cell per 10-microl reaction mixture (Audemard *et al.,* 2004). *Perkinsus* cells can be detected from haemolymph, faeces or water samples using RFTM methods (Bushek *et al.,* 2002; Ellin & Bushek, 2006; Gauthier & Fisher, 1990, Gauthier & Vasta (1995). Diagnostic sensitivity and specificity is not available for these test methods on environmental samples. GAUTHIER J.D. & VASTA G.R. (1995). *In Vitro* Culture of the Eastern Oyster Parasite *Perkinsus marinus:* Optimization of the Methodology. *J. Invertebr. Pathol.*, **66**, 156–168.	Agreed and slightly amended the proposed amendment.

		Rationale: **Change 1:** This proposed sentence is added to support Members to ensure it is clear that there are currently no non-lethal tissue sampling options for *P.marinus*. These other methods are helpful for Member but cannot be used to support self-declarations of freedom. An alternative option is to create 2 subsections withing Section 3.4. to differentiate between non-lethal tissue sampling and non-lethal environmental sampling. **Change 2**: Gauthier & Vasta (1995) is used as a reference in the *P.olseni* chapter to support the use of haemolymph incubation as alternative to the traditional tissue RFTM. If this is the reference chosen for the *P.olseni* chapter, it should be included within the *P.marinus* chapter for completeness. **Change 3:** We also are proposing an additional sentence to this section to address the lack of information available on Diagnostic Sensitivity and specificity for testing environmental samples. This information should be captured within the manual for Members knowledge however, it is not appropriate to add these samples and tests information to tables 4.1., 6.3.1. or 6.3.2.	

3.5. Preservation of samples for submission

For guidance on sample preservation methods for the intended test methods, see Chapter 2.4.0 General information (diseases of molluscs).

3.5.1. Samples for pathogen isolation

The results of bioassay depend strongly on the quality of samples (time since collection and time in storage). Fresh specimens should be kept on ice and preferably sent to the laboratory within 24 hours of collection. To avoid degradation of samples, use alternate storage methods only after consultation with the receiving laboratory.

Standard sample collection, preservation and processing methods for molecular techniques can be found in Section B.5.5 of Chapter 2.4.0 *General information* (diseases of molluscs).

For diagnosis using RFTM, samples must be fresh.

3.5.2. Preservation of samples for molecular detection

For polymerase chain reaction (PCR) assays, samples must be preserved in 70–100% ethanol and not denatured alcohol.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_3.5.2_1	**Australia**	**Category:** change **Proposed amended text:** For polymerase chain reaction (PCR) assays, samples must be preserved in ~~70–100%~~ 80% ethanol and not denatured alcohol. **Rationale:** Better to specify 80% analytical grade ethanol to be consistent with Section 2.4, Chapter 2.4.0, Section B, Part 5.5.1.iii **Supporting evidence:** n/a	Agreed and for consistency added 'analytical grade' [ethanol]

Standard sample collection, preservation and processing methods for molecular techniques can be found in Section B.5.5 of Chapter 2.4.0 *General information* (diseases of molluscs).

3.5.3. Samples for histopathology, immunohistochemistry or *in-situ* hybridisation

For histology, the best preservative is Davidson's AFA, but 10% buffered formalin or other standard histology fixatives are also acceptable.

Standard sample collection, preservation and processing methods for histological techniques can be found in Section B.5.3 of Chapter 2.4.0 *General information* (diseases of molluscs).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_3.5.3_1	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** 3.5.3. Samples for histopathology, immunohistochemistry or in-situ hybridisation (ISH) **Rationale:** The abbreviation "ISH" should be defined on its first use in the text to aid reader comprehension. **Supporting evidence:** not relevant	Agreed.

3.5.4. Samples for other tests

None.

3.6. Pooling of samples

Pooling of samples from more than one individual animal for a given purpose is only recommended where robust supporting data on diagnostic sensitivity and diagnostic specificity have been evaluated and found to be suitable. If the effect of pooling on diagnostic sensitivity has not been thoroughly evaluated, larger specimens should be processed and tested individually. Small life stages such as spat can be pooled to obtain the minimum amount of material for molecular detection. Pooling of very small spat (5–10 depending on size) is acceptable for PCR analyses.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_3.6_1	**Canada**	**Category:** deletion **Proposed amended text:** …Small life stages such as spat can be pooled to obtain the minimum amount of material for molecular detection. ~~Pooling of very small spat (5–10 depending on size) is acceptable for PCR analyses.~~ **Rationale:** Canada has completed a scientific evaluation on the usefulness of small (<6mm) life stages of molluscs for testing of spat for disease freedom which found a gap in the literature of validated studies using spat below 6mm for PCR testing. This evaluation found that there was not enough evidence to support the use of spat smaller than 6mm in sampling protocols. If the pathogenic agent cannot be found in small spat, or that that test methods are not sensitive enough to detect low prevalence infection in spat, these are not appropriate samples. Therefore, Canada questions the inclusion of pooling of very small spat as pooling would further impact the sensitivity and confidence in detection rates. **Supporting evidence:** Suitability of testing of early life stages of *Crassostrea virginica* (larvae to 5 mm spat) by PCR-based assays for the detection of *Haplosporidium nelsoni, Mikrocytos mackini, Perkinsus marinus,* and *Bonamia exitiosa*.	Agreed.

4. Diagnostic methods

The methods currently available for pathogen detection that can be used in i) surveillance of apparently healthy animals, ii) presumptive diagnosis in clinically affected animals and iii) confirmatory diagnostic purposes are listed in Table 4.1. by animal life stage.

Ratings for purposes of use. For each recommended assay a qualitative rating for the purpose of use is provided. The ratings are determined based on multiple performance and operational factors relevant to application of an assay for a defined purpose. These factors include appropriate diagnostic performance characteristics, level of assay validation, availability cost, timeliness, and sample throughput and operability. For a specific purpose of use, assays are rated as:

+++ =	Methods are most suitable with desirable performance and operational characteristics.
++ =	Methods are suitable with acceptable performance and operational characteristics under most circumstances.
+ =	Methods are suitable, but performance or operational characteristics may limit application under some circumstances.
Shaded boxes =	Not appropriate for this purpose.

Validation stage. The validation stage corresponds to the assay development and validation pathway in chapter 1.1.2. The validation stage is specific to each purpose of use. Where available, information on the diagnostic performance of recommended assays is provided in Section 6.3.

WOAH Reference Laboratories welcome feedback on diagnostic performance of recommended assays, in particular PCR methods. Of particular interest are any factors affecting expected assay sensitivity (e.g. tissue components inhibiting amplification) or expected specificity (e.g. failure to detect particular genotypes, detection of homologous sequences within the host genome). These issues should be communicated to the WOAH Reference Laboratories so that advice can be provided to diagnostic laboratories and the standards amended if necessary.

Table 4.1. WOAH recommended diagnostic methods and their level of validation for surveillance of apparently healthy animals and investigation of clinically affected animals

Method	A. Surveillance of apparently healthy animals				B. Presumptive diagnosis of clinically affected animals				C. Confirmatory diagnosis[1] of a suspect result from surveillance or presumptive diagnosis			
	Early life stages[2]	Juveniles[2]	Adults	LV	Early life stages[2]	Juveniles[2]	Adults	LV	Early life stages[2]	Juveniles[2]	Adults	LV
Wet mounts												
Histopathology		++	++	1		+++	+++	NA				
Cell culture												
Transmission electron microscopy									++	++	++	NA
Real-time PCR	+++	+++	+++	2 1	+++	+++	+++	2	+++	+++	+++	2
Conventional PCR	++	++	++	1	++	++	++	1				
Conventional PCR followed by amplicon sequencing									+++	+++	+++	1
In-situ hybridisation										+	+	1
Bioassay												
LAMP												
Ab-ELISA												
Ag-ELISA												
RFTM		++	+++	2		++	+++	NA				

LV = level of validation, refers to the stage of validation in the WOAH Pathway (chapter 1.1.2); PCR = polymerase chain reaction; LAMP = loop-mediated isothermal amplification; Ab- or Ag-ELISA = antibody or antigen enzyme-linked immunosorbent assay, respectively. RFTM = Ray's fluid thioglycollate culture method

[1]For confirmatory diagnoses, methods need to be carried out in combination (see Section 6). [2]Susceptibility of early and juvenile life stages is described in Section 2.2.3.

[3]Specify the test used. Shading indicates the test is inappropriate or should not be used for this purpose.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_Table4.1_1	**Thailand**	**Category:** General (Clarification) We would like to request WOAH to recheck the Level of Validation (LV) in Table 4.1 for the real-time PCR method. If it is confirmed as LV 2, the DSe and DSp values should be specified in Section 6.3. However, if such data are currently unavailable, the LV in Table 4.1 should be revised accordingly **Proposed amended text:** No text proposed **Rationale:** To ensure accuracy **Supporting Document:** -	The Commission rechecked the literature and found that no assays have been validated to level 2;Table 4.1 has been amended accordingly. Tables 6.3.1 and 6.3.2. remain unpopulated. . .
2.4.5_Table4.1_2	**Canada**	**Category:** deletion **Proposed amended text:** Please remove the levels of validation for early life stages in A, B and C or clarify that early life stages must be >6mm due to a lack of validated studies using spat below 6mm for PCR testing. **Rationale:** We note that Section 2.2.3 does not indicate susceptible of early life stages or suitability of early life stages for testing. At what size 'early life stage oysters' transition from seed to juveniles. Canada has completed a scientific evaluation on the usefulness of small (<5mm) life stages of molluscs for testing of spat for disease freedom which found a gap in the literature of validated studies using spat below 6mm for PCR testing. This evaluation found that there was not enough evidence to support the use of spat smaller than 6mm in sampling protocols. If the pathogenic agent cannot be found in small spat, or that that test methods are not sensitive enough to detect low prevalence infection in spat, these are not appropriate samples. **Supporting evidence:** Suitability of testing of early life stages of Crassostrea virginica (larvae to 5 mm spat) by PCR-based assays for the detection of *Haplosporidium nelsoni, Mikrocytos mackini, Perkinsus marinus,* and *Bonamia exitiosa*.	Agreed and amended the scores in Table 4.1. .
2.4.5_Table4.1_3	**Canada**	**Category:** General Canada thanks the Commission for responding to previous comments to indicate that DSe and Dsp are not always published but may be provided by the Reference Laboratory to support the ratings provided within Table 4.1 and therefore DSe and DSp cannot be included within Tables 6.1.2 and 6.1.3. When there is no published information on diagnostic sensitivity in the scientific literature and the rating is based on unpublished reference laboratory validation studies, we would request that a footnote including an explanation for the discrepancy between the rating and the published literature would enhance transparency and clarity for Table 4.1.	Noted for future updates, when relevant. At present, the validation study template has been used for one chapter only: Infection with yellow head virus genotype 1.

2.4.5_Table4.1_4	**Australia**	**Category:** change **Proposed amended text:** n/a **Rationale**: In Table 4.1, for Real-time PCR, the level of validation (LV) to Stage "2" (for both A. Surveillance of healthy animals and B. Presumptive diagnosis of clinically affected animals), means DSe and DSp have been determined so these values need to be added to Table 6.3.1 and Table 6.3.2, respectively. **Supporting evidence:** n/a	See comment 2.4.5_Table4.1_1 above

4.1. Smears

Not recommended as a diagnostic method.

4.2. Histopathology

Samples to be taken consist of fresh, gaping or freshly dead bivalves.

Sections of tissue that include gills, digestive gland, mantle, and gonad should be fixed for 24 hours minimum in a recommended fixative followed by standard processing for histology as described in section 5.3 of Chapter 2.4.0 *General information* (diseases of molluscs). Observations are made at increasing magnifications up to ×1000.

Histology is generally less sensitive than PCR methods (see Sections 6.1. and 6.2).

A positive result is the occurrence of spherical, uninucleated cells, trophozoïtes, ranging from about 2 to 5 µm (sometimes to 10 µm) in diameter with a large vacuole and an eccentrically displaced nucleus. These are typically associated with host gut or sometimes epithelium in lighter infections, with colonisation of connective tissues characteristic of more advanced cases. Binucleated *P. marinus* schizonts (dividing forms) may be observed, though larger schizonts of greater nuclear count are not reliably presented. Many of the parasites may occur within oyster haemocytes. *Perkinsus marinus* cells stain basophilic. A change in *Perkinsus* marinus phenotype (large trophozoïtes at low infection intensity were replaced by abundant tinny cells) was reported since 1986 in oysters from Chesapeake Bay by Carnegie *et al.,* 2021).

4.3. Electron microscopy

Fixed sections reveal large multifocal lesions in gut epithelium or connective tissue of any organ containing *P. marinus* cells (Mackin, 1951). Haemocyte infiltration and phagocytosis of *P. marinus* cells occurs in most infections. In high intensity infections, the gut epithelium may be almost completely destroyed.

Perkinsus marinus zoospores, like zoospores of all species of *Perkinsus,* show an ornamented anterior flagellum with hair-like and spurs-like structures and a glabrous posterior flagellum. The zoospore contains an apical complex consisting of a conoid, subpellicular microtubules, rhoptries, rectilinear micronemes and conoid-associated micronemes. Large vacuoles also occur at the anterior end of the zoospore (Perkins 1976; 1996).

4.4. Nucleic acid amplification

PCR assays should always be run with the controls specified in Section B.5.5 *Molecular methods* Chapter 2.4.0 *General information* (diseases of molluscs). Molluscs are known to potentially contain substances that can inhibit PCR reactions. It is recommended to check for the presence of PCR inhibitors in DNA extracts to avoid false negative results. In case PCR inhibitors are present, DNA samples can be diluted prior to PCR analyses (a 1/10 dilution usually resolves most cases of PCR inhibition). Each sample should be tested in duplicate.

Several PCR assays have been developed for the detection of *Perkinsus* sp. and *P. marinus*, most of them target portions of the rRNA gene complex. Primers that target the NTS region have demonstrated good species specificity for *P. marinus*, however, little is known of the variation within species for the NTS region and there is a risk of false negatives. The sequence variation in the ITS region is more broadly characterised (see the GenBank database) and primers targeting the ITS region are more thoroughly tested for specificity. For these reasons, primers that target the ITS region are recommended

Depending on the context of analysis, *Perkinsus* genus PCR assays be conducted first, and then samples with positive results should be tested with a *P. marinus* specific assay.

Extraction of nucleic acids

Samples to be taken consist of live or freshly dead molluscs. 2–3 mm^2 tissue pieces are excised aseptically from gill, mantle and digestive gland placed into 1.5 ml microcentrifuge. Dissecting utensils should be flamed between samples to prevent cross-contamination.

Different kits and procedures can be used for nucleic acid extraction. DNA is usually extracted by proteinase K digestion at 56°C (generally overnight), and the spin-column methodology using commercially available kits. Faster extraction protocols using Chelex resin has also been described (De Faveri *et al.,* 2009; Piesz *et al.,* 2022). The quality and concentration of the extracted nucleic acid is important and can be checked using a suitable method as appropriate to the circumstances.

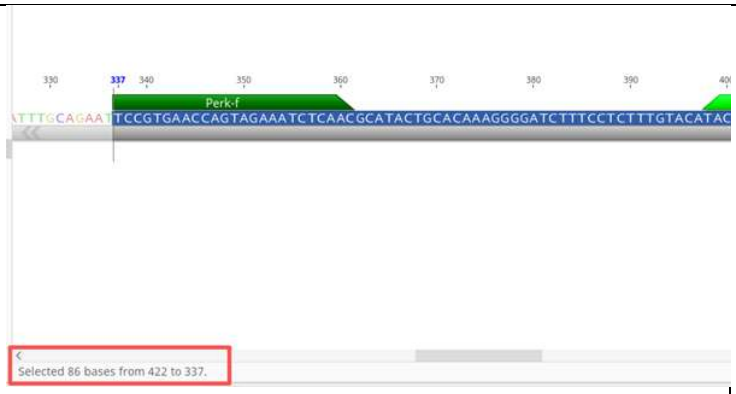
4.4.1. Real-time PCR

***Primers and probes* (*sequences*)**

Pathogen/ target gene	Primer/probe (5'–3')	Concentration	Cycling parameters[a]
Method 1: TaqMan PCR: Gauthier *et al.,* 2006, GenBank Accession No.: AF149876.1			
Perkinsus sp./ITS (detects at least *P. marinus, P. olseni* and *P. chesapeaki*)	PERK-F: TCC-GTG-AAC-CAG-TAG-AAA-TCT-CAA-C PERK-R: GGA-AGA-AGA-GCG-ACA-CTG-ATA-TGT-A PERK Probe: (FAM) CCC-TTT-GTG-CAG-TAT-GC-MGB	0.9 µM 0.9 µM 0.25 µM	40 cycles of: 95°C/15 sec and 60°C/1 min
Method 2: TaqMan PCR: Gauthier *et al.,* 2006; GenBank Accession No.: AF149876.1; amplicon size: 72 bp			
Perkinsus marinus/ITS	PMAR F: TTG-TTA-ACG-CAA-CTC-AAT-GCT-TTG-T PMAR R: AAG-CGC-ACA-TAA-CGA-ACC-ACC PMAR-MGB Probe: (FAM)-GCT-TGA-ACT-AAC-TCT-MGB-	0.9 µM 0.9 µM 0.25 µM	40 cycles of: 95°C/15 sec and 60°C/1 min
Method 3: Real-time PCR: De Faveri *et al.,* 2009; GenBank Accession No.: No data; amplicon size: 90 bp			
Perkinsus marinus/ITS	F: CGC-CTG-TGA-GTA-TCT-CTC-GA R: GTT-GAA-GAG-AAG-AAT-CGC-GTG-AT Probe: [FAM]CGC-AAA-CTC-GAC-TGT-GTT-GTG-GTG	0.2 µM 0.2 µM 0.04 µM	35 cycles at 95°C for 30 sec, and 60°C for 45 sec

[a]A denaturation step prior to cycling has not been included.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_4.4.1_1	**EU**	**Category:** addition **Proposed amended text:** Include the SYBR Green assay described by Audemard *et al.* (2004) using the PmarITS-70F and Pmar-600R primers (amplifying a 509bp product). **Rationale:** This is the assay having the longest history of use.	Agreed. . .
2.4.5_4.4.1_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** Method 1: TaqMan PCR: Gauthier *et al.,* 2006, GenBank Accession No.: AF149876.1; amplicon size: 86 bp **Rationale:** The amplicon size was supplemented. The length was obtained from the sequence downloaded from Genbank (Accession No.: AF149876.1).	Agreed. . .

		 **Supporting evidence:** not relevant

Interpretation of results:

A sample is considered positive when a PCR amplification curve is observed (with the expected Ct value when a cut-off Ct value has been established) with the appropriate melting temperature peak (Tm) in case of SYBR Green PCR assays, with all negative controls being negative and all positive controls being positive.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_4.4.1_3	**Australia**	**Category:** change **Proposed amended text:** A sample is considered positive when a PCR amplification curve is observed (with the expected Ct value when a cut-off Ct value has been established) ~~with the appropriate melting temperature peak (Tm) in case of SYBR Green PCR assays~~, with all negative controls being negative and all positive controls being positive. **Rationale**: Delete as no SYBR Green assays have been described. **Supporting evidence:** n/a	Disagreed as the SYBR green PCR has been reinstated.

Depending on the context/objective of the analysis, a cut-off Ct value can be used to define if samples are positives or negatives. The cut-off Ct value may vary depending on the equipment, reagents and consumables used for the test.

If PCR inhibitors are detected in a sample producing a negative result, this sample should be considered as uninterpretable.

4.4.2. Conventional PCR

Pathogen/ target gene	Primer/probe (5'–3')	Concentration	Cycling parameters[a]
Method 1: Audemard *et al.*, 2004 (primers from Casas *et al.*, 2002); GenBank Accession No.: AF149876.1, amplicon size: 703 bp			
Perkinsus sp./ITS (detects all known *Perkinsus* species *except P. qugwadi*)	PerkITS-85: CCG-CTT-TGT-TTG-GAT-CCC PerkITS-750: ACA-TCA-GGC-CTT-CTA-ATG-ATG	0.5 µM 0.5 µM	40 cycles of: 95°C/1 min and 55°C/1min and 72°C/1min Final elongation 72°C/10 min
Method 2: Audemard *et al.*, 2004; GenBank Accession No.: AF149876.1, amplicon size: 509 bp			
Perkinsus marinus/ITS	PmarITS-70F: CTT-TTG-YTW-GAG-WGT-TGC-GAG-ATG PmarITS-600R: CGA-GTT-TGC-GAG-TAC-CTC-KAG-AG	0.5 µM 0.5 µM	40 cycles of: 95°C/1 min and 57°C/1min and 72°C/3min Final elongation 72°C/10 min

[a]A denaturation step prior to cycling has not been included.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_4.4.2_1	**USA**	**Category:** Addition **Rationale:** We recommend adding an assay to Section 4.4.2. of this chapter specific for targeting non-overlapping regions, to best comply with the statement in Section 6. of this chapter – "When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome."	Noted for future updates should such assays be developed.
2.4.5_4.4.2_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** Method 1: Audemard *et al.*, 2004 (primers from Casas *et al.*, 2002); GenBank Accession No.: AF149876.1, amplicon size: 666 ~~703~~ bp **Rationale:** After querying, the amplified fragment size between PerkITS-85 and PerkITS-750 in AF149876.1 is 666 bp (including primer sequences). The 703 bp fragment mentioned in the literature Audemard et al., 2004 cites Casas et al., 2002 as a reference, but we did not find any description of 703 bp fragment in Casas et al., 2002 or its cited literature Goggin, 1994. **supporting evidence:** Goggin, C. L., 1994. Variation ii	Agreed.

4.4.3. Other nucleic acid amplification methods

None.

4.5. Amplicon sequencing

The size of the PCR amplicon should be verified, for example by agarose gel electrophoresis. Obtained sequences are analysed in comparison with reference sequences.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_4.5_1	**USA**	**Category:** change **Proposed amended texts:** Please see the proposed change in blue –The ~~size of the~~ PCR amplicon of the correct size should be sequenced and compared to ~~verified, for example by agarose gel electrophoresis. Obtained sequences are analysed in comparison with~~ reference sequences. **Rationale:** We recommend the above change for clarity, and to tighten and enhance the flow of the language. Also, by stating that the PCR amplicon is the correct size, it is implied that you verified it was the correct size by (most likely) using agarose gel electrophoresis.	Disagreed, the text is understood as written and is the standard text from the template.

4.6. *In-situ* hybridisation

Samples to be taken consist of live or freshly dead molluscs.

A specific DNA probe that targets the small subunit rRNA gene has been developed for the genus *Perkinsus*: Perksp700DIG (Elston *et al.,* 2004) and a DNA probe that targets the LSU of the rRNA gene has been developed for the specific detection of *P. marinus:* PmarLSU-181 (Moss *et al.,* 2006). Additionally, two other *P. marinus* probes also targeting the LSU region has been developed: Pmar LSU-420 and LSU 560 (Reece *et al.,* 2008), and can be used in cocktails with the first one.The probe should be 5' end-labelled with digoxigenin. Alternatively, probes can be labelled with a fluorochrome (ex. Alexa Fluor 488®)

Pathogen/target	ISH probe type	ISH probe (5'-3')	Reference
Perkinsus sp./SSU (detects all known *Perkinsus* species *except P. qugwadi)*	Labelled oligonucleotide	PerkspSSU-700 CGC-ACA-GTT-AAG-TRC-GTG-RGC-ACG	Elston *et al.* (2004)
Perkinsus marinus/LSU	Labelled oligonucleotide(s) (PmarLSU-181 alone or cocktail of the 3)	PmarLSU-181 GAC-AAA-CGG-CGA-ACG-ACT-C PmarLSU-420 GAA-GAC-AGG-AGC-GAG-CAG-C PmarLSU-560 AAC-CAA-TTC-ACA-GAT-AGC-G	Moss *et al.* (2006); Reece *et al.* (2008)

4.7. Immunohistochemistry

Not available.

4.8. Bioassay

Not available.

4.9. Antibody- or antigen-based detection methods (ELISA, etc.)

Polyclonal antibody-based analyte-capture ELISAs are reported to detect *P. marinus* extracellular secretions (ECP) in tissue homogenates from oysters with infection intensities of one parasite cell/g oyster tissue (Ottinger *et al.,* 2001). The specificity of polyclonal antibodies against *P. marinus* ECP was not tested against other *Perkinsus* spp. or marine mollusc protistan associates.

4.10. Other methods

4.10.1. Ray's fluid thioglycollate culture method (RFTM)

Incubation in thioglycollate is routinely used for surveillance of *P. marinus.* The technique is simple, inexpensive and very sensitive, but not species specific. Trophozoites of *P. marinus* in oyster tissue will enlarge when cultured for at least 5 days in fluid thioglycollate medium containing dextrose that is fortified with antibiotics (penicillin, streptomycin) and an antifungal compound (nystatin) to reduce bacterial and fungal growth. When the tissue is macerated after culture to allow penetration of aqueous iodine solution (Lugol's), the enlarged trophozoites (hypnospores or prezoosporangia in the old terminology) readily take up Lugol's and become easily visible at low power because of their bluish–black colouration and spherical shape.

Samples to be taken consist of live or freshly dead molluscs.

Tissue assay (*Ray, 1966*): tissue samples measuring approximately 5–10 mm are excised, giving preference to rectal, gill and mantle tissue from oysters, and placed in test tubes containing thioglycollate medium (thioglycollate medium containing dextrose 14.6 g; NaCl, 10.0 g; sterile distilled water (dH_2O), 485 ml). A total of 9.5 ml is dispensed into disposable test tubes, which are autoclaved for 15 minutes at 1.2 kg cm^{-2} pressure. The autoclaved solution can be stored in tubes for up to 3 weeks. Dissecting utensils should be rinsed in 95% ethanol and flamed between hosts to prevent carry-over. The recommended antifungal/antibiotics are: 500 units ml^{-1} penicillin G and 500 units ml^{-1} dihydro-streptomycin in media (penicillin, 3.13 g; streptomycin, 6.55 g; 500 ml dH_2O; freeze in 50 ml aliquots; add 0.5 ml to each tube), and 50 µl of mycostatin (nystatin) per

tube. Chloromycetin can be used in place of penicillin/streptomycin. The tube is plugged with a foam rubber or cotton stopper. Incubation is at 22–25°C for between 5 and 7 days, in the dark. After incubation, the fragments of tissue are collected and chopped with a scalpel blade on a glass slide, a drop of Lugol's iodine solution is added (stock Lugol's iodine solution: potassium iodide, 6.0 g; iodine, 4.0 g; dH_2O, 100 ml. Lugol's iodine working solution: dH_2O, 30.0 ml; Lugol's stock solution, 15.0 ml) and the preparation is covered with a cover-slip and allowed to sit for 10 minutes. The preparations are examined in the fresh state.

Whole body burden assay (*Fisher & Oliver, 1996*): the entire host, cut into 2–5 mm pieces, is placed in fluid thioglycollate culture medium and incubated as in the tissue assay above. The solution is centrifuged at 1500 ***g*** for 10 minutes and the supernatant is discarded. 2 M NaOH (20 ml g^{-1} tissue) is added and the solution is incubated at 60°C for 2–6 hours until tissue is digested. The solution is centrifuged at 1500 ***g*** for 10 minutes and the supernatant is discarded. The solution is washed three times in deionised water, the pellet is resuspended in 1 ml Lugol's iodine working solution, and the cells are counted. Serial dilutions may have to be made to reduce the total cell number to a manageable number.

Interpretation of results:

Cultured parasites enlarge from 2–10 to 20–70 µm during incubation. *Perkinsus* spp. cells are spherical and the walls stain blue or bluish-black with Lugol's iodine solution (Bushek *et al.,* 1994; Ray 1966).

In susceptible host species, within the known range of *P. marinus*, a positive result is presumptive evidence of *P. marinus* infection, but should be confirmed in accordance with Section 6.

4.10.2. Cell culture/artificial media

Perkinsus marinus cells are easily cultured in a variety of media (e.g. La Peyre *et al.,* 1993). Culture medium is usually inoculated with heart, haemolymph, or gill tissue. Comparisons of commercially available media have been made (Dungan & Hamilton, 1995) and growth was supported in all media, but was at a maximum in 1/1 DME (Dulbecco's Modified Eagle's)/Ham's F-12 medium.

5. Test(s) recommended for surveillance to demonstrate freedom in apparently healthy populations

Real-time PCR and eventually RFTM tissue or whole-body burden assays should be used for targeted surveillance to declare freedom from infection with *P. marinus*.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_5_1	**USA**	**Category:** editorial **Proposed amended texts:** Please see the proposed change in blue – "Real-time PCR and ~~eventually~~ RFTM tissue…". **Rationale:** We recommend the above change because the adjective "eventually" does not provide additional clarity. If the intent was to highlight that one test method should be used before another, then this should be clearly stated.	Agreed.
2.4.5_5_2	**Canada**	**Category:** Deletion **Proposed amended text:** Real-time PCR ~~and eventually RFTM tissue or whole-body burden assays~~ should be used for targeted surveillance to declare freedom from infection with *P. marinus*. **Rationale:** RFTM should not be used to demonstrate freedom from *Perkinsus* infection. The revised section 6.3. below indicates "Tissue and haemolymph RFTM assays have a limited sensitivity when infection intensity is below 1000 *Perkinsus* cells per g wet tissue (Bushek *et al*., 1994)." Therefore, use in low prevalence	Agreed and slightly modified the proposal.

		populations is unlikely to provide confidence of freedom.	

6. Corroborative diagnostic criteria

This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence of clinical signs (Section 6.2.) but does not evaluate whether the infectious agent is the cause of the clinical event.

The case definitions for a suspect and confirmed case have been developed to support decision making related to trade and confirmation of disease status at the country, zone or compartment level. Case definitions for disease confirmation in endemically affected areas may be less stringent. If a Competent Authority does not have the capability to undertake the necessary diagnostic tests it should seek advice from the appropriate WOAH Reference Laboratory, and if necessary, refer samples to that laboratory for confirmatory testing of samples from the index case in a country, zone or compartment considered free. There are currently no WOAH Reference Laboratory for *Perkinsus marinus*.

When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** n/a **Rationale:** As raised in previous mollusc disease chapters, we would welcome some guidance within this chapter on how these diagnostic criteria apply to species not officially assessed and listed as susceptible to *P. marinus*. Particularly if the criteria if positive results by two PCRs targeting different parts of the genome combined with sequencing are obtained from other species, in the absence of clinical disease or other evidence of infection e.g. ISH, RFTM, or histology. We note that in the current *P. marinus Manual* chapter, that the section for Corroborative diagnostic criteria specifies the criteria applies to known susceptible species within the known geographic range, and that results originating from outside the known geographic or host range should be referred to the reference laboratory. As there is currently no reference laboratory for *P. marinus*, some further guidance on how to interpret results, particularly when the more traditional evidence of infection such as ISH, RFTM, or histology is not available, would be useful. **Supporting evidence:** n/a	Noted, will be addressed in the future updates to the *Aquatic Manual.*

6.1. Apparently healthy animals or animals of unknown health status[1]

Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom.

2.4.5_6.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Apparently healthy animals or animals of unknown health status[1]	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.

1 For example transboundary commodities.

		Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Testing may also be carried out on transboundary commodities, at which time the health status of the animals from which they were derived may be of unknown. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom. **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	. .

6.1.1. Definition of suspect case in apparently healthy animals

The presence of infection with *P. marinus* shall be suspected if at least one of the following criteria is met:

i) Histopathological changes consistent with the presence of the pathogen or the disease

ii) Positive result by conventional PCR

iii) Positive result by real-time PCR

iv) Positive result by RFTM

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.1.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** 6.1.1. Definition of suspect case in apparently healthy animals or animals of unknown health status[1] The presence of infection with *P. marinus* shall be suspected if at least one of the following criteria is met:… **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.
2.4.5_6.1.1_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. marinus* shall be suspected if at least one of the following criteria is met: i) Histopathological changes consistent with the presence of the pathogen or the disease ii) Positive result by conventional PCR iii) Positive result by real-time PCR iv) Positive result by RFTM v) Positive result by *in-situ* hybridisation **Rationale:** We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this manual more comprehensive.	Disagreed: ISH is not used for screening populations. Histopathology would be the screening method with ISH as a confirmatory method provided it has suitable specificity .

		Supporting evidence: not relevant	

6.1.2. Definition of confirmed case in apparently healthy animals

The presence of infection with *P. marinus* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing

ii) Positive result by *In-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing

iii) Positive result by *In-situ* hybridisation and positive result by species specific real-time PCR.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.1.2_1	**UK**	**Category:** Editorial and Addition **Proposed amended text:** i) Positive result by real-time PCR and ~~positive result by~~ conventional PCR followed by amplicon sequencing, targeting two different genomic regions ii) Positive result by ~~i~~In-situ hybridisation and ~~positive result by~~ conventional PCR followed by amplicon sequencing iii) Positive result by ~~i~~In-situ hybridisation and ~~positive result by~~ species specific real-time PCR. **Rationale:** Suggested editorial changes to bring the text in alignment with the rest of the chapter. We also propose the additional text regarding the targeting of two different genomic regions, which is in line with the methodology described here.	Disagreed: this text is already in the introduction to Section 6.
2.4.5_6.1.2_2	**New Zealand**	**Category:** editorial **Proposed amended text:** Definition of confirmed case in apparently healthy animals or animals of unknown health status **Rationale:** Edited to make it explicit that criteria for confirmed case also applied to "animals of unknown health status" as described in 6.1.1. **Supporting evidence** n/a	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown. .
2.4.5_6.1.2_3	**EU**	**Category:** Addition **Proposed amended text:** The presence of infection with *P. marinus* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by histology or *in-situ* hybridisation or RFTM and positive result by PCR combined with sequencing ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing **Rationale:** alignment with the case definitions for *B ostreae, B exitiosa and M refringens*	Disagreed, the Commission agreed that case definitions for a confirmed case in either apparently healthy or clinically affected animals should recommend two specific methods, as is the current approach for other diseases. Molecular methods should either be combined with another diagnostic method or two molecular methods targeting non-overlapping regions of the pathogen genome should be used. Reworded the proposal.

2.4.5_6.1.2_4	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. marinus* is considered to be confirmed if at least one of the following critera is met: i) Positive result by species-specific real-time PCR and positive result by conventional PCR followed by amplicon sequencing ii) Positive result by species-specific *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing iii) Positive result by species-specific *in-situ* hybridisation and positive result by species specific real-time PCR. **Rationale:** One out of three real-time quantitative PCR tests and one out of two conventional PCR tests are not species-specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Agreed: see response to comment 2.4.5_6.1.2_3. .
2.4.5_6.1.2_5	**Australia**	**Category:** change **Proposed amended text:** 6.1.2 Definition of confirmed case in apparently healthy animals … iii) Positive result of real-time PCR followed by positive result by *in-situ* hybridisation ~~and positive result by species specific real-time PCR~~ **Rationale**: Not sure this is consistent with the requirements for a confirmed case (need sequence analysis). Should this be re-written as "Positive result of real-time PCR and followed by positive result by ISH" as modified in Section 6.2.2 ii below **Supporting evidence:** n/a	Agreed: see response to comment 2.4.5_6.1.2_3.

6.2 Clinically affected animals

Clinical signs are not pathognomonic for a single disease; however, they may narrow the range of possible diagnoses.

6.2.1. Definition of suspect case in clinically affected animals

The presence of infection with *P. marinus* shall be suspected if at least one of the following criteria is met:

i) Gross pathology or clinical signs associated with the disease as described in this chapter, with or without elevated mortality

ii) Histopathological changes consistent with the presence of the pathogen or the disease

iii) Positive result by conventional PCR

iv) Positive result by real-time PCR

v) Positive result by RFTM

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.2.1_1	**China (People's Rep. of)**	**Category:** change	Disagreed: ISH is not used for screening populations.

		Proposed amended text: The presence of infection with *P. marinus* shall be suspected if at least one of the following criteria is met: i) Gross pathology or clinical signs associated with the disease as described in this chapter, with or without elevated mortality ii) Histopathological changes consistent with the presence of the pathogen or the disease iii) Positive result by conventional PCR iv) Positive result by real-time PCR v) Positive result by RFTM vi) Positive result by *in-situ* hybridisation **Rationale:** We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this *Manual* more comprehensive. **Supporting evidence:** not relevant	Histopathology would be the screening method with ISH as a confirmatory method provided it has suitable specificity.

6.2.2. Definition of confirmed case in clinically affected animals

The presence of infection with *P. marinus* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by real-time PCR and conventional PCR followed by sequence analysis

ii) Positive result ISH and conventional PCR followed by sequence analysis

ii) Positive result of real-time PCR and ISH

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.2.2_1	**UK**	**Category:** Editorial and Addition **Proposed amended text:** i) Positive result by real-time PCR and conventional PCR followed by ~~sequence analysis~~ amplicon sequencing, targeting two different genomic regions ii) Positive result by in-situ hybridisation ~~ISH~~ and conventional PCR followed by ~~sequence analysis~~ amplicon sequencing iii) Positive result ~~of~~ by species specific real-time PCR and ~~ISH~~ in-situ hybridisation **Rationale:** Suggested editorial changes to bring the text in alignment with the rest of the chapter. We suggest changing 'sequence analysis' to 'amplicon sequencing' to improve clarity and specificity. We also propose the additional text regarding the targeting of two different genomic regions, which is in line with the methodology described here.	Disagreed: this text is already in the introduction to Section 6.
2.4.5_6.2.2_2	**EU**	**Category:** Addition **Proposed amended text:** The presence of infection with *P. marinus* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by histology or *in-situ* hybridisation or RFTM and positive result by PCR combined with sequencing	Partially agreed, the Commission agreed that case definitions for a confirmed case in either apparently healthy or clinically affected animals should recommend two specific methods, as is the current approach for other diseases. Molecular methods should either be combined with another diagnostic

		ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing **Rationale:** alignment with the case definitions for *B ostreae, B exitiosa and M refringens*	method or two molecular methods targeting non-overlapping regions of the pathogen genome should be used. Reworded the proposal. Disagreed on adding histology as it is not a confirmatory test. .
2.4.5_6.2.2_3	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. marinus* is considered to be confirmed if at least one of the following critera is met: i) Positive result by species-specific real-time PCR and conventional PCR followed by sequence analysis ii) Positive result by species-specific ISH and conventional PCR followed by sequence analysis ii) Positive result ~~of~~ by species-specific real-time PCR and ISH **Rationale:** Three out of five real-time quantitative PCR tests and two out of three conventional PCR tests are not species specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Agreed. .
2.4.5_6.2.2_4	**Australia**	**Category:** change **Proposed amended text:** 6.2.2 Definition of confirmed case in apparently healthy animals … iii) Positive result of real-time PCR ~~and~~ followed by positive result by ISH **Rationale**: For clarity of the operational order of conducting the diagnostic confirmation tests **Supporting evidence:** n/a	Agreed: see response to 2.4.5_6.2.2_2.

6.3. Diagnostic sensitivity and specificity for diagnostic tests

The diagnostic performance of tests recommended for surveillance or diagnosis of infection with *P. marinus* are provided in Tables 6.3.1. and 6.3.2 (no data are currently available for either). Data are only presented where tests are validated to at least level 2 of the validation pathway described in Chapter 1.1.2. and the information is available within published diagnostic accuracy studies.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.3_1	**Canada**	**Category:** addition **Proposed amended text:** Some data on analytical performances (stage 1 validation) are available for most diagnostic tests described in this chapter, but are not always complete (for example for inclusivity and exclusivity). RFTM assays and histopathology allow a diagnostic at the genus level and do not allow species discrimination. RFTM assays are considered to be specific for all members of the genus *Perkinsus* except *P. qugwadi*	Agreed.

		(incertae sedis). However, the apparent exclusivity of RFTM assays for detection of only *Perkinsus* species lacks exhaustive confirmation through testing of all other protists (Dungan & Reece, 2020). Molecular tools have been evaluated for their analytical sensitivity and specificity to varying extents. Diagnostic sensitivity (DSe) and specificity (DSp) (stage 2 validation) have generally not been estimated, however data comparing sensitivity performances of the different diagnostic tests are available. Histopathology is considered as less sensitive than PCR and RFTM assays. Whole body burden (WBB) RFTM assay is considered very sensitive, with a theoretical sensitivity of one *Perkinsus* sp. cell per host. Tissue and haemoplymph RFTM assays have a limited sensitivity when infection intensity is below 1000 *perkinsus* cells per g wet tissue (Bushek *et al.*, 1994). **Rationale:** The above information is included within the revised Chapter 2.4.5. Infection with *Perkinsus marinus* but is not specific to Perkinsus marinus. We propose that it should be included for *Perkinsus olseni* as well.	
2.4.5_6.3_2	**Australia**	**Category:** general **Proposed amended text**: n/a **Rationale:** Since "no data are currently available for either" Tables 6.3.1. and 6.3.2, in the absence of this evidence are claims of validation to Stage 2 in Table 4.1 justified? **Supporting evidence**: n/a	The Commission rechecked the literature and found that no assays have been validated to level 2;Table 4.1 has been amended accordingly. Tables 6.3.1 and 6.3.2. remain unpopulated.

Some data on analytical performances (stage 1 validation) are available for most diagnostic tests described in this chapter, but are not always complete (for example for inclusivity and exclusivity). RFTM assays and histopathology allow a diagnostic at the genus level and do not allow species discrimination. RFTM assays are considered to be specific for all members of the genus *Perkinsus* except *P. qugwadi* (incertae sedis). However, the apparent exclusivity of RFTM assays for detection of only *Perkinsus* species lacks exhaustive confirmation through testing of all other protists (Dungan & Reece, 2020). Molecular tools have been evaluated for their analytical sensitivity and specificity to varying extents. Diagnostic sensitivity (DSe) and specificity (DSp) (stage 2 validation) have generally not been estimated, however data comparing sensitivity performances of the different diagnostic tests are available. Histopathology is considered as less sensitive than PCR and RFTM assays. Whole body burden (WBB) RFTM assay is considered very sensitive, with a theoretical sensitivity of one *Perkinsus* sp. cell per host. Tissue and haemoplymph RFTM assays have a limited sensitivity when infection intensity is below 1000 *perkinsus* cells per g wet tissue (Bushek *et al.,* 1994).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.3_2	**Australia**	**Category:** general **Proposed amended text**: n/a **Rationale:** Since "Diagnostic sensitivity (DSe) and specificity (DSp) (stage 2 validation) have generally not been estimated", then claims of validation to Stage 2 in Table 4.1 are not justified. **Supporting evidence**: n/a	The Commission rechecked the literature and found that no assays have been validated to level 2;Table 4.1 has been amended accordingly. Tables 6.3.1 and 6.3.2. remain unpopulated.

6.3.1. For presumptive diagnosis of clinically affected animals (no data are currently available)

Test type	Test purpose	Source populations	Tissue or sample types	Species	DSe (*n*)	DSp (*n*)	Reference test	Citation

DSe = diagnostic sensitivity, DSp = diagnostic specificity, *n* = number of animals used in the validation study, PCR: = polymerase chain reaction.

6.3.2. For surveillance of apparently healthy animals (no data are currently available)

Test type	Test purpose	Source populations	Tissue or sample types	Species	DSe (*n*)	DSp (*n*)	Reference test	Citation

DSe = diagnostic sensitivity, DSp = diagnostic specificity, *n* = number of animals used in the validation study, PCR: = polymerase chain reaction.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.5_6.3_1	**Thailand**	**Category:** General (Clarification) We would like to request WOAH to recheck the Level of Validation (LV) in Table 4.1 for the real-time PCR method. If it is confirmed as LV 2, the DSe and DSp values should be specified in Section 6.3. However, if such data are currently unavailable, the LV in Table 4.1 should be revised accordingly **Proposed amended text:** No text proposed **Rationale:** To ensure accuracy **Supporting Document:** -	The Commission rechecked the literature and found that no assays have been validated to level 2;Table 4.1 has been amended accordingly. Tables 6.3.1 and 6.3.2. remain unpopulated.

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WINNICKI S.M., CARDEN W., HOLOHAN B., RALPH G., WARD E. & B. ALLAM (2008). Establishment of *Perkinsus marinus* infection in *Crassostrea virginica:* insights into the potrtal of entry and the potential role of marine aggregates. *J. Shellfish Res.*, **27**, 1064. (Abstract).

*
* *

NB: Currently (2025) there is no WOAH Reference Laboratory for infection with *Perkinsus marinus* (please consult the WOAH web site: https://www.woah.org/en/what-we-offer/expertise-network/reference-laboratories/#ui-id-3).

NB: FIRST ADOPTED IN 1995 AS PERKINSOSIS. MOST RECENT UPDATES ADOPTED IN 2024 (SECTIONS 2.2.1 AND 2.2.2).

CHAPTER 2.4.6.

INFECTION WITH *PERKINSUS OLSENI*

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_1	**EU**	**Category:** General The EU supports the proposed amendment. For the sake of harmonisation, the EU would encourage the Aquatic Animals Commission to provide the same level of detail notably for the case definitions as it is the case for *Bonamia exitiosa, Bonamia ostreae* or *Marteilia refringens*. Please, see specific comments including to the case definitions below. In addition, in accordance with worms-https://www.marinespecies.org/aphia.php?p=taxdetails&id=625984, *Perkinsus olseni* should now be written *P. olsenii* Finally, please see several comments to improve the readability of the chapter	Agreed: the Commission agreed to stay with the standard approach to retain two confirmatory diagnostic tests as presented in Table 4.1. for the case definitions. The Commission did not agree to change the name to P. olsenii as WORMS did not provide any rationale for the name change
2.4.6_2	**Australia**	**Category:** general **Proposed amended text:** n/a **Rationale**: For most of the mollusc chapters, in Table 4.1 assays are validated to Stage 2. This includes estimates of DSe and DSp which need to be published in peer reviewed literature and added to Table 6.3.1 and/or Table 6.3.2. If there is no data in these tables, there is no peer-reviewed evidence to support claims of validation to Stage 2 (or greater). Tables 6.3.1 and 6.3.2. were added to provide the reader with some evidence of the level of validation of recommended tests – if data exists but is unpublished there is the validation template that can be used. If the AAHSC do not require published estimates of validation to support claims of Stage 2 in Table 4.1 then everyone will just add Stage 2 which defeats the purpose of what the *ad hoc* group and AAHSC was trying to achieve. **Supporting evidence:** n/a	Noted and agreed.

1. Scope

Infection with *Perkinsus olseni* is considered to be infection with the pathogenic agent *Perkinsus. olseni* of the ~~(~~Family Perkinsidae~~)~~.

2. Disease information

2.1. Agent factors

2.1.1. Aetiological agent

The aetiological agent is the protozoan parasite *Perkinsus olseni*.

Current evidence using genomic sequencing data have placed *Perkinsus olseni* in the class Perkinsea of the phylum Perkinsozoa, and Kingdom Alveolata (Reece *et al.,* 1997). *Perkinsus olseni* was formerly known as *Perkinsus atlanticus* in Europe (Azevedo 1989).

As trophozoites grow, cleavage furrows begin to form on the cell surface, signaling the early stages of their subdivision, or the onset of schizogony (Gajamange *et al.,* 2020). As cleavage advances, the number of daughter cells increases, progressing to the morula stage, where approximately 100 irregularly shaped

merozoites are observed. Once schizonts are fully developed, they release hundreds of merozoites upon the rupture or abrasion of the schizont cellular membrane.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.1.1_1	**EU**	**Category:** editorial It should be clarified that the observations in the paragraph above are from *in vitro* culture	Agreed. .

A study investigating genome-wide comparison of five *P. olseni* isolates from Australia (00978-12), New Zealand (PRA 205 and 207), Japan (PRA-179), and Spain (PRA-31) observed differences between different geographical locations. Indeed, *P. olseni* from Oceania (Australia and New Zealand) displayed a high heterozygosity of 5.47–7.71% compared with Eurasia (Spain and Japan) with 0.1 to 0.2% of heterozygosity (Bogema *et al.,* 2021). The gene numbers differed substantially, with the Oceanian isolates sharing a high proportion of unique orthogroups in comparison to the Eurasian isolates (Bogema *et al.,* 2021). The Oceanian isolates had a slightly higher proportion of repetitive content such as tandem gene duplication. The characterised gene contents appear to be highly conserved across *Perkinsus* species such as in *P. olseni*, *P. marinus*, and *P. chesapeaki* (Bogema *et al.,* 2021).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.1.1_2	**EU**	**Category:** editorial The references into brackets to the *P. olseni* isolates should be deleted. **Rationale:** The article referred to above (Bogema *et al.*, 2021) presents five draft *Perkinsus olseni* genomes (obtained from five isolates specified into brackets). These genomes are not the not final versions and might need further validation. Therefore, it is premature to include the isolate codes.	Agreed. .

The ribosomal RNA transcription unit is highly duplicated with variations between the isolates from different parts of the world, which does not make it a suitable region for designing real-time PCR tests to assess the intensity of infection (Bogema *et al.,* 2021).

More studies are needed to confirm the ploidy of *P. olseni*. Indeed, some authors using genome-wide data suggested that *Perkinsus* is polyploid with a ploidy variation between individual cells and populations (Bogema *et al.,* 2021) whereas other using microsatellite loci or restriction fragment length polymorphism proposed a diploidy (Pardo *et al.,* 2011; Reece *et al.,* 1997; 2001; Robledo *et al.,* 1999; Thompson *et al.,* 2011).

2.1.2. Survival and stability in processed or stored samples

Perkinsus olseni can be propagated *in vitro* in various media formulations such as Dulbecco's Modified Eagle medium and Ham's F-12 nutrient mixture (Burreson *et al.,* 2005; Dungan & Reece, 2006) or JL-ODR-2A (La Peyre *et al.,* 2006). Both media are supplemented with various salts, FBS, lipid mixtures, and yeast ultrafiltrates. Isolates can be cryopreserved and stored indefinitely (Dungan et al., 2007).

2.1.3. Survival and stability outside the host

Long-term survival of *P. olseni* outside its bivalve host is not known. but it is at least 4 months for prezoosporangia (Casas *et al.,* 2002b).

2.2. Host factors

2.2.1. Susceptible host species

Species that fulfil the criteria for listing as susceptible to infection with *Perkinsus olseni* according to Chapter 1.5. of the *Aquatic Animal Health Code* (*Aquatic Code*) are:

Family	Scientific name	Common name
Arcidae	*Anadara kagoshimensis*	half-crenated ark
	Anadara trapezia	no common name
Cardiidae	*Tridacna crocea*	crocus giant clam
Haliotidae	*Haliotis laevigata*	greenlip abalone
	Haliotis rubra	blacklip abalone
	Haliotis iris	black paua
Margaritidae	*Pinctada fucata*	Japanese pearl oyster
Mytilidae	*Mytilus galloprovincialis*	Mediterranean mussel
	Perna canaliculus	New Zealand mussel
Veneridae	*Austrovenus stutchburyi*	Stutchbury's venus
	Leukoma jedoensis	Jedo venus
	Paratapes undulatus	undulate venus
	Protapes gallus	rooster venus
	Proteopitar patagonicus	no common name
	Ruditapes decussatus	grooved carpet shell
	Ruditapes philippinarum	Japanese carpet shell

2.2.2. Species with incomplete evidence for susceptibility

Species for which there is incomplete evidence to fulfil the criteria for listing as susceptible to infection with *P. olseni* according to Chapter 1.5. of the *Aquatic Code* are:

Family	Scientific name	Common name
Cardiidae	*Cerastoderma edule*	common edible cockle
Mytilidae	*Mytilus chilensis*	Chilean mussel
Ostreidae	*Crassostrea gasar*	gasar cupped oyster
	Ostrea angasi	Australian mud oyster
Pectinidae	*Pecten novaezelandiae*	New Zealand scallop
Psammobiidae	*Hiatula acuta*	no common name
Veneridae	*Venerupis corrugata*	corrugated venus clam

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.2.2_1	**Perú**	**Categoría:** Adición **Texto modificado propuesto:** Species for which there is incomplete evidence to fulfil the criteria for listing as susceptible to infection with *P. olseni* according to Chapter 1.5. of the Aquatic Code are: *Paphia undulata* **Justificación:** Se reporta la presencia de Perkinsus olseni en ejemplares de Paphia undulata a través de inmunohistoquimica	Disagreed. According to WORMS, *Paphia undulata* is not the accepted taxonomic assignment. The accepted name is *Paratabes undulatus,* which is in the Table in 2.2.1

		Evidencia documentada: Kaewsalabnil S., Buewkeaw A., Watanachote J., Leethochavalit S. & Khongchareonporn N. (2024). Anti Perkinsus olseni Monoclonal Antibody Generation Using Hypnospores as Antigens. *Curr. Appl. Sci. Tech*., e0257223-e0257223. https://doi.org/10.55003/cast.2023.257223	

In addition, pathogen-specific positive polymerase chain reaction (PCR) results have been reported in the following species, but no active infection has been demonstrated:

Family	Scientific name	Common name
Cardiidae	*Cerastoderma glaucum*	olive green cockle
Chamidae	*Chama pacifica*	reflexed jewel box
Haliotidae	*Haliotis diversicolor*	small abalone
Isognomonidae	*Isognomon alatus*	flat tree oyster
	Isognomon sp.	N/A
Margaritidae	*Pinctada imbricata*	Atlantic pearl oyster
Ostreidae	*Crassostrea rhizophorae*	mangrove cupped oyster
	Dendostrea frons	Frons oyster
	Magallana [syn. *Crassostrea*] *gigas*	Pacific oyster
	Magallana [syn. *Crassostrea*] *hongkongensis*	no common name
	Saccostrea sp.	N/A
Pectinidae	*Mimachlamys crassicostata*	noble scallop
Pharidae	*Sinonovacula constricta*	constricted tagelus clam
Veneridae	*Meretrix lyrata*	lyrate hard clam
	Polititapes aureus	golden carpet shell
	Venus verrucosa	warty venus

2.2.3. Likelihood of infection by species, host life stage, population or sub-populations

Perkinsus olseni is known to infect and cause clinical signs in many bivalve species. All stages after settlement are susceptible. *P. olseni* infection intensity increases with host age (Villalba *et al.,* 2005).

2.2.4. Distribution of the pathogen in the host

Perkinsus olseni trophozoites and schizonts can be found in various tissues and organs, including connective tissues, siphons, mantle, gills, digestive system, foot, adductor muscle, and haemolymph (Carella *et al.,* 2023; Park & Choi, 2001; Leethochavalit *et al.,* 2004). In some clams, the infection was heaviest in the digestive gland and gills (Leethochavalit *et al.,* 2004; Park & Choi, 2001). However, infection in the gills of bivalves is representative of the infection throughout the entire body and is therefore considered the organ of choice for diagnosing *P. olseni* infection (Choi *et al.,* 2002; Cui *et al.,* 2018; Dang *et al.,* 2013; Park *et al.,* 1999). *Perkinsus olseni* has also been detected in faeces of infected animals (Park *et al.,* 2010).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.2.4_1	**EU**	**Category:** editorial An introduction explaining that distribution might depend on the host species is needed	Disagreed, the paragraph is general and does not provide specifics for any species.

		Rationale: *Perkinsus olseni* can infect a wide range of species and its distribution depends on the host. As we cannot provide details for all the species, an introduction is needed.	

2.2.5. Aquatic animal reservoirs of infection

None known.

2.2.6. Vectors

None known.

2.3. Disease pattern

2.3.1. Mortality, morbidity and prevalence

Infections in ~~clam hosts~~ shellfish can be lethal depending on environmental conditions, and death may occur 1 or 2 years after infection. Studies have suggested that the impact on the host depends on the intensity of infection but also on host species and locations; for example, in clam hosts it can start being deleterious and lethal at densities of 10^6 parasite cells/g wet tissue (Dang *et al.,* 2013; Waki *et al.,* 2018).

Prevalence is highly variable depending on host and environmental conditions, up to 100% in susceptible host species as determined by histology or polymerase chain reaction (PCR). Prevalence and intensity of infection may be higher in individuals with more than 1 year of exposure to the pathogen because they have been exposed to *P. olseni* for longer, and larger animal have a higher filtration rate (Choi *et al.,* 2002; Villalba *et al.,* 2005). Prevalence can vary according to the season and is often higher in spring (Cui *et al.,* 2018; Villalba *et al.,* 2005).

2.3.2. Clinical signs, including behavioural changes

Perkinsus olseni infection can lead to reduced growth, reproduction issues, and mass mortalities in shellfish populations (Cho & Park, 2010). Clinical signs include mantle retraction, byssal tissue sloughing, gaping or dead molluscs but these clinical signs are not specific to infection with *P. olseni.* Individual bivalves with late-stage infections may exhibit slow responses to stimuli (Sheppard & Phillips, 2008).

2.3.3 Gross pathology

Gross signs are thin, watery tissue, pale digestive gland and nodules in several tissues such as mantle, gills, and foot of some hosts, but these signs are not specific to infection with *P. olseni*.

At high infection intensities the infected molluscs can present several milky-white cysts, abscesses or brown nodules (Azevedo, 1989; Gudkovs *et al.,* 2016; Ruano *et al.,* 2015). The cysts or nodules contain individual and grouped encapsulated trophozoites at different stages of maturation, and result from a natural defensive reaction, the infiltration of hemocytes (Abdel-Baki *et al.,* 2014).

2.3.4. Modes of transmission and life cycle

The life cycle is horizontal, direct from host to host (Villalba *et al.,* 2004). Faecal discharge and decomposition of infected tissues after the host death have been suggested as two transmission pathways for *P. olseni* (Cui *et al.,* 2018; Park *et al.,* 2010).

The life cycle consists of three main life stages: trophozoite, prezoosporangium, and zoospore (Villalba *et al.,* 2004). The trophozoite is a stage occurring in the tissues of the live host where vegetative proliferation occurs. The trophozoite undergoes successive bipartitioning to yield up to 32 daughter cells that stay together in a rosette-like arrangement inside a wall. After rupture of the wall, immature trophozoites enlarge and form a vacuole, becoming mature trophozoites. At the host death, trophozoites evolve into a prezoosporangia, which evolve into zoosporangia when they are released in the environment. Zoosporangia produce zoospores, which are the infective stage.

2.3.5. Environmental factors

Environmental factors such as temperature, salinity, oxygen levels, pH, and nutrient availability influence *Perkinsus* infection dynamics and hypnospore formation (2013; Villalba *et al.,* 2004).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.3.5_1	**EU**	**Editorial:** A reference is missing (2013; Villalba *et al.,* 2004). We suggest to include Deslile *et al.,* 2025 (https://doi.org/10.1016/j.ijpara.2025.04.011) **Rationale:** Deslile *et al.,* 2025 provides further information in another host species, *Perna canaliculus*	Agreed.

Temperature and salinity appear to be the most important environmental factors controlling the transmission of *P. olseni* infection with zoosporulation occurring between 19 and 28°C and increasing with higher temperature and salinity (Kyoung & Ki, 2001; Umeda *et al.,* 2020).

Temperature and salinity also appear to be the two major environmental factors influencing the prevalence and seasonality of *P. olseni* infection, with several studies reporting higher prevalence and intensity of infection in spring (Park & Choi 2001; Soudant *et al.,* 2013; Villalba *et al.,* 2005; Waki & Yoshinaga, 2013; 2015).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.3.5_2	**Perú**	**Categoría:** Adición **Texto modificado propuesto:** …The development of *P. olseni* increased significantly above 20°C and reached an optimal level at 22°C. At 35°C, *Perkinsus* proliferation was considerably reduced, suggesting that the upper thermal limit of *P. olseni* is 30°C < T ≤ 35°C. **Justificación:** Se reporta un estudio que evidencia que la temperatura tuvo un efecto significativo en la propagación del parásito. El desarrollo de *P. olseni* aumentó significativamente por encima de los 20°C y alcanzó un nivel óptimo a 22°C. A 35°C, la proliferación de *Perkinsus* se redujo considerablemente, lo que sugiere que el límite térmico superior de *P. olseni* es 30°C < T ≤ 35°C. **Evidencia documentada:** Delisle L., Bui T., Copedo J., Laroche O., von Ammon U., Lane H. S. & Hutson K.S. (2025). Isolation, culture, and optimal growth conditions for the shellfish protozoan parasite, *Perkinsus olseni*. *Int. J. Parasitol.,* https://doi.org/10.1016/j.ijpara.2025.04.011	Agreed and slightly modified the proposed text.

2.3.6. Geographical distribution

Infections are widespread throughout the tropical Pacific Ocean, Oceania, Asia, Europe and South America (Cremonte *et al.,* 2005; Goggin & Lester, 1995; Villalba *et al.,* 2004). *Perkinsus olseni* is not known from North America.

See WAHIS (https://wahis.woah.org/#/home) for recent information on distribution at the country level.

2.4. Biosecurity and disease control strategies

2.4.1. Vaccination

None.

2.4.2. Chemotherapy including blocking agents

Cyclohexamide, pyrimethamine, deferoxamine (DFO) and 2, 2-bipyridyl inhibit *P. olseni* development/replication *in vitro*, and DFO inhibits *P. olseni* development/infection *in vivo*. (Elandalloussi *et al.,* 2005).

2.4.3. Immunostimulation

None.

2.4.4. Breeding resistant strains

None.

2.4.5. Inactivation methods

Isolated *P. olseni* cells were killed when immersed in freshwater within 10 minutes at room temperature. Another study showed that free zoospores, free prezoosporangia and prezoosporangia in gill tissues were killed after 1 hour incubation with 50, 200, and 3000 ppm of chlorine, respectively (Casas *et al.,* 2002b). *Perkinsus olseni* cells in host tissue were much more resistant to these treatments. UV-C irradiation has been shown to be effective in inactivating *Perkinsus* parasites, with a minimum dose of 94 mJ/cm^2 required to inhibit proliferation and 450 mJ/cm^2 to completely kill all parasites (Fernandez-Boo *et al.,* 2021).

2.4.6. Disinfection of eggs and larvae

Perkinsus olseni is not known to infect eggs or larvae of its hosts, but parasite cells may occur intercellularly.

2.4.7. General husbandry

Management strategies to mitigate perkinsosis impacts include modifying culture procedures such as reducing stocking density, selective breeding for resistant strains, and using triploid or allochthonous oyster species (Villalba *et al.,* 2004).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_2.4.7_1	**New Zealand**	**Category:** general **Proposed amended text:** n/a **Rationale:** "None" is declared under 2.4.4 'Breeding resistant strains', while "selective breeding for resistant strains" is mentioned under 2.4.7 'General husbandry'. These sections seem to be at odds with each other and need reviewing. **Supporting evidence:** n/a	Agreed: deleted the text 'selective breeding for resistant strains,'

3. Specimen selection, sample collection, transportation and handling

This section draws on information in Sections 2.2, 2.3 and 2.4 to identify populations, individuals and samples that are most likely to be infected.

3.1. Selection of populations and individual specimens

Bivalves that are gaping, ~~clams~~ that are found at the sediment surface ~~instead of being~~ when they would normally be buried, or freshly dead animals should be targeted to increase the chances of finding infected animals. Abalone with foot blisters should be sampled. For histology, only live animals should be taken.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_3.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Bivalves that are gaping, ~~clams~~ that are found at the sediment surface ~~instead of being~~ when they would normally be buried, and/or freshly dead animals should be targeted to increase the chances of ~~finding~~ detecting infected animals. Abalone with foot blisters should be sampled. For histology, only live animals should be taken. **Rationale:** Suggesting edits to make the sentence clearer.	Agreed to change to 'detecting'. The term 'and/or' not accepted as 'or' includes the concept of 'and'.

		Supporting evidence: n/a	

3.2. Selection of organs or tissues

Connective tissue of all organs, and haemocytes.

For Ray's fluid thioglycollate medium (RFTM), the whole animal should be ideally sampled if its size allows. If not, pieces of gill followed by mantle or foot for abalone are typically used.

For culture purposes, tissue slices that can include gill, mantle, or foot (for abalone) can be cultured.

For histology, a 5 mm thick section through the visceral mass that includes digestive gland, gill and mantle is used.

For PCR, gill or mantle tissue is recommended.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_3.2_1	**Canada**	**Category:** addition Canada requests the reference(s) for this tissue type selection be included for Member information.	Disagreed: the information is a summary from multiple published sources and Reference Laboratory experience.

3.3. Samples or tissues not suitable for pathogen detection

Rectal tissue is not reliable for PCR assays because of the presence of inhibitors.

3.4. Non-lethal sampling

For *P. marinus*, Gauthier & Vasta (1995) proposed haemolymph incubation in RFTM as an alternative to the traditional tissue RFTM, enabling non-lethal sampling of individuals. This method was further refined by Nickens *et al.* (2002). Rodriguez & Navas (1995) reviewed and compared various RFTM assays for *Perkinsus olseni*, highlighting that whole host body incubation in RFTM is the most sensitive method.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_3.4_1	**Canada**	**Category:** addition **Proposed amended text:** …This method was further refined by Nickens *et al.* (2002). Rodriguez & Navas (1995) reviewed and compared various RFTM assays for *Perkinsus olseni*, highlighting that whole host body incubation in RFTM is the most sensitive method. Diagnostic sensitivity and specificity is not available for these test methods. **Rationale:** We are proposing an additional sentence to this section to address the lack of information available on Diagnostic Sensitivity and specificity for testing non-lethal samples. This information should be captured within the manual for Members knowledge however, it is not appropriate to add this tests information to tables 4.1., 6.3.1. or 6.3.2. because these non-lethal samples are not captured in Section 3.2. as appropriate samples for RFTM testing.	Agreed and slightly amended the proposed sentence.

3.5. Preservation of samples for submission

For guidance on sample preservation methods for the intended test methods, see Chapter 2.4.0 General information (diseases of molluscs).

3.5.1. Samples for pathogen isolation

The results of bioassay depend strongly on the quality of samples (time since collection, time in storage, preservative used). Fresh specimens should be kept on ice and preferably sent to the laboratory within 24 hours of collection. To avoid degradation of samples, use alternative storage methods only after consultation with the receiving laboratory.

3.5.2. Preservation of samples for molecular detection

Tissue samples for PCR testing should be preserved in 80% (v/v) analytical-grade ethanol. The recommended ratio of ethanol to tissue is 10:1 based on studies in terrestrial animal and human health. The use of lower grade (laboratory or industrial grade) ethanol is not recommended. If material cannot be fixed it may be frozen.

Standard sample collection, preservation and processing methods for molecular techniques can be found in Section B.5.5 of Chapter 2.4.0 *General information* (diseases of molluscs).

3.5.3. Samples for histopathology, immunohistochemistry or *in-situ* hybridisation

Standard sample collection, preservation and processing methods for histological techniques can be found in Section B.5.3 of Chapter 2.4.0 *General information* (diseases of molluscs).

3.5.4. Samples for other tests

None.

3.6. Pooling of samples

Pooling of samples from more than one individual animal for a given purpose is only recommended where robust supporting data on diagnostic sensitivity and diagnostic specificity have been evaluated and found to be suitable. If the effect of pooling on diagnostic sensitivity has not been thoroughly evaluated, larger specimens should be processed and tested individually. Small life stages such as spat can be pooled to obtain the minimum amount of material for molecular detection. Pooling of very small spat (5–10 depending on size) is acceptable for PCR analyses.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_3.6_1	**Canada**	**Category:** deletion **Proposed amended text:** …Small life stages such as spat can be pooled to obtain the minimum amount of material for molecular detection. ~~Pooling of very small spat (5–10 depending on size) is acceptable for PCR analyses.~~ **Rationale:** Canada has completed a scientific evaluation on the usefulness of small (<6mm) life stages of molluscs for testing of spat for disease freedom which found a gap in the literature of validated studies using spat below 6mm for PCR testing. This evaluation found that there was not enough evidence to support the use of spat smaller than 6mm in sampling protocols. If the pathogenic agent cannot be found in small spat, or that that test methods are not sensitive enough to detect low prevalence infection in spat, these are not appropriate samples. Therefore, Canada questions the inclusion of pooling of very small spat as pooling would further impact the sensitivity and confidence in detection rates. The evidence provided for *P. marinus* can be extrapolated to *P. olseni*. **Supporting evidence:** Suitability of testing of early life stages of *Crassostrea virginica* (larvae to 5 mm spat) by PCR-based assays for the detection of *Haplosporidium nelsoni, Mikrocytos mackini, Perkinsus marinus,* and *Bonamia exitiosa*.	Agreed.

4. Diagnostic methods

The methods currently available for pathogen detection that can be used in i) surveillance of apparently healthy animals, ii) presumptive diagnosis in clinically affected animals and iii) confirmatory diagnostic purposes are listed in Table 4.1. by animal life stage.

Ratings for purposes of use. For each recommended assay a qualitative rating for the purpose of use is provided. The ratings are determined based on multiple performance and operational factors relevant to application of an assay for a defined purpose. These factors include appropriate diagnostic performance characteristics, level of assay validation, availability cost, timeliness, and sample throughput and operability. For a specific purpose of use, assays are rated as:

+++ = Methods are most suitable with desirable performance and operational characteristics.

++ = Methods are suitable with acceptable performance and operational characteristics under most circumstances.

+ = Methods are suitable, but performance or operational characteristics may limit application under some circumstances.

Shaded boxes = Not appropriate for this purpose.

Validation stage. The validation stage corresponds to the assay development and validation pathway in chapter 1.1.2. The validation stage is specific to each purpose of use. Where available, information on the diagnostic performance of recommended assays is provided in Section 6.3.

WOAH Reference Laboratories welcome feedback on diagnostic performance of recommended assays, in particular PCR methods. Of particular interest are any factors affecting expected assay sensitivity (e.g. tissue components inhibiting amplification) or expected specificity (e.g. failure to detect particular genotypes, detection of homologous sequences within the host genome). These issues should be communicated to the WOAH Reference Laboratories so that advice can be provided to diagnostic laboratories and the standards amended if necessary.

Table 4.1. WOAH recommended diagnostic methods and their level of validation for surveillance of apparently healthy animals and investigation of clinically affected animals

Method	A. Surveillance of apparently healthy animals				B. Presumptive diagnosis of clinically affected animals				C. Confirmatory diagnosis[1] of a suspect result from surveillance or presumptive diagnosis			
	Early life stages[2]	Juveniles[2]	Adults	LV	Early life stages[2]	Juveniles[2]	Adults	LV	Early life stages[2]	Juveniles[2]	Adults	LV
Wet mounts												
Histopathology		++	++	2		+++	+++	NA				
Cell culture												
Transmission electron microscopy												
Real-time PCR	+++	+++	+++	3	+++	+++	+++	NA	+++	+++	+++	3
Conventional PCR	~~++~~+	~~++~~+	~~++~~+	~~3~~ 1	+++	+++	+++	NA				
Conventional PCR followed by amplicon sequencing									+++	+++	+++	~~3~~ 1
In-situ hybridisation										~~++~~+	~~++~~+	~~3~~ 1
Bioassay												
LAMP												
Ab-ELISA												
Ag-ELISA												
RFTM		+++	+++	3		+++	+++	~~3~~ NA				

LV = level of validation, refers to the stage of validation in the WOAH Pathway (chapter 1.1.2); PCR = polymerase chain reaction; LAMP = loop-mediated isothermal amplification; Ab- or Ag-ELISA = antibody or antigen enzyme-linked immunosorbent assay, respectively; RRTM = Ray's fluid thioglycollate medium

[1]For confirmatory diagnoses, methods need to be carried out in combination (see Section 6). [2]Susceptibility of early and juvenile life stages is described in Section 2.2.3.

[3]Specify the test used. Shading indicates the test is inappropriate or should not be used for this purpose.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_Table4.1_1	**Thailand**	**Category:** General (Clarification) We would like to ask for clarification on the rationale for reducing the Level of Validation (LV) of the Conventional PCR method in Table 4.1 from LV 3 to LV 1. **Proposed amended text:** No text proposed **Rationale:** – **Supporting Document:** –	The LV level was reduced as there were no validation data available to support a LV 3.
2.4.6_Table4.1_2	**EU**	Table 4.1 and information presented in section "6.3. Diagnostic sensitivity and specificity for diagnostic tests" should be consistent. **Rationale:** Level of validation of histopathology for the surveillance of healthy animals (LV 2 in the table 4.1) is not documented in section 6.3. Level of validation of real-time PCR assays (LV 3 in the table 4.1) is only documented for one *P. olseni* real-time assay (Rios *et al.,* 2020) and not for the other five real-time PCR assays.	Agreed: could not find such references and so amended the level of validation in Table 4.1.
2.4.6_Table4.1_3	**Canada**	**Category:** deletion **Proposed amended text:** Please remove the levels of validation for early life stages in A, B and C or clarify that early life stages must be >6mm due to a lack of validated studies using spat below 6mm for PCR testing. **Rationale:** We note that Section 2.2.3 does not indicate susceptible of early life stages or suitability of early life stages for testing. At what size 'early life stage oysters' transition from seed to juveniles. Canada has completed a scientific evaluation on the usefulness of small (<5mm) life stages of molluscs for testing of spat for disease freedom which found a gap in the literature of validated studies using spat below 6mm for PCR testing. This evaluation found that there was not enough evidence to support the use of spat smaller than 6mm in sampling protocols. If the pathogenic agent cannot be found in small spat, or that that test methods are not sensitive enough to detect low prevalence infection in spat, these are not appropriate samples. **Supporting evidence:** Suitability of testing of early life stages of Crassostrea virginica (larvae to 5 mm spat) by PCR-based assays for the detection of *Haplosporidium nelsoni, Mikrocytos mackini, Perkinsus marinus,* and *Bonamia exitiosa*.	Agreed and amended the scores in Table 4.1.
2.4.6_Table4.1_4	**Canada**	**Category:** General Canada thanks the Commission for responding to previous comments to indicate that DSe and Dsp are not always published but may be provided by the Reference Laboratory to support the ratings provided within Table 4.1 and therefore DSe and DSp cannot be included within Tables 6.1.2 and 6.1.3.	Noted for future updates, when relevant. At present, the validation study template has been used for one chapter only: Infection with yellow head virus genotype 1.

		When there is no published information on diagnostic sensitivity in the scientific literature and the rating is based on unpublished reference laboratory validation studies, we would request that a footnote including an explanation for the discrepancy between the rating and the published literature would enhance transparency and clarity for Table 4.1.	

4.1. Smears

Not recommended as a diagnostic method.

4.2. Histopathology

Samples to be taken consist of live or moribund bivalves.

Sections of tissues that include gills, digestive gland and mantle should be fixed for a minimum of 24 hours in a recommended fixative followed by standard processing for histology as described in Section 5.3 of Chapter 2.4.0 *General information* (diseases of molluscs). Observations are made at increasing magnifications up to ×400.

Fixed sections reveal large multifocal lesions in connective tissue containing *P. olseni* cells. Haemocyte infiltration (haemocytosis) occurs in most infections. In clam hosts, *P. olseni* cells are often encapsulated by a thick layer of eosinophilic material derived from haemocyte degranulation (Villalba *et al.,* 2004).

The occurrence of spherical, uninucleate cells ranging from approximately 5 to 15 µm in diameter with a large vacuole and an eccentrically displaced nucleus with a prominent nucleolus, may indicate infection with *Perkinsus olseni*. Multinucleate schizonts (dividing forms) often accompany the uninucleate trophozoites. Cells may be phagocytosed by host haemocytes Cells may be phagocytosed by host haemocytes. *Perkinsus olseni* cells stain lightly basophilic.

4.3. Electron microscopy

Ultrastructural data show that the lysis of haemocytes and coalescence of metachromatic granules result in the nodule that encapsulates trophozoites (Sagrista *et al.,* 1995).

4.4. Nucleic acid amplification

Samples to be taken consist of live or freshly dead molluscs. 2–3 mm^2 tissue pieces are excised aseptically from gill and mantle and placed into 1.5 ml microcentrifuge tubes containing 80% ethanol. Dissecting utensils should be flamed between samples to prevent cross-contamination.

PCR assays should always be run with the controls specified in Section B.5.5 *Molecular methods* Chapter 2.4.0 *General information* (diseases of molluscs). Each sample should be tested in duplicate.

Extraction of nucleic acids

DNA is extracted by proteinase K digestion overnight at 56°C and the spin-column methodology using commercially available kits. Different kits and procedures can be used for nucleic acid extraction. The quality and concentration of the extracted nucleic acid is important and can be checked using a suitable method as appropriate to the circumstances.

4.4.1. Real-time PCR

A real-time *Perkinsus* genus PCR assay targeting the ITS region has been developed for use with host tissue (Gauthier *et al.,* 2006). It has been tested only with *P. marinus*, *P. olseni* and *P. chesapeaki*, and was shown to be more sensitive in a limited validation against the RFTM assay. This assay needs to be tested more thoroughly for specificity, but may be useful for laboratories that possess the necessary equipment.

Several real-time PCR assays all targeting the ITS region have been developed to detect and quantify *P. olseni* (Cui *et al.,* 2018; Gajamange *et al.,* 2011; Itoïz *et al.,* 2021; Ríos *et al.,* 2020; Umeda & Yoshinaga, 2012). One needs to be careful when attempting to quantify *P. olseni* as genome sequencing revealed that this region is highly duplicated,

which does not make it the ideal region for quantifying parasite infection. Of all those assays, only the assay developed by Ríos *et al.* (2020) presented some level of validation such as sensitivity and reproducibility. More validation is required before a real-time PCR assay can be recommended (see chapter 1.1.2 *Principles and methods of validation of diagnostic assays for infectious diseases*). In addition, Ríos *et al.* (2020) detected non-specific hybridisation when using the Umeda & Yoshinaga (2012) and Gajamange *et al.* (2011) assays.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_4.4.1_1	**Australia**	**Category:** general **Proposed amended text:** n/a **Rationale**: As indicated in the text of section 4.4.1 above: "Of all those assays, only the assay developed by Ríos *et al.* (2020) presented some level of validation such as sensitivity and reproducibility. More validation is required before a real-time PCR assay can be recommended". Then Table 4.1 cannot claim assays are validated to Stage 3. **Supporting evidence:** n/a	Agreed and amended the text. .

***Primers and probes* (*sequences*)**

Pathogen/ target gene	Primer/probe (5'–3')	Concentration	Cycling parameters[a]
Method 1: TaqMan PCR Gauthier *et al.,* 2006; GenBank Accession No.: AF149876.1			
Perkinsus sp./ITS (detects at least *P. marinus, P. olseni* and *P. chesapeaki*).	PERK-F: CGT-GAA-CCA-GTA-GAA-ATC-TCA-A PERK-R: ACA-TAT-CAG-TGT-CGC-TCT-TCT-TCC Probe: GCA-TAC-TGC-ACA-AAG-GG	0.9 µM 0.9 µM 0.25 µM	40 cycles of: 95°C/15 sec and 60°C/60 sec
Method 2: TaqMan PCR Itoiz *et al.,* 2021; GenBank Accession No.: MW187111; amplicon size: 76 bp			
P. olseni / ITS 2	PolsITS2-F: CAC-CAC-AAC-ACA-GTC-GGA-C PolsITS2-R: CGT-ATT-GTA-GCC-CCT-CCG-A PolsITS2-probe: GAC-ACT-CAC-AGG-CGC-GGT-CC	0.2 µM 0.2 µM 0.5 µM	45 cycles of: 95°C/10 sec and 55°C/20 sec
Method 3: SYBR Green PCR Rios *et al.,* 2020; GenBank Accession No.[b]: ~~???~~; amplicon size 346 bp			
P. olseni / ITS	Perk-ITS-qF1: CTG-ACC-GCC-TTA-ACG-GGC Perk-ITS-qR2: CTA-TCT-CCG-AAG-AGT-TAG-TCC-	10 µM 10 µM	40 cycles of: 95°C/30 sec and 60°C/60 sec
Method 4: Cui *et al*., 2018; GenBank Accession No.: ~~???~~ AF369975; amplicon size 217 bp			
P. olseni / ITS	PO-F: GAG-TGT-CTC-TGG-TTG-CTC-GCA PO-R: ACA-TCA-GGC-CTT-CTA-ATG-ATG	10 µM 10 µM	40 cycles of: 95°C/15 sec and 60°C/60 sec
Method 6: Umeda &Yoshinaga, 2012; GenBank Accession No.~~???~~[b]			
P. olseni / ITS	OF: CTT-AAC-GGG-CCG-TGT-TA OR: CAT-AAC-GAA-CTA-TCT-CCG-AAG	0.5 µM 0.5 µM	40 cycles of: 98°C/2 sec and 60°C/5 sec
Method 5: Gajamange *et al.,* 2011; GenBank Accession No.: ~~???~~ AF473840, amplicon size: 97 bp			
P. olseni / 5.8S & ITS2	PK-ITS-F: CAG-AAT-TCC-GTG-AAC-CAG-TAG-A PK-ITS-R: TGT-CGC-TCT-TCT-TCC-CGA-TA Probe: TCA-ACG-CAT-ACT-GCA-CAA-AGG-GGA	10 pM 10 pM 10 pM	40 cycles of: 95°C/10 sec and 56°C/30 sec

[a]A denaturation step prior to cycling has not been included.

[b]For GenBank Accession numbers see original publications

<table>
<tr><th>Reference</th><th>Member</th><th>Comment</th><th>Aquatic Animals Commission response</th></tr>
<tr><td>2.4.6_4.1.1_1</td><td>China (People's Rep. of)</td><td>Category: Change
<table>
<tr><th>Pathogen/ target gene</th><th>Primer/probe (5'–3')</th><th>Concentration</th><th>Cycling parameters[a]</th></tr>
<tr><td colspan="4">Method 1: TaqMan PCR Gauthier et al., 2006; GenBank Accession No.: AF149876.1; amplicon size: 86 bp</td></tr>
<tr><td>Perkinsus sp./ITS (detects at least P. marinus, P. olseni and P. chesapeaki).</td><td>PERK-F: CGT-GAA-CCA-GTA-GAA-ATC-TCA-A
PERK-R: ACA-TAT-CAG-TGT-CGC-TCT-TCT-TCC
Probe: GCA-TAC-TGC-ACA-AAG-GG
PERK-F: TCC-GTG-AAC-CAG-TAG-AAA-TCT-CAA-C
PERK-R: GGA-AGA-AGA-GCG-ACA-CTG-ATA-TGT-A
Probe: (FAM)-CCC-TTT-GTG-CAG-TAT-GC-MGB</td><td>0.9 µM
0.9 µM
0.25 µM</td><td>40 cycles of: 95°C/15 sec and 60°C/60 sec</td></tr>
<tr><td colspan="4">Method 2: TaqMan PCR Itoiz et al., 2021; GenBank Accession No.: MW187111; amplicon size: 76 bp</td></tr>
<tr><td>P. olseni / ITS 2 ITS 2</td><td>PolsITS2-F: CAC-CAC-AAC-ACA-GTC-GGA-C
PolsITS2-R: CGT-ATT-GTA-GCC-CCT-CCG-A
PolsITS2-probe: GAC-ACT-CAC-AGG-CGC-GGT-CC</td><td>0.2 µM
0.2 µM
0.5 µM</td><td>45 cycles of: 95°C/10 sec and 55°C/20 sec</td></tr>
<tr><td colspan="4">Method 3: SYBR Green PCR Rios et al., 2020; GenBank Accession No.[b]; ???; amplicon size 346 bp</td></tr>
<tr><td>P. olseni / ITS ITS</td><td>Perk-ITS-qF1: CTG-ACC-GCC-TTA-ACG-GGC
Perk-ITS-qR2: CTA-TCT-CCG-AAG-AGT-TAG-TCC-</td><td>10 µM
10 µM</td><td>40 cycles of: 95°C/30 sec and 60°C/60 sec</td></tr>
<tr><td colspan="4">Method 4: Cui et al., 2018; GenBank Accession No.: ??? AF369975; amplicon size 217 bp</td></tr>
<tr><td>P. olseni / ITS ITS</td><td>PO-F: GAG-TGT-CTC-TGG-TTG-CTC-GCA
PO-R: ACA-TCA-GGC-CTT-CTA-ATG-ATG</td><td>10 µM
10 µM</td><td>40 cycles of: 95°C/15 sec and 60°C/60 sec</td></tr>
<tr><td colspan="4">Method 65: Umeda &Yoshinaga, 2012; GenBank Accession No. ???[b]</td></tr>
<tr><td>P. olseni / ITS ITS</td><td>OF: CTT-AAC-GGG-CCG-TGT-TA
OR: CAT-AAC-GAA-CTA-TCT-CCG-AAG</td><td>0.5 µM
0.5 µM</td><td>40 cycles of: 98°C/2 sec and 60°C/5 sec</td></tr>
<tr><td colspan="4">Method 56: Gajamange et al., 2011; GenBank Accession No.: ??? AF473840, amplicon size: 97 bp</td></tr>
<tr><td>P. olseni / 5.8S & ITS2 5.8S & ITS2</td><td>PK-ITS-F: CAG-AAT-TCC-GTG-AAC-CAG-TAG-A
PK-ITS-R: TGT-CGC-TCT-TCT-TCC-CGA-TA
Probe: TCA-ACG-CAT-ACT-GCA-CAA-AGG-GGA</td><td>10 pM
10 pM
10 pM</td><td>40 cycles of: 95°C/10 sec and 56°C/30 sec</td></tr>
</table>
Rationale:
Updated the table to correct errors as listed below based on the source literature.
1) After the publication of the paper by Gauthier et al., 2006, a correction notice was issued, modifying the primer and probe sequences. This modification has been</td><td>Agreed.</td></tr>
</table>

		adopted in 'Chapter 2.4.5. Infection with *Perkinsus marinus*' and the relevant information here needs to be updated. The amplification product size has been added. 2) ITS, ITS2, and 5.8S should not be italicised. 3) Method 5 and Method 6 are reversed.	

4.4.2. Conventional PCR

Pathogen/ target gene	Primer/probe (5'–3')	Concentration	Cycling parameters[a]
Method 1: Moss *et al.,* 2006; GenBank Accession No.: ~~???~~ AF369975; amplicon size: [bp] ???			
P. olseni	PolsITS-140F: GAC-CGC-CTT-AAC-GGG-CCG-TGT-T PolsITS-600R: GGR-CTT-GCG-AGC-ATC-CAA-AG 450 bp	0.1 µM 0.1 µM	40 cycles of: 94°C/60 sec and 62°C/60 sec and 65°C/180 sec
Method 2: Casas *et al.,* 2002a; GenBank Accession No.: ~~???~~ AF369975; amplicon size: 655 bp			
Perkinsus spp. (except *P. qugwadi*)	PerkITS-85: CCG-CTT-TGT-TTG-GAT-CCC PerkITS-750: ACA-TCA-GGC-CTT-CTA-ATG-ATG 675bp product	0.1 µM 0.1 µM	40 cycles of: 95°C/1 min and 55°C/1 min and 72°C/1 min

[a]A denaturation step prior to cycling has not been included.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_4.1.1_1	**China (People's Rep. of)**	**Category:** Change **Rationale:** (table below) Updated the table to correct errors as listed below based on the source literature. 1) Method 1: The amplification product size for section 4.4.2 has been added. In accordance with the new formatting requirements, the "450bp" below the primer sequences has been deleted, and an extension step after PCR cycling has been added. 2) Method 2: The primer pair PerkITS-85 and PerkITS-750 has also been adopted in "Chapter 2.4.5. Infection with *Perkinsus marinus*". The details here have been revised to align with the content in section 2.4.5, including the amplification	Partially agree, Table 4.4.1. and 4.4.2. do not include information on final elongation parameters.

Pathogen/ target gene	Primer/probe (5'–3')	Concentration	Cycling parameters[a]
Method 1: Moss *et al.,* 2006; GenBank Accession No.: ~~???~~ AF369975; amplicon size: 457 bp ~~[bp]~~ ~~???~~			
P. olseni	PolsITS-140F: GAC-CGC-CTT-AAC-GGG-CCG-TGT-T PolsITS-600R: GGR-CTT-GCG-AGC-ATC-CAA-AG ~~450 bp~~	0.1 µM 0.1 µM	40 cycles of: 94°C/60 sec and 62°C/60 sec and 65°C/180 sec Final elongation 65°C/10 min
Method 2: ~~Method 1~~: Audemard *et al.*, 2004 (primers from Casas *et al.*, 2002); GenBank Accession No.: AF149876.1, amplicon size: 666 bp **~~Casas *et al.,* 2002a; GenBank Accession No.: ??? ; amplicon size: bp~~**			
Perkinsus spp. (except *P. qugwadi*)	PerkITS-85: CCG-CTT-TGT-TTG-GAT-CCC PerkITS-750: ACA-TCA-GGC-CTT-CTA-ATG-ATG ~~675bp product~~	~~0.1~~0.5 µM ~~0.1~~0.5 µM	40 cycles of: 95°C/1 min and 55°C/1 min and 72°C/1 min Final elongation 72°C/10 min

		conditions. According to the new formatting requirements, the "675bp" below the primer sequences has been deleted.	

4.4.3. Other nucleic acid amplification methods

A PCR-restriction fragment length polymorphism (RFLP) assay has been developed that may be useful for specific diagnoses of *P. olseni* (Abollo *et al.,* 2006), although it has not been tested for specificity against all known *Perkinsus* species.

A LAMP assay was developed by (Feng *et al.,* 2013) targeting ITS-2 and appeared to be rapid, sensitive (detection limit of 10 copies of plasmid DNA), and specific for *Perkinsus* spp. detection.

4.5. Amplicon sequencing

The size of the PCR amplicon should be verified, for example by agarose gel electrophoresis. Both DNA strands of the PCR product must be sequenced and analysed in comparison with reference sequences.

4.6. *In-situ* hybridisation

Samples to be taken consist of live or freshly dead molluscs.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_4.6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** ~~Samples to be taken consist of~~ Live or freshly dead molluscs should be targeted for sampling. **Rationale:** Edited sentence as it appeared incomplete. **Supporting evidence:** n/a	Agreed.

A specific DNA probe that targets the small subunit rRNA gene has been developed for the genus *Perkinsus*: Perksp700DIG (Elston *et al.,* 2004) and a DNA probe that targets the LSU of the rRNA gene has been developed for the specific detection of *P. olseni:* PolsLSU-464DIG (Moss *et al.,* 2006). Muznebin *et al.* (2023) also used a DIG labelled probe using primers targeting the ITS gene region.

Pathogen/target	ISH probe type	ISH probe (5'-3')	Reference
Perkinsus sp./SSU (detects all known *Perkinsus* species *except P. qugwadi)*	Labelled oligonucleotide	PerkspSSU-700-CGC-ACA-GTT-AAG-TRC-GTG-RGC-ACG	Elston *et al.* (2004)
Perkinsus olseni/LSU	Labelled oligonucleotide	PolsLSU-464DIG-CTC-ACA-AGT-GCC-AAA-CAA-CTG	Moss *et al.* (2006);
Perkinsus olseni/ITS	Labelled primers	Pols140F-GAC-CGC-CTT-AAC-GGG-CCG-TGT-T and PolsITS-600R-GGR-CTT-GCG-AGC-ATC-CAA-AG-3	Muznebin *et al.,* (2023)

~~***Perkinsus* genus assays (PCR and *in situ* hybridisation)~~

~~For *in situ* hybridisation (ISH), probes have been developed that target the small subunit (SSU) of the rRNA gene complex (Elston *et al.,* 2004).~~

~~***Perkinsus* genus-specific *in situ* hybridisation~~

~~Samples to be taken: follow the procedure for 'fixed sections' above, except that tissue sections must be placed on positively charged glass slides or slides coated with 3-aminopropyl-triethoxylane, without staining. Deparaffinise~~

sections in xylene for 10 minutes and then rehydrate in an alcohol series. Wash sections twice for 5 minutes in phosphate-buffered saline (PBS).

A specific DNA probe that targets the small subunit rRNA gene has been developed for the genus *Perkinsus* (Elston *et al.*, 2004): Perksp700DIG (5′ CGC ACA GTT AAG TRC GTG RGC ACG 3′). The probe should be 5′ end-labelled with digoxigenin.

The tissue sections are treated with 125 µg ml−1 pronase in PBS, at 37°C for 30 minutes. The reaction is then stopped by washing the sections in PBS with 0.2% glycine for 5 minutes. The sections are then placed in 2× SSC (standard saline citrate; 20× SSC = 3 M NaCl; 0.3 M Na-citrate; pH 7.0) for 10 minutes.

The sections are prehybridised for 1 hour at 42°C in prehybridisation solution (4× SSC, 50% formamide, 5× Denhardt's solution, 0.5 mg ml−1 yeast tRNA, and 0.5 mg ml−1 heat-denatured salmon sperm DNA) in a humid chamber.

The prehybridisation solution is then replaced with prehybridisation buffer containing 7 ng µl−1 of the digoxigenin-labelled *Perkinsus* genus probe. The sections are covered with in-situ hybridisation plastic cover-slips and placed on a heating block at 90°C for 12 minutes. The slides are then cooled on ice for 1 minute before hybridisation overnight at 42°C in a humid chamber.

The sections are washed twice for 5 minutes each in 2× SSC at room temperature, twice for 5 minutes each in 1× SSC at room temperature, and twice for 10 minutes each in 0.5× SSC at 42°C. The sections are then placed in Buffer 1 (100 mM Tris, pH 7.5, 150 mM NaCl) for 1–2 minutes.

The sections are placed in Buffer 1 (see above) supplemented with 0.3% Triton X-100 and 2% sheep serum for 30 minutes. Anti-digoxigenin alkaline phosphatase antibody conjugate is diluted 1/500 (or according to the manufacturer's recommendations) in Buffer 1 supplemented with 0.3% Triton X-100 and 1% sheep serum and applied to the tissue sections. The sections are covered with in-situ hybridisation cover-slips and incubated for 3 hours at room temperature in a humid chamber.

The slides are washed twice in Buffer 1 for 5 minutes each and twice in Buffer 2 (100 mM Tris, pH 9.5, 100 mM NaCl, 50 mM $MgCl_2$) for 5 minutes each. The slides are then placed in colour development solution (337.5 µg ml−1 nitroblue tetrazolium, 175 µg ml−1 5-bromo-4-chloro-3-indolylphosphate p-toluidine salt, 240 µg ml^{-1} levamisole in Buffer 2) for 2 hours in the dark. The colour reaction is stopped by washing in TE buffer (10 mM Tris, pH 8.0, 1 mM EDTA [ethylene diamine tetra-acetic acid]).

The slides are then rinsed in sterile distilled water (dH_2O). The sections are counterstained with Bismarck Brown Y, rinsed in dH_2O, and cover-slips are applied using an aqueous mounting medium.

Positive/negative controls: these are compulsory. Positive controls are tissue sections from any Perkinsus sp. infected mollusc. Negative controls are either no-probe assays or assays with uninfected oysters.

***Perkinsus olseni* specific *in situ* hybridisation**

The probe should be end-labelled with digoxygenin. The ISH procedures are the same as for the *Perkinsus* genus probe presented above.

Positive/negative controls: these are compulsory. Positive controls are tissue sections from any susceptible host infected with *P. olseni*. Negative controls are either no-probe assays or assays with uninfected oysters.

A DNA probe that targets the LSU of the rRNA gene of *P. olseni* has been developed (Mo*ss et al.,* 2006) (PolsLSU-464DIG 5′ CTC ACA AGT GCC AAA CAA CTG 3′).

Muznebin *et al.,* (2023) also used ISH using the DIG PCR probe synthesis kit (Roche). A DIG-labelled probe was generated using the PCR primers Pols140F (5′ GAC CGC CTT AAC GGG CCG TGT T 3′) and PolsITS 600R (5′ GGR CTT GCG AGC ATC CAA AG 3′) (Mo*ss et al.,* 2006). The DIG-labelled probe was used at a concentration of 5 ng/µl.

4.7. Immunohistochemistry

Not available.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_4.7_1	**Perú**	**Categoría:** Adición **Texto modificado propuesto:** ~~Not available.~~ The use of monoclonal antibodies is reported for the detection of *Perkinsus olseni* trophozoites in *Paphia undulata* in Thailand (Kaewsalabnil *et al*., 2024) **Justificación:** Se reporta el uso de anticuerpos monoclonales para la detección de trofozoitos de *Perkinsus olseni* en *Paphia undulata* en Tailandia. **Evidencia documentada:** Kaewsalabnil S., Buewkeaw A., Watanachote J., Leethochavalit S. & Khongchareonporn N. (2024). Anti Perkinsus olseni Monoclonal Antibody Generation Using Hypnospores as Antigens. *Curr. Appl. Sci. Tech*., e0257223-e0257223. https://doi.org/10.55003/cast.2023.257223	Agreed and amended the proposed text. .

4.8. Bioassay

Not available.

4.9. Antibody- or antigen-based detection methods (ELISA, etc.)

Not currently available for diagnostic purposes but monoclonal antibodies have been developed (Hanrio et al., 2021; 2022; Park et al., 2010;).

4.10. Other methods

4.10.1. Ray's fluid thioglycollate culture method (RFTM)

Incubation in thioglycollate is routinely used for surveillance of *P. olseni.* The technique is simple, inexpensive and very sensitive, but not species-specific. Trophozoites of *P. olseni* in host tissue will enlarge when cultured for at least 5 days in fluid thioglycollate medium containing dextrose that is supplemented with antibiotics (penicillin, streptomycin) and an antifungal compound (nystatin) to reduce bacterial and fungal growth. When the tissue is macerated after culture to allow penetration of aqueous iodine solution (Lugol's), the enlarged trophozoites (hypnospores or prezoosporangia in the old terminology) readily take up Lugol's and they easily become visible at low power because of their generally bluish–black coloration and their spherical shape.

Samples to be taken consist of live or freshly dead molluscs.

Tissue assay (Ray, 1966): tissue samples measuring approximately 5–10 mm are excised giving preference to rectal, gill and mantle tissue from oysters and clams, and adductor or foot muscles or mantle for abalone, and placed in test tubes containing thioglycollate medium (thioglycollate medium containing dextrose 14.6 g; NaCl, 10.0 g; sterile distilled water (dH_2O), 485 ml). A total of 9.5 ml is dispensed into disposable test tubes, which are autoclaved for 15 minutes at 1.2 kg cm^{-2} pressure. The autoclaved solution can be stored in tubes for up to 3 weeks. Dissecting utensils should be rinsed in 95% ethanol and flamed between hosts to prevent carry-over. The recommended antifungal/antibiotics are: 500 units ml^{-1} penicillin G and 500 units ml^{-1} dihydro-streptomycin in media (penicillin, 3.13 g; streptomycin, 6.55 g; 500 ml dH_2O; freeze in 50 ml aliquots; add 0.5 ml to each tube), and 50 µl of mycostatin (nystatin) per tube. Chloromycetin can be used in place of penicillin/streptomycin. The tube is plugged with a foam rubber or cotton stopper. Incubation is at 22–25°C for between 5 and 7 days, in the dark. After incubation, the fragments of tissue are collected and chopped with a scalpel blade on a glass slide, a drop of Lugol's iodine solution is added (stock Lugol's iodine solution: potassium iodide, 6.0 g; iodine, 4.0 g; dH_2O, 100 ml. Lugol's iodine working solution: dH_2O, 30.0 ml; Lugol's stock solution, 15.0 ml) and the preparation is covered with a cover-slip and allowed to sit for 10 minutes. The preparations are examined in the fresh state.

Whole body burden assay (Fisher & Oliver, 1996): the entire host, cut into 2–5 mm pieces, is placed in fluid thioglycollate culture medium and incubated as in the tissue assay above. If host organisms are too large to use the entire host, then selected target tissue can be used. The solution is centrifuged at 1500 ***g*** for 10 minutes and the supernatant is discarded. 2 M NaOH (20 ml g^{-1} tissue) is added and the solution is incubated at 60°C for 2–6 hours until tissue is digested. The solution is centrifuged at 1500 ***g*** for 10 minutes and the supernatant is discarded. The solution is washed three times in deionised water, the pellet is resuspended in 1 ml Lugol's iodine working solution, and the cells are counted. Serial dilutions may have to be made to reduce the total cell number to a manageable number.

Interpretation of results:

Cultured parasites enlarge from 2–10 to 20–70 µm during incubation. *Perkinsus* spp. cells are spherical and the walls stain blue or bluish-black with Lugol's iodine solution (Bushek *et al.*, 1994; Ray 1966).

In susceptible host species, within the known range of *P. olseni*, a positive result is presumptive evidence of *P. olseni* infection, but should be confirmed in accordance with Section 6.

5. Test(s) recommended for surveillance to demonstrate freedom in apparently healthy populations

Real-time PCR and RFTM tissue or whole-body burden assays are recommended for targeted surveillance to declare freedom from infection with *P. olseni*.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_5_1	**Canada**	**Category:** Deletion **Proposed amended text:** Real-time PCR ~~and RFTM tissue or whole-body burden assays~~ ~~is~~ recommended for targeted surveillance to declare freedom from infection with *P. olseni*. **Rationale:** RFTM should not be used to demonstrate freedom from *Perkinsus* infection. Section 6.3. of the revised Chapter 2.4.5. Infection with *Perkinsus marinus* indicates "Tissue and haemolymph RFTM assays have a limited sensitivity when infection intensity is below 1000 *Perkinsus* cells per g wet tissue (Bushek *et al.*, 1994)." Therefore, use in low prevalence populations is unlikely to provide confidence of freedom.	Agreed and slightly modified the proposal (kept RFTM as scores Table 4.1). .

6. Corroborative diagnostic criteria

This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence of clinical signs (Section 6.2.) but does not evaluate whether the infectious agent is the cause of the clinical event.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence (Section 6.2.) of clinical signs ~~(Section 6.2.)~~ but does not evaluate whether the infectious agent is the cause of the clinical event. **Rationale:** Edited so that reference to section is consistent. **Supporting evidence:** n/a	Agreed. . .

The case definitions for a suspect and confirmed case have been developed to support decision making related to trade and confirmation of disease status at the country, zone or compartment level. Case definitions for disease confirmation in endemically affected areas may be less stringent. If a Competent Authority does not have the capability to undertake the

necessary diagnostic tests it should seek advice from the appropriate WOAH Reference Laboratory, and if necessary, refer samples to that laboratory for confirmatory testing of samples from the index case in a country, zone or compartment considered free. There are currently no WOAH Reference Laboratory for *Perkinsus olseni*.

When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6_2	**New Zealand**	**Category:** general comment **Proposed amended text:** n/a **Rationale:** As raised in the *Perkinsus marinus* chapter, we would welcome some guidance within this chapter on how these diagnostic criteria apply to species not officially assessed and listed as susceptible to *P. olseni*. Particularly if positive results by two PCRs targeting different parts of the genome combined with sequencing are obtained from other species, in the absence of clinical disease or other evidence of infection e.g. ISH, RFTM, or histology. We note that in the current *P. olseni* Manual Chapter, that the section for Corroborative diagnostic criteria specifies the criteria applies to known susceptible species within the known geographic range, and that results originating from outside the known geographic or host range should be referred to the reference laboratory. As there is currently no reference laboratory for *P. olseni*, some further guidance on how to interpret results, particularly when the more traditional evidence of infection such as ISH, RFTM, or histology is not available, would be useful. **Supporting evidence** n/a	Noted, will be addressed in the future updates to the *Aquatic Manual.*

6.1. Apparently healthy animals or animals of unknown health status[1]

Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Apparently healthy animals or animals of unknown health status[1] Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Testing may also be carried out on transboundary commodities, at which time the health status of the animals from which they were derived may be of unknown. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom. **Rationale:**	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown.

1 For example transboundary commodities.

		As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	

6.1.1. Definition of suspect case in apparently healthy animals

The presence of infection with *P. olseni* shall be suspected if at least one of the following criteria is met:

i) Histopathological changes consistent with the presence of the pathogen or the disease

ii) Positive result by conventional PCR

iii) Positive result by real-time PCR

iv) Positive result by RFTM

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6.1.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** 6.1.1. Definition of suspect case in apparently healthy animals or animals of unknown health status[1] The presence of infection with *P. olseni* shall be suspected if at least one of the following criteria is met:… **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown.
2.4.6_6.1.1_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. olseni* shall be suspected if at least one of the following criteria is met: i) Histopathological changes consistent with the presence of the pathogen or the disease ii) Positive result by conventional PCR iii) Positive result by real-time PCR iv) Positive result by RFTM v) Positive result by *in-situ* hybridisation **Rationale:** We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this *Manual* more comprehensive. **Supporting evidence:** not relevant	Disagreed: ISH is not used for screening populations. Histopathology would be the screening method with ISH as a confirmatory method provided it has suitable specificity.

6.1.2. Definition of confirmed case in apparently healthy animals

The presence of infection with *P. olseni* is considered to be confirmed if at least one of the following ~~criterion~~ criteria is met:

i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing

ii) Positive result by *In-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing

iii) Positive result by *In-situ* hybridisation and positive result by species specific real-time PCR.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6.1.2_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Definition of confirmed case in apparently healthy animals or animals of unknown health status **Rationale:** Edited to make it explicit that criteria for confirmed case also applied to "animals of unknown health status" as described in 6.1.1. **Supporting evidence:** n/a	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown.
2.4.6_6.1.2_2	**EU**	**Category:** Addition **Proposed amended text:** The presence of infection with *P. olsenii s* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by histology or *in-situ* hybridisation or RFTM and positive result by PCR combined with sequencing ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing **Rationale:** alignment with the case definitions for *B ostreae, B exitiosa and M refringens*	Agreed and slightly modified the proposal.
2.4.6_6.1.2_3	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. olsenii* is considered to be confirmed if at least one of the following critera is met: i) Positive result by species-specific real-time PCR and positive result by conventional PCR followed by amplicon sequencing ii) Positive result by species-specific *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing iii) Positive result by species-specific *in-situ* hybridisation and positive result by species specific real-time PCR. **Rationale:** One out of six real-time quantitative PCR tests, one out of two conventional PCR tests, and one out of two *in-situ* hybridisation methods are not species-specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Agreed: see response to comment 2.4.6_6.1.2_2.
2.4.6_6.1.2_4	**Australia**	**Category:** change **Proposed amended text:** The presence of infection with *P. olseni* is considered to be confirmed if at least one of the following ~~criterion~~ criteria is met: i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing ii) Positive result by *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing iii) Positive result of real-time PCR followed by positive result by *in-situ* hybridisation ~~and positive result by real-time PCR~~	Agreed: see response to comment 2.4.6_6.1.2_2. .

		Rationale: Change to reflect the correct operational order of using these testing methods, as real-time PCR is not specific, whereas In-situ hybridisation (ISH) is specific and used for confirmation testing, so ISH is used following positive real-time PCR results. It does not make sense to undertake non-specific PCR testing if you already have specific positive test results by ISH. **Supporting evidence:** n/a	

6.2 Clinically affected animals

Clinical signs are not pathognomonic for a single disease; however, they may narrow the range of possible diagnoses.

6.2.1. Definition of suspect case in clinically affected animals

The presence of infection with *P. olseni* shall be suspected if at least one of the following criteria is met:

i) Gross pathology or clinical signs associated with the disease as described in this chapter, with or without elevated mortality

ii) Histopathological changes consistent with the presence of the pathogen or the disease

iii) Positive result by conventional PCR

iv) Positive result by real-time PCR

v) Positive result by RFTM

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6.2.1_1	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. olseni* shall be suspected if at least one of the following criteria is met: i) Gross pathology or clinical signs associated with the disease as described in this chapter, with or without elevated mortality ii) Histopathological changes consistent with the presence of the pathogen or the disease iii) Positive result by conventional PCR iv) Positive result by real-time PCR v) Positive result by RFTM vi) Positive result by *in-situ* hybridisation **Rationale:** We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this *Manual* more comprehensive. **Supporting evidence:** not relevant	Disagreed: ISH is not used for screening populations. Histopathology would be the screening method with ISH as a confirmatory method provided it has suitable specificity.

6.2.2. Definition of confirmed case in clinically affected animals

The presence of infection with *P. olseni* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by real-time PCR and conventional PCR followed by sequence analysis

ii) Positive result ISH and conventional PCR followed by sequence analysis

ii) Positive result of real-time PCR and ISH

2.4.6_6.2.2_1	**EU**	**Category:** Addition	Partially agreed,the Commission agreed

		Proposed amended text: The presence of infection with *P. olsenii s* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by histology or *in-situ* hybridisation or RFTM and positive result by PCR combined with sequencing ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing **Rationale:** alignment with the case definitions for *B ostreae, B exitiosa and M refringens*	that case definitions for a confirmed case in either apparently healthy or clinically affected animals should recommend two specific methods, as is the current approach for other diseases. Molecular methods should either be combined with another diagnostic method or two molecular methods targeting non-overlapping regions of the pathogen genome should be used. Reworded the proposal.
2.4.6_6.2.2_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *P. olseni* is considered to be confirmed if at least one of the following critera is met: i) Positive result by species-specific real-time PCR and conventional PCR followed by sequence analysis ii) Positive result by species-specific ISH and conventional PCR followed by sequence analysis ii) Positive result ~~of~~ by species-specific real-time PCR and ISH **Rationale:** One out of six real-time quantitative PCR tests, one out of two conventional PCR tests, and one out of two *in-situ* hybridisation methods are not species-specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Agreed. .
2.4.6_6.2.2_2	**Australia**	**Category:** change **Proposed amended text:** The presence of infection with *P. olseni* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing. ii) Positive result by *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing. iii) Positive result of real-time PCR followed by positive result by *in-situ* hybridisation (ISH) **Rationale**: For clarity and consistency with comment above for 6.1.2 iii) **Supporting evidence:** n/a	Agreed: see response to comment 2.4.6_6.2.2_1.

6.3. Diagnostic sensitivity and specificity for diagnostic tests

The diagnostic performance of tests recommended for surveillance or diagnosis of infection with *P. olseni* are provided in Tables 6.3.1. (no data are currently available) and 6.3.2 ~~(no data are currently available for either)~~. Data are only presented where tests are validated to at least level 2 of the validation pathway described in Chapter 1.1.2. and the information is available within published diagnostic accuracy studies.

6.3.1. For presumptive diagnosis of clinically affected animals (no data are currently available)

Test type	Test purpose	Source populations	Tissue or sample types	Species	DSe (*n*)	DSp (*n*)	Reference test	Citation

DSe = diagnostic sensitivity, DSp = diagnostic specificity, *n* = number of animals used in the validation study, PCR: = polymerase chain reaction.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.6_6.3.1_1	**Australia**	**Category:** general	Agreed.
		Proposed amended text: n/a.	
		Rationale: Text above indicates "no data are currently available" however, Table 4.1 states assays are validated to Stage 3 so estimates of DSe and DSp must be provided in Table 6.3.1 if this LV is to be retained.	
		Supporting evidence: –	

6.3.2. For surveillance of apparently healthy animals ~~(no data are currently available)~~

Test type	Test purpose	Source populations	Tissue or sample types	Species	DSe (*n*)	DSp (*n*)	Reference test	Citation
Real-time PCR	Targeted surveillance	Wild populations	Gill tissue Haemolymph	*Ruditapes philippinarum*	100 79	50 12.5	RFTM	Rios *et al.*, 2020

DSe = diagnostic sensitivity, DSp = diagnostic specificity, *n* = number of animals used in the validation study, PCR: = polymerase chain reaction; RFTM = Ray's fluid thioglycollate medium.

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* *

NB: Currently (2025) there is no WOAH Reference Laboratory for infection with *Perkinsus olseni* (please consult the WOAH web site: https://www.woah.org/en/what-we-offer/expertise-network/reference-laboratories/#ui-id-3).

NB: FIRST ADOPTED IN 1995 AS PERKINSOSIS. MOST RECENT UPDATES ADOPTED IN 2015.

CHAPTER 2.4.7

INFECTION WITH *XENOHALIOTIS CALIFORNIENSIS*

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_1	**EU**	**Category:** General The EU supports the proposed amendments. For the sake of harmonisation, the EU would encourage the Aquatic Animals Commission to provide the same level of detail notably for the case definitions as it is the case for *Bonamia exitiosa, Bonamia ostreae* or *Marteilia refringens*. Please see below comments to the case definitions.	Noted.
2.4.7_2	**Australia**	**Category:** general **Proposed amended text:** n/a **Rationale**: For most of the mollusc chapters, in Table 4.1 assays are validated to Stage 2. This includes estimates of DSe and DSp which need to be published in peer-reviewed literature and added to Table 6.3.1 and/or Table 6.3.2. If there are no data in these tables, there is no peer-reviewed evidence to support claims of validation to Stage 2 (or greater). Tables 6.3.1 and 6.3.2. were added to provide the reader with some evidence of the level of validation of recommended tests – if data exists but is unpublished there is the validation template that can be used. If the AAHSC do not require published estimates of validation to support claims of Stage 2 in Table 4.1 then everyone will just add Stage 2 which defeats the purpose of what the ad hoc group and AAHSC was trying to achieve. **Supporting evidence:** n/a	Noted.

1. Scope

Infection with *Xenohaliotis californiensis* means infection with the pathogenic agent *Candidatus* Xenohaliotis californiensis of the Family Anaplasmataceae. For the purposes of this chapter, the pathogenic agent will be referred to as *Xenohaliotis californiensis.*

2. Disease information

2.1. Agent factors

2.1.1. Aetiological agent

Xenohaliotis californiensis is an intracellular bacterium in the family Anaplasmataceae (Dumler *et al.,* 2001) and is closely related to members of the genera *Ehrlichia, Anaplasma* and *Cowdria* (Friedman *et al.,* 2000). The disease caused by this bacterium is known as withering syndrome (Friedman *et al.,* 2002; Haaker *et al.,* 1992) and may be more appropriately termed abalone rickettsiosis. Some *X. californiensis* may be infected with a phage (Friedman & Crosson, 2012). The dimorphic rod-to-spherical-shaped bacterium measures an average of 332 × 1550 nm in the bacillus form and an average of 1405 nm in the spherical morphotype. The bacterium reproduces within intracytoplasmic vacuoles 14–56 µm in diameter within gastrointestinal epithelia (Friedman *et al.,* 2000).

Reference	Member	Comment	Aquatic Animals Commission response

2.4.7_2.1.1_1	**China (People's Rep. of)**	**Category:** Change **Proposed amended text :** *Xenohaliotis californiensis* is an intracellular bacterium in the ~~family~~ Family Anaplasmataceae (Dumler *et al.,* 2001) and is closely related to members of the ~~genera~~ Genera *Ehrlichia, Anaplasma* and *Cowdria* (Friedman *et al.,* 2000). **Rationale:** When used as taxonomic ranks, the first letters of "Family" and "Genera" should be capitalised.	Disagreed: these words do not need to be capitalised. .

2.1.2. Survival and stability in processed or stored samples

As the pathogen has not been cultured the survival and stability in stored samples is unknown.

2.1.3. Survival and stability outside the host

Although *X. californiensis* is thought to be an obligate intracellular organism, the bacterium may survive outside the host for ~~an undetermined period of time~~ as evidenced by water-borne transmission studies (Balseiro *et al.*, 2006; Braid *et al.*, 2005; Friedman *et al.*, 2002; 2007; Rosenblum *et al.*, 2008).

~~For inactivation methods, see Section 2.4.5.~~

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_2.1.3_1	**New Zealand**	**Category:** General **Proposed amended text:** n/a **Rationale:** Suggest adding specific information on how long the bacterium may survive in point 2.1.3, if it is available, as it would be useful information **Supporting evidence:** n/a	Noted for future updates.

2.2. Host factors

2.2.1. Susceptible host species

Species that fulfil the criteria for listing as susceptible to infection with *Xenohaliotis californiensis* according to Chapter 1.5. of the *Aquatic Animal Health Code* (*Aquatic Code*) are:

Family	Scientific name	Common name
Haliotidae	*Haliotis corrugata*	pink abalone
	Haliotis cracherodii	black abalone
	Haliotis discus discus	Japanese abalone
	Haliotis diversicolor	small abalone
	Haliotis fulgens	green abalone
	Haliotis kamtschatkana	pinto abalone
	Haliotis rufescens	red abalone
	Haliotis rufescens X *Haliotis discus hannai* hybrid	hybrid red and Japanese abalone
	Haliotis sorenseni	white abalone
	Haliotis tuberculata	tuberculate abalone

2.2.2. Species with incomplete evidence for susceptibility

Species for which there is incomplete evidence to fulfil the criteria for listing as susceptible to infection with *X. californiensis* according to Chapter 1.5. of the *Aquatic Code* is:

Family	Scientific name	Common name
Haliotidae	*Haliotis gigantea*	giant abalone

In addition, pathogen-specific positive polymerase chain reaction (PCR) results have been reported in the following species, but no active infection has been demonstrated:

Family	Scientific name	Common name
Haliotidae	*Haliotis discus hannai*	Japanese disc abalone

2.2.3. Likelihood of infection by species, host life stage, population or sub-populations

The bacterium divides by binary fission (Friedman *et al.,* 2000) and has direct, horizontal transmission (Braid *et al*., 2005; Friedman *et al*., 2002; Moore *et al*., 2001). Although not typically observed in farmed abalones until they are in grow-out conditions (>2.5 cm maximum size), polymerase chain reaction (PCR) examination of exposed 6-week-old abalones suggested that 1–2 mm abalones may become infected (Moore *et al*., unpublished observations). Probability of detection increases with increasing abalone size. Animals less than 10 mm in size have a reduced probability of detection using histology but equal probability of detection using PCR (Friedman *et al*., 2007; Moore *et al*., 2011).

While all post-larval life stages have been demonstrated susceptible to infection with *X. californiensis*, clinical disease is typically observed in animals >1 years of age in farmed abalones (Friedman, unpublished observations) and all abalone size classes observed in wild populations surveyed to date (e.g. Balseiro *et al*., 2006; Braid *et al*., 2005; Friedman *et al*., 1997; Haaker *et al*., 1992; Steinbeck *et al*., 1992; Van Blaricom *et al*., 1993).

2.2.4. Distribution of the pathogen in the host

Xenohaliotis californiensis infects the gastrointestinal epithelial cells of the posterior oesophagus, digestive gland and, to a lesser extent, intestine (Friedman *et al*., 2000).

2.2.5. Aquatic animal reservoirs of infection

None.

2.2.6. Vectors

None.

2.3. Disease pattern

2.3.1. Mortality, morbidity and prevalence

Susceptibility varies with species as the bacterium is known to cause disease in *H. cracherodii* (up to 99% mortality; Moore *et al*., 2009), *H. sorenseni* (up to 100% mortality; Friedman & McCormick, unpublished observations), *H. rufescens* (up to 35% mortality; Moore *et al*., 2000; 2001), *H. corrugata* and *H. fulgens* (Tinajero *et al*., 2002). Unlike the other abalone species studied to date, the magnitude of abalone mortality is not well documented in *H. corrugata* and *H. fulgens*. However, in Baja California, Mexico, up to 100% of *H. fulgens* and 63% of *H. corrugata* may be infected, with up to 43% of *H. fulgens* and 71% of *H. corrugata* having microscopic signs of disease (degenerated or metaplastic digestive gland; Tinajero *et al*., 2002).

The incubation period varies with temperature but typically involves a prolonged 3- to 7-month prepatent period. Mortality typically occurs 1–2.5 months after the onset of visible clinical signs (Friedman *et al*., 1997). Prevalence has not been well documented but up to 61% of *H. diversicolor supertexta* were infected at a farm in Thailand, however, like the European abalone, *H. tuberculata*, no abalones exhibited clinical signs of withering syndrome (Balseiro *et al*., 2001).

Infections may persist for long periods without the development of clinical disease when the host is maintained at cool water temperatures (e.g. 15°C for *H. rufescens*), and exposure to elevated seawater temperatures (e.g. >17°C for *H. rufescens*, *H. cracherodii* and *H. sorenseni*) typically results in clinical disease (Friedman & Finley, 2003; Moore *et al*., 2000; Steinbeck *et al*., 1992). Varying seawater temperatures with a lower mean temperature (e.g. 16.5°C for *H. rufescens*) may exacerbate losses (Moore *et al*., 2011). There is some suggestion that species, especially those inhabiting warmer waters may harbour the bacterium without the development of clinical disease (Wetchateng, 2008; Wetchateng *et al*., 2010).

2.3.2. Clinical signs, including behavioural changes

This intracellular pathogen infects the gastrointestinal epithelial cells, leading to clinical signs of starvation, including pedal and digestive gland atrophy. Abalones with *X. californiensis* infections may be sub-clinically infected during the prepatent period or at water temperatures ≤15°C. Infected individuals may be slightly to severely emaciated (atrophied) under permissive water temperatures.

During an epidemic, affected abalones will often cling to horizontal (as opposed to vertical or inverted) substrates and appear weak (easily removed from the substrate by hand) and emaciated (withered) (Haaker *et al*., 1992). Farmed abalones will also be anorexic. In addition, the presence of an abnormally high number of fresh shells may also indicate disease.

2.3.3 Gross pathology

Clinical disease is characterised by morphological changes in the digestive gland, which vary between species and may include degeneration (atrophy of tubules, increase in connective tissues and inflammation) and/or metaplasia of the digestive tubules. Metaplasia involves the replacement of terminal secretory/absorptive acini with absorptive/transport ducts similar in appearance to the post-oesophagus. These morphological changes are accompanied by anorexia, depletion of glycogen reserves, followed by use of the foot muscle as an energy source and subsequent death (Balseiro *et al*., 2006; Braid *et al*., 2005; Friedman *et al*., 1997; 2007; Kismohandaka *et al*., 1995; Moore *et al*., 2000; 2001). The foot of affected abalones contains fewer and less organised muscle bundles, abundant connective tissue and may contain more cerous cells than unaffected individuals (Friedman *et al*., 2007; Moore *et al*., 2000; Van Blaricom *et al*., 1993). Surviving abalones appear to remain infected, even in low water temperature environments, such as in northern California (Friedman & Finley, 2003).

2.3.4. Modes of transmission and life cycle

Transmission of *X. californiensis* is horizontal and is postulated to be via a faecal–oral route. Exposure of abalones to seawater containing infectious material is sufficient for transmission of the bacterium, and no direct animal contact is required (Balseiro *et al*., 2006; Braid *et al*., 2005; Friedman *et al*., 2002; 2007; Moore *et al*., 2000; 2001; Rosenblum *et al.,* 2008). Temperatures below 13°C have been demonstrated to limit transmission of the bacterium (i.e. less than 1% transmission) relative to those held at ~18°C (72–94% transmission) (Braid *et al*., 2005).

2.3.5. Environmental factors

Disease (withering syndrome) occurs at elevated water temperatures (~18–25°C in abalones with moderate to severe infections (Braid *et al*., 2005; Friedman *et al*., 2000; 2002; Moore *et al*., 2000; 2011; Rosenblum *et al*., 2008). Parasite transmission is enhanced in fed (94%) as opposed to starved. (72%) abalones (Balseiro *et al*., 2006; Braid *et al*., 2005). Subclinical infections have been observed in *H. diversicolor supertexta* raised at 27–29°C. As abalones are obligate marine species, salinity tolerances of the Rickettsia-like organism (RLO) have not been investigated.

2.3.6. Geographical distribution

Xenohaliotis californiensis occurs along the south-west coast of North America. However, as infected abalones have been transported to South America, Asia-Pacific, and Europe and possibly other regions, the geographical range of the aetiological agent is suspected to be broad where California red abalones, *Haliotis rufescens*, are cultured or areas where native species have been exposed to red abalones (e.g. Wetchateng, 2008)

See WAHIS (https://wahis.woah.org/#/home) for recent information on distribution at the country level.

2.4. Biosecurity and disease control strategies

2.4.1. Vaccination

No vaccines are available.

2.4.2. Chemotherapy including blocking agents

Reducing densities and application of an oxytetracycline-medicated diet may reduce losses (Friedman *et al*., 2003; 2007; Rosenblum *et al*., 2008). Oral administration of 12–19% TM-100 (90–100 mg kg^{-1}) in a medicated diet for 10 or 20 days provides protection against bacterial re-infection for several months. A single day oral administration of 12% TM-100 may reduce bacterial infections from 80% to 10% prevalence and mean infection intensity from 1.4 to 0.1 on a scale of 0–3 (Friedman *et al*., 2000; 2007).

2.4.3. Immunostimulation

No immunostimulants are currently known

2.4.4. Breeding resistant strains

Interest in selecting for resistant abalones, particularly for restoration purposes, is increasing. Wild black abalones have continued to recruit along the California Channel Islands since 2002 and some recruits survive, suggesting that these individuals may be more resistant to this rickettsial disease (Tinajero *et al*., 2002).

2.4.5. Inactivation methods

Based on successful decontamination in the laboratory, this bacterium is readily inactivated by immersion in <10% bleach. In addition, exposure of seawater containing the bacterium to >10 mg litre–1 [ppm] calcium hypochlorite and disinfection of equipment in a bath of 1% tamed iodine in freshwater for 1 hour are effective disinfectants based on the use of these disinfection methods at a marine laboratory with flow-through seawater and a lack of detection of this pathogen in adjacent abalone populations (Friedman & Finley, 2003).

2.4.6. Disinfection of eggs and larvae

No attempts to disinfect eggs and larvae have been undertaken.

2.4.7. General husbandry

Husbandry practices to control *X. californiensis* are typical of those for any bacterial disease and include the purchase of inspected seed (devoid of evidence of infection), maintaining separate families or groups (i.e. avoid high grading and mixing of disparate groups), rinsing hands and equipment in freshwater or iodinated water and drying them in between uses. Isolation of infected groups is recommended if possible.

3. Specimen selection, sample collection, transportation and handling

This section draws on information in Sections 2.2, 2.3 and 2.4 to identify populations, individuals and samples that are most likely to be infected.

3.1. Selection of populations and individual specimens

To optimise detection (targeted sampling), selection of abalones exhibiting the clinical sign of reduced weight (atrophied pedal muscle) is recommended. If possible, animals should be sampled after exposure to a period (e.g. 30 days) of warm water (e.g. >18°C).

3.2. Selection of organs or tissues

The best target tissue is the posterior oesophagus, and the second-best tissue is the digestive gland/intestine complex.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_3.2_1	**Canada**	**Category:** addition	Agreed.

		Canada requests the reference(s) for this tissue type selection be included for Member information.	.

3.3. Samples or tissues not suitable for pathogen detection

Non-digestive tissues do not contain rickettsial DNA and should be avoided.

3.4. Non-lethal sampling

None

3.5. Preservation of samples for submission

3.5.1. Samples for pathogen isolation

Not applicable.

3.5.2. Preservation of samples for molecular detection

~~Tissue samples for PCR testing should be preserved in 80% (v/v) analytical grade ethanol. The recommended ratio of ethanol to tissue is 10:1 based on studies in terrestrial animal and human health. The use of lower grade (laboratory or industrial grade) ethanol is not recommended. If material cannot be fixed it may be frozen.~~

Standard sample collection, preservation and processing methods for molecular techniques can be found in Section B.5.5 of Chapter 2.4.0 *General information* (diseases of molluscs).

3.5.3. Samples for histopathology, immunohistochemistry or *in-situ* hybridisation

Standard sample collection, preservation and processing methods for histological techniques can be found in Section B.5.3 of Chapter 2.4.0 *General information* (diseases of molluscs).

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_3.5.2_1	**Thailand**	**Category:** General We would like to request WOAH to review the formatting of Section 3.5.2 to ensure consistency with other *Manual* chapters. It is observed that chapters for other mollusc diseases that were recently updated (e.g. Chapters 2.4.2 Infection with *Bonamia exitiosa* and 2.4.5 Infection with *Perkinsus marinus*) still have the detail of sample preservation provided in this section. **Proposed amended text:** No text proposed **Rationale:** To ensure consistency of the *Manual* chapters **Supporting document:** –	Agreed: for consistency, the AAC agreed not to duplicate any information in the disease-specific chapters that is already in the general chapter unless there are specific methods that apply to specific diseases.

3.5.4. Samples for other tests

None.

3.6. Pooling of samples

Pooling of samples from more than one individual animal for a given purpose is only recommended where robust supporting data on diagnostic sensitivity and diagnostic specificity have been evaluated and found to be suitable. If the effect of pooling on diagnostic sensitivity has not been thoroughly evaluated, specimens should be processed and tested individually. Small life stages such as spat can be pooled to obtain the minimum amount of material for virus isolation or molecular detection.

4. Diagnostic methods

The methods currently available for pathogen detection that can be used in i) surveillance of apparently healthy animals, ii) presumptive diagnosis in clinically affected animals and iii) confirmatory diagnostic purposes are listed in Table 4.1. by animal life stage.

Ratings for purposes of use. For each recommended assay a qualitative rating for the purpose of use is provided. The ratings are determined based on multiple performance and operational factors relevant to application of an assay for a defined purpose. These factors include appropriate diagnostic performance characteristics, level of assay validation, availability cost, timeliness, and sample throughput and operability. For a specific purpose of use, assays are rated as:

+++ = Methods are most suitable with desirable performance and operational characteristics.

++ = Methods are suitable with acceptable performance and operational characteristics under most circumstances.

+ = Methods are suitable, but performance or operational characteristics may limit application under some circumstances.

Shaded boxes = Not appropriate for this purpose.

Validation stage. The validation stage corresponds to the assay development and validation pathway in chapter 1.1.2. The validation stage is specific to each purpose of use. Where available, information on the diagnostic performance of recommended assays is provided in Section 6.3.

WOAH Reference Laboratories welcome feedback on diagnostic performance of recommended assays, in particular PCR methods. Of particular interest are any factors affecting expected assay sensitivity (e.g. tissue components inhibiting amplification) or expected specificity (e.g. failure to detect particular genotypes, detection of homologous sequences within the host genome). These issues should be communicated to the WOAH Reference Laboratories so that advice can be provided to diagnostic laboratories and the standards amended if necessary.

Table 4.1. *WOAH recommended diagnostic methods and their level of validation for surveillance of apparently healthy animals and investigation of clinically affected animals*

Method	A. Surveillance of apparently healthy animals				B. Presumptive diagnosis of clinically affected animals				C. Confirmatory diagnosis[1] of a suspect result from surveillance or presumptive diagnosis			
	Early life stages[2]	Juveniles[2]	Adults	LV	Early life stages[2]	Juveniles[2]	Adults	LV	Early life stages[2]	Juveniles[2]	Adults	LV
Wet mounts												
Histopathology	+	++	++	1	+	+++	+++	1				
Cell cultures												
Real-time PCR	+++	+++	+++	3	+++	+++	+++	NA~~3~~	+++	+++	+++	3
Conventional PCR	+++	+++	+++	1	+++	+++	+++	1				
Conventional PCR followed by amplicon sequencing									+++	+++	+++	1
In-situ hybridisation					+	+	+	1	+	+	+	1
Bioassay												
LAMP												
Ab-ELISA												
Ag-ELISA												
Other antigen detection methods[3]												
Other methods[3]												

LV = level of validation, refers to the stage of validation in the WOAH Pathway (chapter 1.1.2); PCR = polymerase chain reaction; LAMP = loop-mediated isothermal amplification; Ab- or Ag-ELISA = antibody or antigen enzyme-linked immunosorbent assay, respectively; IFAT = indirect fluorescent antibody test.
[1]For confirmatory diagnoses, methods need to be carried out in combination (see Section 6). [2]Susceptibility of early and juvenile life stages is described in Section 2.2.3.
[3]Specify the test used. Shading indicates the test is inappropriate or should not be used for this purpose.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_Table4.1_1	**Canada**	**Category:** addition Canada thanks the Commission for responding to previous comments indicating that DSe and Dsp are not always published but may be provided by the Reference Laboratory to support the ratings provided within Table 4.1 and therefore DSe and DSp cannot be included within Tables 6.1.2 and 6.1.3. When there is no published information on diagnostic sensitivity in the scientific literature and the rating is based on unpublished reference laboratory validation studies, we would request that a footnote including an explanation for the discrepancy between the rating and the published literature would enhance transparency and clarity for Table 4.1.	Noted for future updates, when relevant. At present, the validation study template has been used for one chapter only: Infection with yellow head virus genotype 1.
2.4.7_Table4.1_2	**Australia**	**Category:** change **Proposed amended text:** n/a **Rationale**: In Table 4.1, the LV for real-time PCR for A). Surveillance of apparently healthy animals is not supported by data currently presented in Table 6.3.2. The LV level should be changed from "3" to "N/A" in Table 4.1, following Australia's suggested changes to Table 6.3.2. The assay seems to have been validated for clinically diseased animals only but not apparently healthy animals. In addition, the LV for Real-time PCR for B). Presumptive diagnosis of clinically affected animals should be changed from "N/A" to "3" in Table 4.1, following Australia's suggested corrections to data presented in Table 6.3.1 **Supporting evidence:** n/a	Disagreed: the quoted paper does not explicitly mention clinically affected animals rather just exposure. Changed the description of the source animals in Section 6.2.3 to remove confusion.

4.1. Wet mounts

Not applicable.

4.2. Histopathology and cytopathology

The presence of basophilic, ovoid intracytoplasmic bacterial inclusions in digestive epithelia (posterior oesophagus, transport ducts and metaplastic epithelia of the digestive gland, and/or intestine). Metaplastic changes in the digestive gland that include the transformation of the terminal digestive tubules into absorptive/transport epithelia occurs in abalones infected with *X. californiensis* (Balseiro *et al*., 2006; Braid *et al*., 2005; Friedman *et al*., 2002; Moore *et al*., 2000; 2001). Although metaplasia has been observed in all affected species examined to date, the response to infection may vary between hosts. Red abalones and white abalones, for example, typically respond with a metaplastic change (Balseiro *et al*., 2006; Braid *et al*., 2005; Moore *et al*., 2000), while black abalones generally respond with a combination of metaplasia, digestive tubule degeneration and inflammation (Friedman *et al*., 1997; 2002). Affected individuals contain less pedal glycogen and fewer muscle bundles than do unaffected individuals (Balseiro *et al*., 2006; Braid *et al*., 2005; Gardner *et al*., 1995). In some abalones, an increase in serous cells may be observed in the foot muscle (Van Blaricom *et al*., 1993), but these signs are not pathognomonic for this disease. Juvenile white abalone may contain an apparently metaplastic digestive without presence of *X. californiensis* (Friedman *et al*., 2007). Thus, the presence of *X. californiensis* in digestive epithelia in conjunction with the morphological changes noted above indicate the presence of clinical withering syndrome.

4.3. Cell culture for isolation

Not applicable.

4.4. Nucleic acid amplification

PCR assays should always be run with the controls specified in Section B.5.5 *Molecular methods* Chapter 2.4.0 *General information* (diseases of molluscs). Each sample should be tested in duplicate.

Extraction of nucleic acids

Different kits and procedures can be used for nucleic acid extraction. The quality and concentration of the extracted nucleic acid is important and can be checked using a suitable method as appropriate to the circumstances.

4.4.1. Real-time PCR

Pathogen/ target gene	Primer/probe (5'–3')	Concentration	Cycling parameters
Method 1: Friedman *et al.,* 2014; GenBank Accession No.: AF133090, amplicon size: 147 bp			
16S rDNA	Fwd WSN1 F: AGT-TTA-CTG-AAG-GCA-AGT-AGC-AGA Rev WSN1 R: TCT-AAC-TTG-GAC-TCA-TTC-AAA-AGC Probe WS-RLO_P: TGC-TTG-GAA-ATC-TAC-TCA-GAA-GAC-ATG-A	320 nM 320 nM 200 nM	45 cycles of: 95°C/15 sec and 60°C/30 sec

4.4.2. Conventional PCR

Pathogen/ target gene	Primer (5'–3')	Concentration	Cycling parameters[(a)]
Method 1: Andree *et al.,* 2000; GenBank Accession No.: AF133090, amplicon size: 160 bp			
16S rDNA	Fwd RA 5-1: GTT-GAA-CGT-GCC-TTC-AGT-TTA-C Rev RA 3-6**:** ACT-TGG-ACT-CAT-TCA-AAA-GCG-GA	500 nM 500 nM	40 cycles of: 95°C/60 sec, 50°C/30 sec and 72°C/30 sec
Method 2: Cicala *et al.,* 2017; GenBank Accession No.: KU645900, amplicon size: 426 bp			
16S rDNA	Fwd ss16S-F: GCC-TCA-GTT-TGG-CTG-GGT-TCT-TCA Rev ss16S-R: GAA-TTG-CCA-CTT-TAA-AGT-ATG-GAC-GG	300 nM 300 nM	40 cycles of: 94°C/60 sec, 66°C/30 sec and 72°C/30 sec

4.4.3. Other nucleic acid amplification methods

Not applicable.

4.5. Amplicon sequencing

The size of the PCR amplicon should be verified, for example by agarose gel electrophoresis. Both DNA strands of the PCR product must be sequenced and analysed in comparison with reference sequences.

4.6. *In-situ* hybridisation

Use of Rickettsiales-like prokaryotes specific DNA probes with histological sections is useful to demonstrate the presence of *X. californiensis* nucleic acid in infected cells (Antonio *et al*., 2000). See Chapter 2.4.0 Section 5.5.4 for general comments on *in-situ* hybridisation.

Antonio *et al.* (2000) developed an ISH method targeting the 16S rDNA gene. This method allows the detection of Rickettsiales-like prokaryotes in tissue sections. Although this method has not been formally validated, tests for specificity using several bivalve and fish rickettsial organisms suggested that the test was specific for *X. californiensis*.

Reference	Pathogen/target gene	ISH probe	Probe size
Antonio *et al.* (2000)	16S rDNA	RA 5-1: GTT-GAA-CGT-GCC-TTC-AGT-TTA-C RA 3-6: ACT-TGG-ACT-CAT-TCA-AAA-GCG-GA; RA 3-8: CCA-CTG-TGA-GTG-GTT-ATC-TCC-TG; RA 5-6: GAA-GCA-ATA-TTG-TGA-GAT-AAA-GCA.	oligo nucleotide probes

Reference	**Member**	**Comment**	**Aquatic Animals Commission response**
2.4.7_4.6_1			Agreed.

	China (People's Rep. of)	**Category:** change **Proposed amended text:** Change "Probe ~~size~~" to "Probe type" in the table heading **Rationale:** to reflect the table content. **Supporting evidence:** not relevant	.

4.7. Immunohistochemistry

Not applicable.

4.8. Bioassay

Not applicable.

4.9. Antibody- or antigen-based detection methods (ELISA, etc.)

Not applicable.

4.10. Other methods

Not applicable.

5. Test(s) recommended for surveillance to demonstrate freedom in apparently healthy populations

The recommended method for surveillance is real-time PCR using the assay by Friedman *et al.* (2014).

6. Corroborative diagnostic criteria

This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence of clinical signs (Section 6.2.) but does not evaluate whether the infectious agent is the cause of the clinical event.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence (Section 6.2.) of clinical signs ~~(Section 6.2.)~~ but does not evaluate whether the infectious agent is the cause of the clinical event. **Rationale:** Edited so that reference to section is consistent. Supporting evidence n/a	Agreed.

The case definitions for a suspect and confirmed case have been developed to support decision making related to trade and confirmation of disease status at the country, zone or compartment level. Case definitions for disease confirmation in endemically affected areas may be less stringent. If a Competent Authority does not have the capability to undertake the necessary diagnostic tests it should seek advice from the appropriate WOAH Reference Laboratory, and if necessary, refer samples to that laboratory for confirmatory testing of samples from the index case in a country, zone or compartment considered free. There are currently no WOAH Reference Laboratories for *Xenohaliotis californiensis*.

6.1. Apparently healthy animals or animals of unknown health status[1]

1 For example transboundary commodities.

Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Apparently healthy animals or animals of unknown health status[1] Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Testing may also be carried out on transboundary commodities, at which time the health status of the animals from which they were derived may be of unknown. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom. **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.

6.1.1. Definition of suspect case in apparently healthy animals

The presence of infection with *X. californiensis* shall be suspected if at least one of the following criteria is met:

i) Histopathological changes consistent with the presence of the pathogen or the disease

ii) Positive result by conventional PCR

iii) Positive result by real-time PCR

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6.1.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** 6.1.1. Definition of suspect case in apparently healthy animals or animals of unknown health status[1] The presence of infection with *X. californiensis* shall be suspected if at least one of the following criteria is met:… **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown. .

6.1.2. Definition of confirmed case in apparently healthy animals

The presence of infection with *X. californiensis* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing

ii) Positive result by *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing

iii) Positive result by *in-situ* hybridisation and positive result by real-time PCR

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6.1.2_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Definition of confirmed case in apparently healthy animals or animals of unknown health status **Rationale:** Edited to make it explicit that criteria for confirmed case also applied to "animals of unknown health status" as described in 6.1.1. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown. . .
2.4.7_6.1.2_2	**EU**	**Category:** Addition **Proposed amended text:** The presence of infection with *X. californiensis* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by histology or *in-situ* hybridisation and positive result by PCR combined with sequencing ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing **Rationale:** alignment with the case definitions for *B ostreae, B exitiosa and M refringens*	Partially agreed, the Commission agreed that case definitions for a confirmed case in either apparently healthy or clinically affected animals should recommend two specific methods, as is the current approach for other diseases. Molecular methods should either be combined with another diagnostic method or two molecular methods targeting non-overlapping regions of the pathogen genome should be used. Reworded the proposal. Disagreed on adding histology as it is not a confirmatory test. . .
2.4.7_6.1.2_2	**Australia**	**Category:** change **Proposed amended text:** 6.1.2. Definition of confirmed case in apparently healthy animals The presence of infection with *X. californiensis* is considered to be confirmed if at least one of the following ~~criterion~~ criteria is met: i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing ii) Positive result by *In-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing iii) Positive result of real-time PCR followed by positive result by *in-situ* hybridisation ~~and positive result by real-time PCR~~ **Rationale**: Change for clarity to reflect the correct operational order of using these testing methods, as real-time PCR is not specific, whereas *in-situ* hybridisation (ISH) is specific and used for confirmation testing, so ISH is used following positive real-time PCR results. It does not make	Agreed. .

		sense to undertake non-specific PCR testing if you already have specific positive test results by ISH. **Supporting evidence:** n/a	

6.2 Clinically affected animals

Clinical signs are not pathognomonic for a single disease; however they may narrow the range of possible diagnoses.

6.2.1. Definition of suspect case in clinically affected animals

The presence of infection with *X. californiensis* shall be suspected if at least one of the following criteria is met:

i) Histopathological changes consistent with the presence of the pathogen or the disease

ii) Positive result by *in-situ* hybridisation

iii) Positive result by conventional PCR

iv) Positive result by real-time PCR

6.2.2. Definition of confirmed case in clinically affected animals

The presence of infection with *X. californiensis* is considered to be confirmed if at least at least one of the following criteria is met:

i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing.

ii) Positive result by *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing.

iii) Positive result by *In-situ* hybridisation and positive result by real-time PCR.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6.2.2_1	**EU**	**Category:** Addition **Proposed amended text:** The presence of infection with *X. californiensis* is considered to be confirmed if at least one of the following criteria is met: i) Positive result by histology or *in-situ* hybridisation and positive result by PCR combined with sequencing ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing **Rationale:** alignment with the case definitions for *B ostreae, B exitiosa and M refringens*	Partially agreed, the Commission agreed that case definitions for a confirmed case in either apparently healthy or clinically affected animals should recommend two specific methods, as is the current approach for other diseases. Molecular methods should either be combined with another diagnostic method or two molecular methods targeting non-overlapping regions of the pathogen genome should be used. Reworded the proposal. Disagreed on adding histology as it is not a confirmatory test.
2.4.7_6.2.2_2	**Australia**	**Category:** change **Proposed amended text:** The presence of infection with *X. californiensis* is considered to be confirmed if at least one of the following criteria is met:	Agreed.

		i) Positive result by real-time PCR and positive result by conventional PCR followed by amplicon sequencing. ii) Positive result by *in-situ* hybridisation and positive result by conventional PCR followed by amplicon sequencing. iii) Positive result of real-time PCR followed by positive result by *in-situ* hybridisation ~~and positive result by real-time PCR~~ **Rationale**: For clarity and consistency with comment above for 6.1.2 iii) above **Supporting evidence:** n/a	

6.3. Diagnostic sensitivity and specificity for diagnostic tests

The diagnostic performance of tests recommended for surveillance or diagnosis of infection with *Xenohaliotis californiensis* are provided in Tables 6.3.1. [no data currently available] and 6.3.2). Data are only presented where tests are validated to at least level 2 of the validation pathway described in Chapter 1.1.2. and the information is available within published diagnostic accuracy studies.

6.3.1. For presumptive diagnosis of clinically affected animals (no data are currently available)

Test type	Test purpose	Source populations	Tissue or sample types	Species	DSe (*n*)	DSp (*n*)	Reference test	Citation
~~Real-time PCR~~	~~Diagnosis~~	~~Clinically diseased abalone from farms~~	~~Posterior esophagus and digestive gland tissue~~	~~*Haliotis rufescens, H. sorensoni, H. fulgens, H. discushannai, H. cracherodii, H. kamtschatkana*~~	~~100 (518)~~	~~99.7 (518)~~	~~Histopathology~~	~~Friedman *et al.* (2014)~~

DSe = diagnostic sensitivity, DSp = diagnostic specificity, *n* = number of animals used in the validation study, PCR: = polymerase chain reaction.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6.3.1_1	**Australia**	**Category:** change **Proposed amended text:** The deleted data/text in Table 6.3.1 should be retained. **Rationale:** see comment below for Table 6.3.2 **Supporting evidence:** –	Disagreed the study population is not clinically affected described as exposed or unexposed.

6.3.2. For surveillance of apparently healthy animals ~~(no data are currently available)~~

Test type	Test purpose	Source populations	Tissue or sample types	Species	DSe (*n*)	DSp (*n*)	Reference test	Citation
Real-time PCR	Diagnosis	Clinically diseased abalone from farms	Posterior esophagus and digestive gland tissue	*Haliotis rufescens, H. sorensoni, H. fulgens, H. discushannai, H. cracherodii, H. kamtschatkana*	100 (518)	99.7 (518)	Histopathology	Friedman *et al.* (2014)

DSe = diagnostic sensitivity, DSp = diagnostic specificity, *n* = number of animals used in the validation study, PCR: = polymerase chain reaction.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.7_6.3.2_1	**Thailand**	**Category:** General We would like to request WOAH to review the scientific validity and appropriateness of designating histopathology as a confirmatory method for PCR-positive samples. Given the difference in sensitivity between the two methods, this may affect the accuracy of the confirmatory diagnosis. **Proposed amended text:** No text proposed **Rationale:** To ensure scientific accuracy **Supporting Document:** –	Agreed: acknowledge that histopathology is not a reference test (but it is the reference test used in the manuscript), which is why a reference test is included in the table, and the performance of a test should always be considered in relation to a reference test. This topic is covered in the validation chapter
2.4.7_6.3.2_2	**Australia**	**Category:** change **Proposed amended text:** The data in Table 6.3.2 should be deleted **Rationale**: It is not appropriate to use clinically diseased abalone to determined estimates of Dse in apparently healthy animals. This data should be deleted from Table 6.3.2 and retained in Table 6.3.1. **Supporting evidence:** –	Disagreed: the quoted paper does not explicitly mention clinically affected animals rather just exposure. Changed the description of the source animals in Section 6.3.2 to remove confusion.

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* *

NB: Currently (2025) there is no WOAH Reference Laboratory for infection with *Xenohaliotis californiensis* (please consult the WOAH web site: https://www.woah.org/en/what-we-offer/expertise-network/reference-laboratories/#ui-id-3).

NB: FIRST ADOPTED IN 2006. MOST RECENT UPDATES ADOPTED IN 2012.

Update on viral taxonomy in Scope of Chapters 2.2.3., 2.2.5., 2.2.6., 2.2.7., 2.2.8., 2.2.10., 2.3.2., 2.3.4., 2.3.5., 2.3.6., 2.3.8., 2.3.9., 2.3.10., and 2.4.1.

Reference	Member	Comment	Aquatic Animals Commission response
Annex20_1	**EU**	**Category:** General The EU supports the revised scientific names. However, the EU would kindly request further clarifications about the definition of infection with infectious salmon anaemia virus (ISAV). Please, see specific question below. In addition, please see several editorial comments to Chapter 2.3.6 (infection with Koi herpesvirus).	Noted and generally agreed.
Annex20_2	**Canada**	**Comment Category:** General We note that in Annex 13, infection with yellowhead virus was included to ensure the text in Article 9.10.1. was harmonised with other diseases. However, the name of the pathogenic agent was updated in 2022 *to Okavirus flavicapitis* and this should also be reflected in the *Aquatic Manual*. Additionally, in Section 2.2.1. of the *Manual* there is a description of the different YHV genogroups that may require updating as ICTV now lists YHV8 as a different viral species *Okavirus orientale*. It is unclear where YHV2-7 are placed (see Genus: Okavirus | ICTV).	Noted and will be addressed in the next update of the yellow head chapter.

CHAPTER 2.2.3.

INFECTION WITH DECAPOD IRIDESCENT VIRUS 1

1. Scope

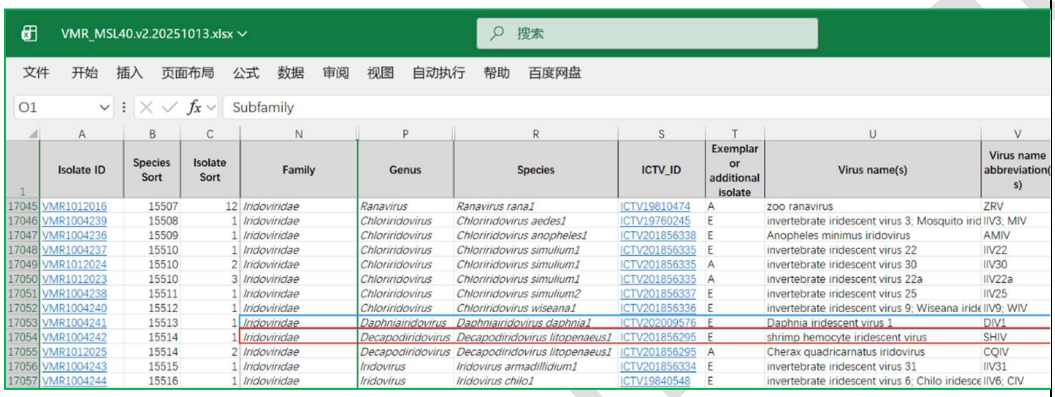
Infection with decapod iridescent virus 1 (DIV1) means infection with the pathogenic agent *Decapodiridovirus litopenaeus1* ~~decapod iridescent virus 1 (DIV1), Genus *Decapodiridovirus*,~~ of the Subfamily *Betairidovirinae*, Family *Iridoviridae*.

Reference	Member	Comment	Aquatic Animals Commission response
2.2.3_1_1	**China (People's Rep. Of)**	**Category:** general We agree with WOAH here to change decapod iridescent virus 1 (DIV1) to *Decapodiridovirus litopenaeus1*. However, it is inappropriate to use decapod iridescent virus 1 (DIV1) as the chapter title and the name of the virus in the *Aquatic Code* and *Aquatic Manual*. We suggest changing the chapter title from "infection with decapod iridescent virus 1" to "infection with *Decapodiridovirus litopenaeus1*" or "infection with shrimp hemocyte iridescent virus". The virus name "decapod iridescent virus 1" and its abbreviation DIV1 should be discarded, and the virus name should be changed to *Decapodiridovirus litopenaeus1* or shrimp hemocyte iridescent virus (SHIV). Reasons for revision are as follows:	Disagreed: infection with decapod iridescent virus 1 and the abbreviation DIV1 is widely known for several years. Changing it would cause confusion.

(1) In 2023, ICTV changed the scientific name of this virus from *Decapod iridescent virus 1* to *Decapodiridovirus litopenaeus1*. The scientific name *Decapod iridescent virus 1* has been discontinued by ICTV and is no longer in use.

(2) The abbreviation DIV1 has been officially used by ICTV for *Daphnia iridescent virus 1* (the exemplar isolate of species *Daphniairidovirus daphnia1*), and ICTV has never abbreviated decapod iridescent virus 1 as DIV1. Abbreviating Decapod iridescent virus 1 (a name that has been discarded by ICTV) as DIV1 can cause confusion. Therefore, DIV1 should no longer be used to refer to *Decapodiridovirus litopenaeus1*.

(3) The exemplar isolate of species *Decapodiridovirus litopenaeus1* is "shrimp hemocyte iridescent virus (SHIV)" according to ICTV.


VMR_MSL40.v2.20251013.xlsx

搜索

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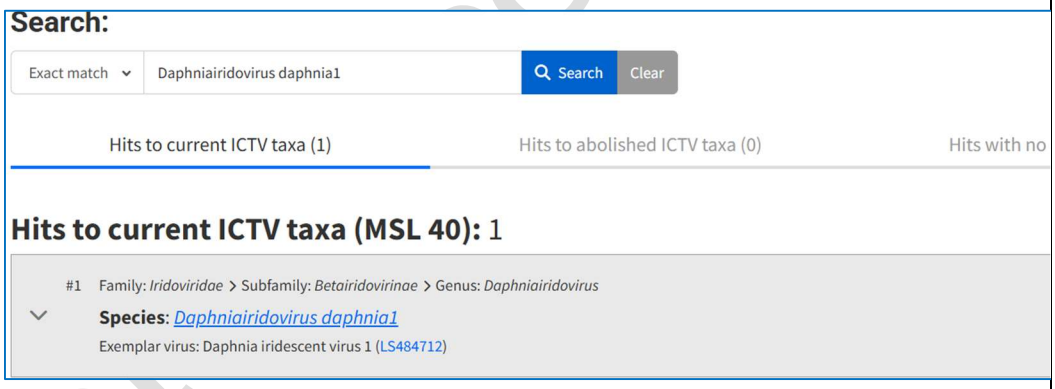
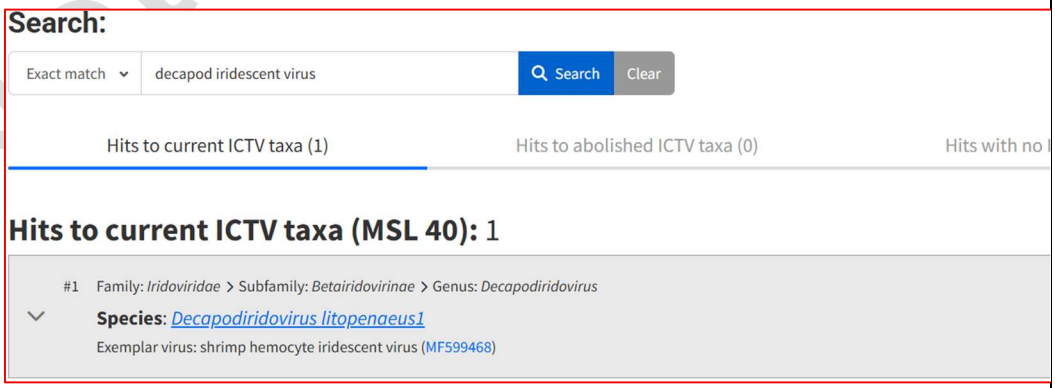
O1 Subfamily

	A	B	C	N	P	R	S	T	U	V
1	Isolate ID	Species Sort	Isolate Sort	Family	Genus	Species	ICTV_ID	Exemplar or additional isolate	Virus name(s)	Virus name abbreviation(s)
17045	VMR1012016	15507	12	*Iridoviridae*	*Ranavirus*	*Ranavirus rana1*	ICTV19810474	A	zoo ranavirus	ZRV
17046	VMR1004239	15508	1	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus aedes1*	ICTV19760245	E	invertebrate iridescent virus 3; Mosquito irid	IIV3; MIV
17047	VMR1004236	15509	1	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus anopheles1*	ICTV201856338	E	Anopheles minimus iridovirus	AMIV
17048	VMR1004237	15510	1	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus simulium1*	ICTV201856335	E	invertebrate iridescent virus 22	IIV22
17049	VMR1012024	15510	2	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus simulium1*	ICTV201856335	A	invertebrate iridescent virus 30	IIV30
17050	VMR1012023	15510	3	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus simulium1*	ICTV201856335	A	invertebrate iridescent virus 22a	IIV22a
17051	VMR1004238	15511	1	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus simulium2*	ICTV201856337	E	invertebrate iridescent virus 25	IIV25
17052	VMR1004240	15512	1	*Iridoviridae*	*Chloriridovirus*	*Chloriridovirus wiseana1*	ICTV201856336	E	invertebrate iridescent virus 9; Wiseana iride	IIV9; WIV
17053	VMR1004241	15513	1	*Iridoviridae*	*Daphniairidovirus*	*Daphniairidovirus daphnia1*	ICTV202009576	E	Daphnia iridescent virus 1	DIV1
17054	VMR1004242	15514	1	*Iridoviridae*	*Decapodiridovirus*	*Decapodiridovirus litopenaeus1*	ICTV201856295	E	shrimp hemocyte iridescent virus	SHIV
17055	VMR1012025	15514	2	*Iridoviridae*	*Decapodiridovirus*	*Decapodiridovirus litopenaeus1*	ICTV201856295	A	Cherax quadricarnatus iridovirus	CQIV
17056	VMR1004243	15515	1	*Iridoviridae*	*Iridovirus*	*Iridovirus armadillidium1*	ICTV201856334	E	invertebrate iridescent virus 31	IIV31
17057	VMR1004244	15516	1	*Iridoviridae*	*Iridovirus*	*Iridovirus chilo1*	ICTV19840548	E	invertebrate iridescent virus 6; Chilo iridesce	IIV6; CIV

(from ICTV database file 2025: VMR_MSL40.v2.20251013.xlsx)

See also: ICTV Database Search Results





Supporting evidence: not relevant

CHAPTER 2.2.5.

INFECTION WITH HYPODERMAL AND HAEMATOPOIETIC NECROSIS VIRUS

Reference	Member	Comment	Aquatic Animals Commission response
2.2.5_1	**Canada**	**Category:** correction **Proposed amended text: amend the title to:** INFECTION WITH INFECTIOUS HYPODERMAL AND HAEMATOPOIETIC NECROSIS VIRUS **Rationale:** Editorial to match the name of the listed disease	Agreed.

1. Scope

Infection with infectious hypodermal and haematopoietic necrosis virus (IHHNV) means infection with the pathogenic agent *Penstylhamaparvovirus decapod 1* ~~Decapod penstylhamaparvovirus 1~~, of the ~~Genus *Penstylhamaparvovirus* and~~ Family *Parvoviridae*.

[…]

CHAPTER 2.2.6.

INFECTION WITH INFECTIOUS MYONECROSIS VIRUS

1. Scope

Infection with infectious myonecrosis virus (IMNV) means infection with the pathogenic agent infectious myonecrosis virus ~~(IMNV) that is tentatively assigned to the Family *Totiviridae*~~ of the genus Artivirus and Family Artiviridae.

Reference	Member	Comment	Aquatic Animals Commission response
2.2.6_1_1	**Japan**	Editorial **Proposed amended text**: Infection with infectious myonecrosis virus (IMNV) means infection with the pathogenic agent infectious myonecrosis virus of the genus *Artivirus* ~~Artivirus~~ and Family *Artiviridae* ~~Artiviridae~~. **Rational**: Japan proposes to revise as above to align the format with other chapters (italics).	Agreed.
2.2.6_1_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** Infection with infectious myonecrosis virus (IMNV) means infection with the pathogenic agent ~~infectious myonecrosis virus~~ *Artivirus ichi* ~~(IMNV) that is tentatively assigned to the Family *Totiviridae*~~ of ~~the genus Artivirus and~~ Family Artiviridae. **Rational**: In 2023, ICTV updated the classification of IMNV from an unassigned genus in the family Totiviridae to a member of the genus Artivirus in the family Artiviridae. The scientific name of IMNV is "*Artivirus ichi*", and the exemplar isolate of species *Artivirus ichi* is Penaeid shrimp infectious myonecrosis virus (PSIMNV, AY570982). Here we recommend using the scientific name of this virus *Artivirus ichi*.	Agreed.
2.2.6_1_3	**Canada**	**Category:** change **Proposed amended text:** Infection with infectious myonecrosis virus (IMNV) means infection with the pathogenic agent *Artivirus ichi* ~~infectious myonecrosis virus (IMNV) that is tentatively assigned to the Family *Totiviridae*~~ of the genus Artivirus and Family Artiviridae. **Rationale:** A search for IMNv (Penaeid shrimp infectious myonecrosis virus) within ICTV returns the virus species *Artivirus ichi.* The ICTV report in family Artiviridea (Family: Artiviridae (Interim Report) | ICTV) lists Penaeid shrimp infectious myonecrosis virus as *Artivirus ichi*	Agreed.

2. Disease information

2.1. Agent factors

2.1.1. Aetiological agent

IMNV is tentatively assigned to the family Artiviridae (ICTV, 2025) ~~*Totiviridae* (Lightner, 2011; Nibert, 2007; Poulos *et al.*, 2006; Wickner *et al.*, 2011)~~.

Reference	Member	Comment	Aquatic Animals Commission response
2.2.6_1_2	**China (People's Rep. Of)**	**Category:** change **Proposed amended text:** IMNV ~~is~~ ~~tentatively~~ was assigned ~~to~~ by the International Committee on Taxonomy of Viruses (ICTV) as a member of the genus *Artivirus* within the family Artiviridae (ICTV, 2025) ~~*Totiviridae* (Lightner, 2011; Nibert, 2007; Poulos *et al.*, 2006; Wickner *et al.*, 2011)~~. **Rationale:** In 2023, ICTV updated the classification of IMNV from an unassigned genus in the family Totiviridae to a member of the genus *Artivirus* in the family Artiviridae. The scientific name of IMNV is "*Artivirus ichi*", and the exemplar isolate of species *Artivirus ichi* is Penaeid shrimp infectious myonecrosis virus (PSIMNV, AY570982). Here we recommend using the scientific name of this virus *Artivirus ichi*.	Agreed and slightly modified the proposed text.

IMNV particles are icosahedral in shape and 40 nm in diameter, with a buoyant density of 1.366 g ml^{-1} in caesium chloride. The genome consists of a single, double-stranded (ds) RNA molecule of 8226–8230 bp (Loy *et al*., 2015; Naim *et al.,* 2015). Sequencing of the viral genome reveals two non-overlapping open reading frames (ORFs). The first ORF (ORF1, 470–5596 nt) encodes a putative RNA-binding protein and a capsid protein. The coding region of the RNA-binding protein is located in the first half of ORF1 and contains a dsRNA-binding motif in the first 60 amino acids. The second half of ORF1 encodes a capsid protein, as determined by amino acid sequencing, with a molecular mass of 106 kDa. The second ORF (ORF2, 5884–8133 nt) encodes a putative RdRp (Poulos *et al.,* 2006). The most variable region of IMNV genome is located in the first half of ORF1, coinciding with a region which probably encodes the capsid protrusions (Dantas *et al*., 2015).

The complete genomes of IMNV types originating from Brazil and Indonesia have been sequenced and found to be 99.6% identical at the nucleotide level (Poulos *et al.,* 2006; Senapin *et al.,* 2007). The 99.6% full genome sequence identity (and anecdotal information on the introduction of *Penaeus vannamei* stocks from Brazil) indicate that the disease was introduced from Brazil to Indonesia in 2006. A new genotype was analysed in infected samples in 2018 in Indonesia, including an isolate that contains a deletion of 622 amino acids (Mai *et al.,* 2019).

[…]

CHAPTER 2.2.7.

INFECTION WITH MACROBRACHIUM ROSENBERGII NODAVIRUS (WHITE TAIL DISEASE)

1. Scope

Infection with Macrobrachium rosenbergii nodavirus means infection with the pathogenic agent Macrobrachium rosenbergii nodavirus (MrNV) in the Family *Nodaviridae*. The disease is commonly known as white tail disease (WTD).

Extra small virus (XSV) is associated with disease, but its role has not been determined.

2. Disease information

2.1. Agent factors

2.1.1. Aetiological agent

Two viruses are associated with WTD, namely MrNV (primary) and extra small virus (XSV) (associate) (Qian *et al.,* 2003; Romestand & Bonami, 2003). MrNV is a necessary cause of WTD in prawns, however, the role of XSV in pathogenicity remains unclear.

MrNV belongs in the family *Nodaviridae* (Bonami *et al.,* 2005). While the physico-chemical properties of MrNV are consistent with those of other members of the *Nodaviridae*, it differs structurally and genetically from other nodaviruses within the two recognised genera, *Alphanodavirus* and *Betanodavirus* (Ho *et al*., 2017, 2018; Naveenkumar *et al*., 2013). Consequently, a third genus, *Gammanodavirus*, has been proposed for nodaviruses that infect crustaceans, including MrNV and *Penaeus vannamei* nodavirus (*Pv*NV) (Naveenkumar *et al*., 2013).

XSV is the first sequenced satellite virus in aquatic animals and it is also the first record of a satellite-nodavirus association (Bonami *et al.,* 2005). XSV has been classified by the ICTV as *Macronovirus macrobrachii* ~~*Macrobrachium satellite virus 1*~~ of the family *Sarthroviridae.*

[…]

CHAPTER 2.2.8.

INFECTION WITH TAURA SYNDROME VIRUS

1. Scope

Infection with Taura syndrome virus (TSV) means infection with the pathogenic agent *Aparavirus tauraense* ~~Taura syndrome virus (TSV)~~, ~~Genus *Aparavirus*,~~ of the Family *Dicistroviridae*, Order *Picornavirales*.

[…]

CHAPTER 2.3.4.

INFECTION WITH HRP-DELETED OR HPR0 INFECTIOUS SALMON ANAEMIA VIRUS

Reference	Member	Comment	Aquatic Animals Commission response
2.3.4_1	**EU**	**Category:** General The EU supports the revised Section. However, the EU would seek clarification as regards the reasons to remove the "genus isavirus" as it is still in the ICTV description https://ictv.global/taxonomy/taxondetails?taxnode_id=202403964&taxon_name=Isavirus%20salaris	Noted.

1. Scope

Infection with infectious salmon anaemia virus (ISAV) means infection with the pathogenic agent *Isavirus salaris* of the ~~Genus *Isavirus* and~~ Family *Orthomyxoviridae*. Infection with ISAV includes two genotypes; highly polymorphic region (HPR)-deleted ISAV, or the non-pathogenic HPR0 ISAV (non-deleted HPR) ~~ISAV of the Genus Isavirus and Family Orthomyxoviridae~~.

Reference	Member	Comment	Aquatic Animals Commission response
2.3.4_1_1	**Canada**	**Comment Category:** Addition **Proposed amended text:** Infection with infectious salmon anaemia virus (ISAV) means infection with the pathogenic agent *Isavirus salaris* of the ~~Genus *Isavirus* and~~ Family *Orthomyxoviridae*. Infection with ISAV includes two genotypes; highly polymorphic region ~~(HPR)~~-deleted ISAV (HPR-deleted ISAV), or ~~the non-pathogenic~~ full-length highly polymorphic region-0 ~~HPR0~~ ISAV (~~non-deleted HPR~~ HPR0 ISAV) ~~ISAV of the Genus Isavirus and Family Orthomyxoviridae~~. **Rationale:** For consistency and clarity, the code should state the name in full then provide the acronym (in brackets) for both cases. • highly polymorphic region-deleted ISAV (HPR-deleted ISAV), • full-length highly polymorphic region-0 ISAV (HPR0 ISAV) Additionally, non-pathogenic is a qualifier, not part of the name of the strain and therefore should not be included within the name of the strain within the first paragraph. Additionally, non-pathogenic is a qualifier, not part of the name of the strain and therefore should not be included within the name of the strain within the first paragraph. It is, and is more appropriately, included within the third paragraph describing the link between HPR0 and HPR-deleted ISA.	Agreed.

HPR-deleted ISAV may cause disease in Atlantic salmon (Salmo salar), which may progress to a generalised and lethal condition characterised by severe anaemia, and variable haemorrhages and necrosis in several organs.

Detection of HPR0 ISAV has not been associated with clinical signs of disease in Atlantic salmon (Christiansen et al., 2011). A link between non-pathogenic HPR0 ISAV and pathogenic HPR-deleted ISAV has been suggested, with some disease outbreaks potentially occurring as a result of the emergence of HPR-deleted ISAV from HPR0 ISAV (Cardenas et al., 2014; Christiansen et al., 2017; Cunningham et al., 2002; Gagne & Leblanc, 2017; Mjaaland, et al., 2002).

[…]

CHAPTER 2.3.5.

INFECTION WITH INFECTIOUS HAEMATOPOIETIC NECROSIS VIRUS

1. Scope

Infection with infectious haematopoietic necrosis virus (IHNV) means infection with the pathogenic agent *Novirhabdovirus salmonid* ~~Salmonid novirhabdovirus (commonly known as infectious haematopoietic necrosis virus [IHNV]) Genus *Novirhabdovirus* and~~ of the Family *Rhabdoviridae*.

Reference	Member	Comment	Aquatic Animals Commission response
2.3.5_1_1	**China (People's Rep. Of)**	**Category:** General China supports modifying the names of disease pathogens in *Aquatic Code* and *Aquatic Manual* based on the latest changes in ICTV. This helps international standards closely follow the latest scientific developments, fully reflecting the scientific character of WOAH standards. It is recommended to add the name of the pathogen that was previously used in the standard in each disease chapter, in parentheses, so that users can understand and trace it. For example: **Proposed amended text:** Infection with infectious haematopoietic necrosis virus (IHNV) means infection with the pathogenic agent *Novirhabdovirus salmonid* (previously know as infectious haematopoietic necrosis virus, and Salmonid novirhabdovirus) ~~Salmonid novirhabdovirus (commonly known as infectious haematopoietic necrosis virus [IHNV]) Genus *Novirhabdovirus* and~~ of the Family *Rhabdoviridae*.	Agreed.

[…]

CHAPTER 2.3.6.

INFECTION WITH KOI HERPESVIRUS

Reference	Member	Comment	Aquatic Animals Commission response
2.3.6_1	**EU**	**Category:** Editorial CyHV-1 and CyHV-2 when mentioned for the first time should be fully spelled out as CyHV-1 (carp pox virus), CyHV-2 (goldfish haematopoietic necrosis virus.	Agreed.

1. Scope

Infection with koi herpesvirus (KHV) means infection with the pathogenic agent *Cyvirus cyprinidallo 3* (previously known as cyprinid herpesvirus-3 [CyHV-3]), of ~~the Genus *Cyprinivirus* in~~ the Family *Alloherpesviridae*.

Reference	Member	Comment	Aquatic Animals Commission response
2.3.6_1_1	**Canada**	**Comment Category:** change **Proposed amended text:** *Cyvirus cyprinidallo3* ~~*cyprinidallo 3*~~ **Rationale:** Remove space before 3 to be consistent with ICTV, throughout.	Agreed.

2. Disease information

2.1. Agent factors

2.1.1. Aetiological agent

KHV, also known as carp interstitial nephritis and gill necrosis virus (CNGV) (Ilouze *et al.,* 2010), has been classified as *Cyvirus cyprinidallo3* ~~cyprinid herpesvirus-3 (CyHV-3) following the nomenclature of other cyprinid herpesviruses: CyHV-1 (carp pox virus, fish papilloma virus) and CyHV-2 (goldfish haematopoietic necrosis virus)~~. Analysis of the complete genome has shown that CyHV-3 is closely related to CyHV-1, CyHV-2, anguillid herpesvirus-1 (AngHV-1) and distantly related to channel catfish virus (Ictalurid herpesvirus: IcHV-1) and Ranid (frog) herpesvirus (RaHV-1) (Waltzek *et al.,* 2005). CyHV-3 was designated the type species of the new *Cyprinivirus* genus within the *Alloherpesviridae* family, that also contains CyHV-1 and CyHV-2. However, the designation KHV has been retained in the *Aquatic Code* and *Aquatic Manual* for reasons of continuity and is used here synonymously with CyHV-3.

The size of the KHV genome is now confirmed as 295 kbp. Virus nucleocapsids have been measured at 100–110 nm in diameter and are surrounded by an envelope (Ilouze *et al.,* 2010). The enveloped virions range in size from 170 to 230 nm in the different infected cell types (Hedrick *et al.,* 2000; Miwa *et al.,* 2007; Miyazaki *et al,* 2008a). Aoki *et al.* (2007) initially described the complete genome sequence of three isolates of KHV and the genome includes 164 open reading frames (ORFs) of which 156 are unique protein-coding genes. They suggested that the finding that 15 KHV genes are homologous with genes in IcHV-1 confirms the proposed place of KHV in the family Herpesviridae. Forty viral proteins and 18 cellular proteins are incorporated into mature virions.

Engelsma *et al.* (2013) detected novel strains of cyprinid herpesvirus closely related to KHV. These strains may represent low or non-pathogenic variants of CyHV-3, but further investigation is required to establish the true genetic relationship between these strains, and KHV.

CHAPTER 2.3.8.

INFECTION WITH SALMONID ALPHAVIRUS

1. Scope

Infection with salmonid alphavirus (SAV) means infection with any genotype of the pathogenic agent Alphavirus salmon ~~SAV~~, ~~Genus *Alphavirus*~~ and Family *Togaviridae*.

Reference	Member	Comment	Aquatic Animals Commission response
2.3.8_1_1	**Thailand**	**Category:** Editorial **Proposed amended text:** "Infection with salmonid alphavirus (SAV) means infection with any genotype of the pathogenic agent *Alphavirus salmon* ~~SAV,~~ ~~Genus Alphavirus~~ ~~and~~ of the Family *Togaviridae*." **Rationale:** Scientific name of a pathogen should be italicised **Supporting Document:** –	Agreed.

[…]

CHAPTER 2.3.9.

INFECTION WITH SPRING VIRAEMIA OF CARP VIRUS

1. Scope

Infection with spring viraemia of carp virus (SVCV) means infection with the pathogenic agent *agent Sprivivirus cyprinus* ~~(commonly known as spring viraemia of carp virus [SVCV])~~, of ~~the Genus *Sprivivirus* and~~ the Family *Rhabdoviridae*.

[…]

CHAPTER 2.3.10.

INFECTION WITH VIRAL HAEMORRHAGIC SEPTICAEMIA VIRUS

1. Scope

Infection with viral haemorrhagic septicaemia virus (VHSV) means infection with the pathogenic agent ***Novirhabdovirus piscine*** ~~viral haemorrhagic septicaemia virus~~ of the ~~Genus *Novirhabdovirus* and~~ Family *Rhabdoviridae*.

[…]

CHAPTER 2.4.1.

INFECTION WITH ABALONE HERPESVIRUS

1. Scope

Infection with abalone herpesvirus (AbHV) means infection with the pathogenic agent *Aurivirus haliotidmalaco1* (previously known as *Haliotid herpesvirus 1*, and *abalone herpesvirus* [AbHV]) of ~~the genus *Aurivirus* and~~ the Family *Malacoherpesviridae*.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.1_1_1	**China (People's Rep. Of)**	**Category:** change **Proposed amended text :** Infection with abalone herpesvirus (AbHV) means infection with the pathogenic agent *Aurivirus haliotidmalaco1* (previously known as *Haliotid herpesvirus 1*~~, and *abalone herpesvirus* [AbHV]~~) of ~~the genus *Aurivirus* and~~ the Family *Malacoherpesviridae*. **Rationale:** Abalone herpesvirus (AbHV) is a common name and has never been classified as a formal taxonomic name. Therefore, it should not appear in the paragraph related to viral taxonomic status, and should not be italicised. **supporting evidence:** not relevant.	Agreed.

[…]

CHAPTER 2.4.2.

INFECTION WITH *BONAMIA EXITIOSA*

. . .

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_1	**EU**	**Category:** General The EU supports the proposed amendments and thanks the Aquatic Animals Commission for addressing the previous EU comments.	Noted.
2.4.2_2	**Australia**	**Category:** General **Proposed amended text:** n/a **Rationale**: There is a comment earlier in this report (Page 24, Item 8.2.1, notes for the September 2025 meeting) regarding *Bonamia* sp., where the Reference Laboratory expert "clarified that for these diseases, molecular methods may identify endogenous DNA and therefore should either be combined with another diagnostic method or two species-specific molecular methods targeting non-overlapping regions of the pathogen genome should be used". Australia asks if there is any evidence of Bonamia incorporation into the host genome and, if there is, that this could be provided. If there is no evidence for *Bonamia* incorporation into the host genome, claims of endogenous DNA should not be included (i.e. without reference to a peer-reviewed publication). Therefore, the requirement for two conventional PCRs and sequence analysis for a confirmed case is not justified. There is also inconsistency within the requirements for a confirmed case for apparently healthy animals and clinically affected animals. For 6.2.2.i, only 1 conventional PCR and sequence analysis is required but 6.2.2ii requires two conventional PCRs targeting different regions of the genomes and sequence analysis. Sequence requirements for a confirmed case should be consistent, regardless of the assay used to identify a suspect case **Supporting evidence:** n/a	The September 2025 Commission meeting report mistakenly stated that molecular methods may identify endogenous DNA. To confirm, molecular methods **do not** identify endogenous (oyster) DNA, but they could detect environmental *Bonamia* DNA 'retained' by the oysters during filtration. Disagreed that there is inconsistency within the requirements for a confirmed case for apparently healthy animals and clinically affected animals: the requirement for confirmation is positive results from two tests with sufficient specificity.
2.4.2_3	**Australia**	**Category:** General **Proposed amended text:** n/a **Rationale**: For most of the mollusc chapters, in Table 4.1 assays are validated to Stage 2. This includes estimates of DSe and DSp which need to be published in peer reviewed literature and added to Table 6.3.1 and/or Table 6.3.2. If there is no data in these tables, there is no peer-reviewed evidence to support claims of validation to Stage 2 (or greater). Tables 6.3.1 and 6.3.2. were added to provide the reader with some evidence of the level of validation of recommended tests – if data exists but is unpublished there is the validation template that can be used. If the AAHSC do not require published estimates of validation to support claims of Stage 2 in Table 4.1 then everyone will just add Stage 2 which defeats the purpose of what the ad hoc group and AAHSC was trying to achieve **Supporting evidence:** n/a	Noted: this comment will be addressed when the chapter is next updated. Neither Table 4.1. nor Tables 6.3.1 and 6.3.2 were circulated for comment.

6. Corroborative diagnostic criteria

This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence of clinical signs (Section 6.2.) but does not evaluate whether the infectious agent is the cause of the clinical event.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence (Section 6.2.) of clinical signs ~~(Section 6.2.)~~ but does not evaluate whether the infectious agent is the cause of the clinical event. **Rationale:** Edited so that reference to section is consistent. Supporting evidence n/a	Agreed.

The case definitions for a suspect and confirmed case have been developed to support decision-making related to trade and confirmation of disease status at the country, zone or compartment level. Case definitions for disease confirmation in endemically affected areas may be less stringent. If a Competent Authority does not have the capability to undertake the necessary diagnostic tests it should seek advice from the appropriate WOAH Reference Laboratory, and if necessary, refer samples to that laboratory for confirmatory testing of samples from the index case in a country, zone or compartment considered free.

When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_6_2	**USA**	**Category:** General **Rationale:** We agree with the addition of this sentence to Chapter 2.4.2. We also recommend adding an assay to Section 4.5. of this chapter specific for targeting non-overlapping regions.	Noted: this comment will be addressed when the chapter is next updated. Section 4.5 was not circulated for comment.
2.4.2_6_3	**Thailand**	**Category:** General We would like WOAH to consider including additional molecular test methods in the *Aquatic Manual* that target different genomic regions (other than 18S rDNA). Currently, the *Aquatic Manual* specifies conventional PCR methods targeting only 18S rDNA, which is not sufficient to meet the stated conditions. **Proposed amended text:** No text proposed **Rationale:** To ensure the requirements are clear and practical for implementation **Supporting Document:** -	Noted: this comment will be addressed when the chapter is next updated. The Section on molecular methods was not circulated for comment.

6.1. Apparently healthy animals or animals of unknown health status[1]

Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or

1 For example transboundary commodities.

equipment, etc., from a known infected population equate to an epidemiological link. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_6.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Apparently healthy animals or animals of unknown health status[1] Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Testing may also be carried out on transboundary commodities, at which time the health status of the animals from which they were derived may be of unknown. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom. **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.

6.1.1. Definition of suspect case in apparently healthy animals

The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met:

i) Observation of parasite cells in tissue imprints

ii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen

iii) Positive result by conventional PCR

iv) Positive result by real-time PCR

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_6.1.1_1	**New Zealand**	**Category:** editorial 6.1.1. Definition of suspect case in apparently healthy animals or animals of unknown health status[1] The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met:… **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.
2.4.2_6.1.1_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met: i) Observation of parasite cells in tissue imprints	Disagreed: ISH is not used for surveillance of apparently healthy animals (Table 4.1). Histopathology would be the screening method for a suspect case with ISH as a

		ii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen iii) Positive result by conventional PCR iv) Positive result by real-time PCR v) Positive result by *in-situ* hybridisation **Rationale:** None of the three existing in-situ hybridisation methods are species specific. The current *Manual* specifies that *in-situ* hybridisation is applicable for detecting suspected cases in samples with clinical signs. We recommend including this method for detecting suspected cases in apparently healthy individuals to make the *Manual* more comprehensive. **Supporting evidence:** not relevant	confirmatory method provided it has suitable specificity.

6.1.2. Definition of confirmed case in apparently healthy animals

The presence of infection with *B. exitiosa* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by tissue imprints, histology or *in-situ* hybridisation and ~~real-time~~ positive result by PCR ~~and conventional PCR and~~ combined with sequencing

ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing

~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_6.1.2_1	**USA**	**Category:** General **Rationale:** We agree with the proposed edits to 6.1.2.ii) of Chapter 2.4.2. We also recommend adding an assay to Section 4.5. of this chapter specific for targeting non-overlapping regions.	Noted: this comment will be addressed when the chapter is next updated. Section 4.5 was not circulated for comment.
2.4.2_6.1.2_2	**UK**	**Category:** Addition **Proposed amended text:** ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different ~~parts~~ regions of the genome combined with sequencing, when visualisation of parasite cells using a microscopy method is not possible **Rationale:** We suggest an editorial change to "regions" to align with other texts and methodology. We also suggest adding this text on visualisation to clarify that, while this method of using two PCRs is acceptable, visualisation of the parasite is the preferred method.	Disagreed: not necessary as visualisation methods are included in point i) and are preferred for confirming a case in apparently healthy animals.
2.4.2_6.1.2_3	**New Zealand**	**Category:** editorial **Proposed amended text:** Definition of confirmed case in apparently healthy animals or animals of unknown health status **Rationale:** Edited to make it explicit that criteria for confirmed case also applied to "animals of unknown health status" as described in 6.1.1. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.

2.4.2_6.1.2_4	**New Zealand**	**Category:** general comment **Proposed amended text:** n/a **Rationale:** We would welcome some guidance within this chapter on how to apply these diagnostic criteria, especially to species not officially assessed and listed as susceptible. In particular, we are concerned that *Bonamia exitiosa* could occur incidentally in filter feeders that co-exist with infected populations of flat oysters and be detected via molecular methods. We note that pathogen-specific positive PCR results for *Bonamia ostreae* have been reported from four other non-mollusc hosts and while not listed as susceptible, are mentioned in the *Manual* Chapter. If the criteria "…. positive results by two PCRs targeting different parts of the genome combined with sequencing" were obtained from a species not listed as susceptible, would this meet the definition of a "confirmed case" and need to be notified under Article 1.1.3? Some guidance as to under what circumstances the Reference Laboratory should be consulted would also be welcomed, as have been present previously in some mollusc disease *Manual* chapters. **Supporting evidence** n/a	Noted: this comment will be addressed when the chapter is next updated.
2.4.2_6.1.2_5	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. ~~ostreae~~ exitiosa* is considered to be confirmed if at least one of the following critera is met: i) Positive result by tissue imprints, histology or *in-situ* hybridisation and positive result by ~~real-time~~ species-specific PCR ~~and conventional PCR and~~ combined with sequencing ii) Positive results by ~~real-time~~ two species-specific PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing ~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~ **Rationale:** Three out of five real-time quantitative PCR tests and two out of three conventional PCR tests are not species specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Disagreed: sequence analysis adds the required level of specificity to these assays.
2.4.2_6.1.2_6	**Australia**	**Category:** change **Proposed amended text:** 6.1.2. Definition of confirmed case in apparently healthy animals ii) Positive results by ~~real-time~~ two real-time PCRs or *in-situ* hybridisation and conventional PCR and sequence analysis ~~and *in-situ* hybridisation~~ targeting different parts of the genome ~~combined with sequencing~~ **Rationale:** If there is no evidence of endogenous DNA, then this case definition should only require confirmatory testing using one conventional PCR method and sequence analysis of amplicons.	Disagreed: a positive result by two independent tests is required for confirmation.

		Supporting evidence: not relevant	

6.2 Clinically affected animals

Clinical signs are not pathognomonic for a single disease; however they may narrow the range of possible diagnoses.

6.2.1. Definition of suspect case in clinically affected animals

The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met:

i) Gross pathology or clinical signs associated with the disease as described in this chapter

ii) Observation of parasite cells in tissue imprints

iii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen

iv) Positive result by real-time PCR

v) Positive result by conventional PCR

vi) Positive result by *in-situ* hybridisation

6.2.2. Definition of confirmed case in clinically affected animals

The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by tissue imprints, histology or in-situ hybridisation and positive result by ~~real-time~~ PCR ~~and conventional PCR and~~ combined with sequencing

ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing

~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~

Reference	Member	Comment	Aquatic Animals Commission response
2.4.2_6.2.2_1	**USA**	**Category:** General **Rationale:** We agree with the proposed edits to 6.2.2.ii) in Chapter 2.4.2. We also recommend adding an assay to Section 4.5. of this chapter specific for targeting non-overlapping regions.	Noted: this comment will be addressed when the chapter is next updated. Section 4.5 was not circulated for comment.
2.4.2_6.2.2_2	**UK & Thailand**	**Category:** Editorial **Proposed amended text:** The presence of infection with *B. ~~ostreae~~ exitiosa* is considered to be confirmed if at least one of the following critera is met: **Rationale:** Species correction	Agreed.
2.4.2_6.2.2_3	**UK**	**Category:** Addition **Proposed amended text:** ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different ~~parts~~ regions of the genome combined with sequencing, when visualisation of parasite cells using a microscopy method is not possible **Rationale:** We suggest an editorial change to "regions" to align with other texts and	Disagreed: not necessary as visualisation methods are included in point i) and are preferred for confirming a case in clinically affected animals.

		methodology. We also suggest adding this text on visualisation to clarify that, while this method of using two PCRs is acceptable, visualisation of the parasite is the preferred method.	
2.4.2_6.2.2_4	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following critera is met: i) Positive result by tissue imprints, histology or *in-situ* hybridisation and positive result by ~~real-time~~ species-specific PCR ~~and conventional PCR and~~ combined with sequencing ii) Positive results by ~~real-time~~ two species-specific PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing ~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~ **Rationale:** Three out of five real-time quantitative PCR tests and two out of three conventional PCR tests are not species specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Disagreed: sequence analysis adds the required level of specificity to these assays.
2.4.2_6.2.2_5	**Australia**	**Category:** Change **Proposed amended text:** 6.2.2. Definition of confirmed case in clinically affected animals The presence of infection with ~~*B. ostreae*~~ *B. exitiosa* is considered to be confirmed if at least one of the following criteria is met: … ii) Positive results by ~~real-time~~ two real-time PCRs or *in-situ* hybridisation and conventional PCR and sequence analysis ~~and *in-situ* hybridisation~~ ~~targeting different parts of the genome combined with sequencing~~ **Rationale**: If there is no evidence of endogenous DNA, then this case definition should only require confirmatory testing using one conventional PCR method and sequence analysis of amplicons **Supporting evidence:** N/A	Disagreed: a positive result by two independent tests is required for confirmation.

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CHAPTER 2.4.3.

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6. Corroborative diagnostic criteria

This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence of clinical signs (Section 6.2.) but does not evaluate whether the infectious agent is the cause of the clinical event.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence (Section 6.2.) of clinical signs ~~(Section 6.2.)~~ but does not evaluate whether the infectious agent is the cause of the clinical event. **Rationale:** Edited so that reference to section is consistent. Supporting evidence n/a	Agreed.

The case definitions for a suspect and confirmed case have been developed to support decision making related to trade and confirmation of disease status at the country, zone or compartment level. Case definitions for disease confirmation in endemically affected areas may be less stringent. If a Competent Authority does not have the capability to undertake the necessary diagnostic tests it should seek advice from the appropriate WOAH Reference Laboratory, and if necessary, refer samples to that laboratory for confirmatory testing of samples from the index case in a country, zone or compartment considered free.

When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6_1	**USA**	**Category:** General **Rationale:** We agree with the addition of this sentence to Chapter 2.4.3. We also recommend adding an assay to Section 4.5. of this chapter specific for targeting non-overlapping regions.	Noted: this comment will be addressed when the chapter is next updated. Section 4.5 was not circulated for comment.
2.4.2_6_2	**Thailand**	**Category:** General We would like WOAH to consider including additional molecular test methods in the *Aquatic Manual* that target different genomic regions (other than 18S rDNA). Currently, the *Aquatic Manual* specifies conventional PCR methods targeting only 18S rDNA, which is not sufficient to meet the stated conditions. **Proposed amended text:** No text proposed **Rationale:** To ensure the requirements are clear and practical for implementation **Supporting Document:** -	Noted: this comment will be addressed when the chapter is next updated. The Section on molecular methods was not circulated for comment.

6.1. Apparently healthy animals or animals of unknown health status[2]

Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** Apparently healthy animals or animals of unknown health status[1] Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Testing may also be carried out on transboundary commodities, at which time the health status of the animals from which they were derived may be of unknown. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom. **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown.

6.1.1. Definition of suspect case in apparently healthy animals

The presence of infection with *B. ostreae* shall be suspected if at least one of the following criteria is met:

i) Observation of parasite cells in tissue imprints

ii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen

iii) Positive result by conventional PCR

iv) Positive result by real-time PCR

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6.1.1_1	**New Zealand**	**Category:** editorial **Proposed amended text:** 6.1.1. Definition of suspect case in apparently healthy animals or animals of unknown health status[1] The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met:… **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown.

2 For example transboundary commodities.

2.4.3_6.1.1_2	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met: i) Observation of parasite cells in tissue imprints ii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen iii) Positive result by conventional PCR iv) Positive result by real-time PCR v) Positive result by *in-situ* hybridisation **Rationale:** We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this *Manual* more comprehensive. **Supporting evidence:** not relevant	Disagreed: ISH is not used for screening populations. Histopathology would be the screening method for a suspect case with ISH as a confirmatory method provided it has suitable specificity. .

6.1.2. Definition of confirmed case in apparently healthy animals

The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by tissue imprints, histology or *in-situ* hybridisation and ~~real-time~~ positive result by PCR ~~and conventional PCR and~~ combined with sequencing

ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing

~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6.1.2_1	**USA**	**Category:** General **Rationale:** We agree with the proposed edits to 6.1.2.ii) of Chapter 2.4.3. We also recommend adding an assay to Section 4.5. of this chapter specific for targeting non-overlapping regions.	Noted: this comment will be addressed when the chapter is next updated. Section 4.5 was not circulated for comment.
2.4.3_6.1.2_2	**UK**	**Category:** Addition **Proposed amended text:** ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different ~~parts~~ regions of the genome combined with sequencing, when visualisation of parasite cells using a microscopy method is not possible **Rationale:** We suggest an editorial change to "regions" to align with other texts and methodology. We also suggest adding this text on visualisation to clarify that, while this method of using two PCRs is acceptable, visualisation of the parasite is the preferred method.	Disagreed: not necessary as visualisation methods are included in point i) and are preferred for confirming a case in apparently healthy animals. .
2.4.3_6.1.2_3	**New Zealand**	**Category:** editorial **Proposed amended text:** Definition of confirmed case in apparently healthy animals or animals of unknown health status	Disagreed as the Section covers apparently healthy animals, the health status of which is unknown.

		Rationale: It was unclear whether these criteria for confirmed case also applied to "animals of unknown health status" as described in 6.1.1 – if so we think it should be specified. **Supporting evidence** n/a	
2.4.2_6.1.2_4	**New Zealand**	**Category:** general comment **Proposed amended text:** n/a **Rationale:** As commented in the *Bonamia exitiosa* chapter, we would welcome some guidance within this chapter on how to apply these diagnostic criteria, especially to species not officially assessed and listed as susceptible. In particular, we are concerned that *Bonamia ostreae* could occur incidentally in filter feeders that co-exist with infected populations of flat oysters and be detected via molecular methods. We note that pathogen-specific positive PCR results for *Bonamia ostreae* have been reported from four other non-mollusc hosts and while not listed as susceptible, are mentioned in the *Manual* Chapter. We would like to understand if the criteria "…. positive results by two PCRs targeting different parts of the genome combined with sequencing" were obtained from a species not listed as susceptible, would this meet the definition of a "confirmed case" and need to be notified under Article 1.1.3 ? Some guidance as to under what circumstances the Reference Laboratory should be consulted would also be welcomed. **Supporting evidence** n/a	Noted: this comment will be addressed when the chapter is next updated.
2.4.3_6.1.2_5	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following critera is met: i) Positive result by tissue imprints, histology or *in situ* hybridisation and ~~real-time~~ positive result by species-specific PCR ~~and conventional PCR~~ and combined with sequencing ii) Positive results by ~~real-time~~ two species-specific PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing ~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~ **Rationale:** Three out of five real-time quantitative PCR tests and two out of three conventional PCR tests are not species specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Disagreed: sequence analysis adds the required level of specificity to these assays.
2.4.3_6.1.2_6	**Australia**	**Category:** Change **Proposed amended text:** 6.1.2. Definition of confirmed case in apparently healthy animals … ii) Positive results by ~~real-time~~ two real-time PCRs or	Disagreed: a positive result by two independent tests is required for confirmation.

		in-situ hybridisation and conventional PCR and sequence analysis ~~and *in-situ* hybridisation~~ ~~targeting different parts of the genome~~ ~~combined with sequencing~~ **Rationale**: If there is no evidence of endogenous DNA, then this case definition should only require confirmatory testing using one conventional PCR method and sequence analysis of amplicons. **Supporting evidence:** N/A	

6.2 Clinically affected animals

Clinical signs are not pathognomonic for a single disease; however they may narrow the range of possible diagnoses.

6.2.1. Definition of suspect case in clinically affected animals

The presence of infection with *B. ostreae* shall be suspected if at least one of the following criteria is met:

i) Gross pathology or clinical signs associated with the disease as described in this chapter

ii) Observation of parasite cells in tissue imprints

iii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen

iv) Positive result by real-time PCR

v) Positive result by conventional PCR

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6.2.1_1	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. exitiosa* shall be suspected if at least one of the following criteria is met: i) Gross pathology or clinical signs associated with the disease as described in this chapter ii) Observation of parasite cells in tissue imprints iii) Observation of parasite cells in tissue sections with or without histopathology characteristic of the pathogen iv) Positive result by real-time PCR v) Positive result by conventional PCR vi) Positive result by *in-situ* hybridisation **Rationale:** We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this *Manual* more comprehensive. **Supporting evidence:** not relevant	Disagreed: ISH is not used for screening populations. Histopathology would be the screening method for a suspect case with ISH as a confirmatory method provided it has suitable specificity..

6.2.2. Definition of confirmed case in clinically affected animals

The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following critera is met:

i) Positive result by tissue imprints, histology or in-situ hybridisation and positive result by ~~real-time~~ PCR ~~and conventional PCR and~~ combined with sequencing

ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing

~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~

Reference	Member	Comment	Aquatic Animals Commission response
2.4.3_6.2.2_1	**USA**	**Category:** General **Rationale:** We agree with the proposed edits to 6.2.2.ii) of Chapter 2.4.3. We also recommend adding an assay to Section 4.5. of this chapter specific for targeting non-overlapping regions.	Noted: this comment will be addressed when the chapter is next updated. Section 4.5 was not circulated for comment.
2.4.3_6.2.2_2	**UK**	**Category:** Addition **Proposed amended text:** ii) Positive results by ~~real-time~~ two PCRs ~~and *in-situ* hybridisation~~ targeting different ~~parts~~ regions of the genome combined with sequencing, when visualisation of parasite cells using a microscopy method is not possible **Rationale:** We suggest an editorial change to "regions" to align with other texts and methodology. We also suggest adding this text on visualisation to clarify that, while this method of using two PCRs is acceptable, visualisation of the parasite is the preferred method.	Disagreed: not necessary as visualisation methods are included in point i) and are preferred for confirming a case in clinically affected animals..
2.4.2_6.2.2_3	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following critera is met: i) Positive result by tissue imprints, histology or *in-situ* hybridisation and positive result by ~~real-time~~ species-specific PCR ~~and conventional PCR and~~ combined with sequencing ii) Positive results by ~~real-time~~ two species-specific PCRs ~~and *in-situ* hybridisation~~ targeting different parts of the genome combined with sequencing ~~iii) Positive result by tissue imprints, histology or *in-situ* hybridisation, and positive result by conventional PCR followed by sequence analysis~~ **Rationale:** Three out of five real-time quantitative PCR tests and two out of three conventional PCR tests are not species specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Disagreed: sequence analysis adds the required level of specificity to these assays.
2.4.2_6.2.2_4	**Australia**	**Category:** Change **Proposed amended text:** 6.2.2. Definition of confirmed case in clinically affected animals The presence of infection with *B. ostreae* is considered to be confirmed if at least one of the following criteria is met:	Disagreed: a positive result by two independent tests is required for confirmation.

		… ii) Positive results by ~~real-time~~ ~~two~~ real-time PCR~~s~~ or *in-situ* hybridisation and conventional PCR and sequence analysis ~~and *in-situ* hybridisation~~ ~~targeting different parts of the genome~~ ~~combined with sequencing~~ **Rationale:** If there is no evidence of endogenous DNA, then this case definition should only require confirmatory testing using one conventional PCR method and sequence analysis of amplicons. **Supporting evidence:** N/A	

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CHAPTER 2.4.4.

INFECTION WITH *MARTEILIA REFRINGENS*

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Reference	Member	Comment	Aquatic Animals Commission response
2.4.4_1	**EU**	**Category:** General The EU supports the proposed amendments.	Noted.
2.4.4_2	**Australia**	**Category:** General **Proposed amended text:** n/a **Rationale**: For this chapter on *Marteilia*, there seems to have been a cut and paste from the *Bonamia* chapter as a confirmed case also requires positive results by two PCRs targeting different parts of the genome combined with sequencing but there is no mention of endogenous DNA in this chapter. **Supporting evidence:** n/a	The September 2025 Commission meeting report mistakenly stated that molecular methods may identify endogenous DNA. To confirm, molecular methods **do not** identify endogenous (oyster) DNA, but they could detect environmental *Marteilia* DNA 'retained' by the oysters during filtration. .
2.4.4_3	**Australia**	**Category:** General **Proposed amended text:** n/a **Rationale**: For most of the mollusc chapters, in Table 4.1 assays are validated to Stage 2. This includes estimates of DSe and DSp which need to be published in peer reviewed literature and added to Table 6.3.1 and/or Table 6.3.2. If there is no data in these tables, there is no peer-reviewed evidence to support claims of validation to Stage 2 (or greater). Tables 6.3.1 and 6.3.2. were added to provide the reader with some evidence of the level of validation of recommended tests – if data exists but is unpublished there is the validation template that can be used. If the AAHSC do not require published estimates of validation to support claims of Stage 2 in Table 4.1 then everyone will just add Stage 2 which defeats the purpose of what the ad hoc group was trying to achieve. **Supporting evidence:** n/a	Noted: this comment will be addressed when the chapter is next updated. Neither Table 4.1. nor Tables 6.3.1 and 6.3.2 were circulated for comment.

6. Corroborative diagnostic criteria

This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence of clinical signs (Section 6.2.) but does not evaluate whether the infectious agent is the cause of the clinical event.

The case definitions for a suspect and confirmed case have been developed to support decision making related to trade and confirmation of disease status at the country, zone or compartment level. Case definitions for disease confirmation in endemically affected areas may be less stringent. If a Competent Authority does not have the capability to undertake the necessary diagnostic tests it should seek advice from the appropriate WOAH Reference Laboratory, and if necessary, refer samples to that laboratory for confirmatory testing of samples from the index case in a country, zone or compartment considered free.

When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.4_6_1	**New Zealand**	**Category:** editorial **Proposed amended text:** This section only addresses the diagnostic test results for detection of infection in the absence (Section 6.1.) or in the presence (Section 6.2.) of clinical signs ~~(Section 6.2.)~~ but does not evaluate whether the infectious agent is the cause of the clinical event. **Rationale:** the reference to Section 6.2 should be moved after "presence" to make more sense and be consistent with how reference to Section 6.1 was used. Supporting evidence n/a	Agreed. .
2.4.4_6_2	**Australia**	**Category:** change **Proposed amended text:** ~~When two molecular methods are used for confirmation of a case, it is preferable to use two species-specific methods targeting non-overlapping regions of the pathogen genome.~~ **Rationale:** This seems to be a cut and paste error from the Bonamia chapters. Supporting evidence n/a	Disagreed: this is standard text that has been added to the chapter template (not necessarily for *Bonamia* chapters only).

6.1. Apparently healthy animals or animals of unknown health status[1]

Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population, equate to an epidemiological link. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom.

Reference	Member	Comment	Aquatic Animals Commission response
2.4.4_6.1_2	**New Zealand**	**Category:** general comment **Proposed amended text:** Apparently healthy animals or animals of unknown health status[1] Apparently healthy populations may fall under suspicion, and therefore be sampled, if there is an epidemiological link(s) to an infected population. Hydrographical proximity to, or movement of animals or animal products or equipment, etc., from a known infected population equate to an epidemiological link. Testing may also be carried out on transboundary commodities, at which time the health status of the animals from which they were derived may be of unknown. Alternatively, healthy populations are sampled in surveys to demonstrate disease freedom. **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.

1 For example transboundary commodities.

		health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	

6.1.1. Definition of suspect case in apparently healthy animals

The presence of infection with *M. refringens* shall be suspected if at least one of the following criteria is met:

i) Positive result by a recommended molecular detection test

ii) Visual observation of the pathogen by microscopy

Reference	Member	Comment	Aquatic Animals Commission response
2.4.4_6.1.1_1	**UK**	**Category:** Change **Proposed amended text:** ~~i) Positive result by a recommended molecular detection test~~ ~~ii) Visual observation of the pathogen by microscopy~~ i) Positive result by wet mounts ii) Positive result by tissue imprints iii) Positive result by histopathology iv) Positive result by real-time PCR v) Positive result by conventional PCR **Rationale:** We suggest this more detailed list, in line with section 6.2.1 of this chapter, for clarity.	Partially agreed, included all the diagnostic methods in Table 4.1. Wet mounts not included as it is not in Table 4.1.
2.4.4_6.1.1_2	**New Zealand**	**Category:** general comment **Proposed amended text:** 6.1.1. Definition of suspect case in apparently healthy animals or animals of unknown health status[1] The presence of infection with *M. refringens* shall be suspected if at least one of the following criteria is met:… **Rationale:** As commented in other mollusc disease chapters, while the concept of "apparently healthy populations" was explained in the text, the concept of "animals of unknown health status" was buried in a footnote. Suggest moving this detail up into the text. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.

6.1.2. Definition of confirmed case in apparently healthy animals

The presence of infection with *M. refringens* is considered to be confirmed if at least one of the following ~~criterion~~ criteria is met:

i) Positive result by tissue imprints, histology or *in-situ* hybridisation and ~~real-time~~ positive result by PCR ~~and conventional PCR followed by sequence analysis~~ combined with sequencing

ii) Positive results by two PCRs targeting different parts of the genome combined with sequencing

Reference	Member	Comment	Aquatic Animals Commission response

2.4.4_6.1.2_1	**UK**	**Category:** Addition **Proposed amended text:** ii) Positive results by two PCR assays targeting different regions of the genome combined with nucleotide sequencing, when visualisation of parasite cells using a microscopy method is not possible **Rationale:** This additional text clarifies that visualisation of the parasite cells should be done if possible, and that utilising two PCR results as detailed here, while acceptable, is not the preferred methodology.	Disagreed: not necessary as visualisation methods are included in point i) and are preferred for confirming a case in apparently healthy animals.
2.4.4_6.1.2_2	**New Zealand**	**Category:** editorial **Proposed amended text:** Definition of confirmed case in apparently healthy animals or animals of unknown health status **Rationale:** Edited to make it explicit that criteria for confirmed case also applied to "animals of unknown health status" as described in 6.1.1. **Supporting evidence** n/a	Disagreed: the Section covers apparently healthy animals, the health status of which is unknown.
2.4.4_6.1.2_3	**New Zealand**	**Category:** general comment **Proposed amended text:** n/a **Rationale:** As commented in the *Bonamia* sp. chapters, we would welcome some guidance within this chapter on how these diagnostic criteria apply to species not officially assessed and listed as susceptible. Particularly if the criteria if positive results by two PCRs targeting different parts of the genome combined with sequencing are obtained from other species, in the absence of clinical disease. We note that pathogen-specific positive PCR results for *M. refringens* have been reported from other non-mollusc hosts and while not listed as susceptible, are mentioned in the *Manual* Chapter. Some guidance as to under what circumstances the Reference Laboratory should be consulted would also be welcomed, as have been present previously in some mollusc disease *Manual* chapters. **Supporting evidence** n/a	Noted: this comment will be addressed when the chapter is next updated.
2.4.4_6.1.2_4	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *M. refringens* is considered to be confirmed if at least one of the following ~~criterion~~ criteria is met: i) Positive result by tissue imprints, histology or *in-situ* hybridisation and ~~real-time~~ positive result by species-specific PCR ~~and conventional PCR followed by sequence analysis~~ combined with sequencing ii) Positive results by two species-specific PCRs targeting different parts of the genome combined with sequencing **Rationale:** One out of three real-time quantitative PCR tests and all three conventional PCR tests are not species-	Disagreed: sequence analysis adds the required level of specificity to these assays.

		specific. Therefore, species-specific PCR should be emphasized when confirming cases.. **Supporting evidence:** not relevant	
2.4.4_6.1.2_5	**Australia**	**Category:** editorial **Proposed amended text:** ii) Positive results by ~~two~~ conventional PCR~~s targeting different parts of the genome~~ combined with sequencing **Rationale:** What evidence is there to justify the case definition at 6.1.2.ii)? There is a comment earlier in this report (Page 24, Item 8.2.1, notes for the September 2025 meeting) regarding *Bonamia* sp., where the Reference Laboratory expert "clarified that for these diseases, molecular methods may identify endogenous DNA and therefore should either be combined with another diagnostic method or two species-specific molecular methods targeting non-overlapping regions of the pathogen genome should be used". This same logic has not been stated anywhere for *Marteilia* sp. Further, what is the evidence for this? **Supporting evidence** n/a	Partially agreed, did not agree to include "conventional" before PCR because there are other available PCR methods. Agreed to remove "targeting different parts of the genome", this information is already included in the text. .

6.2 Clinically affected animals

Clinical signs are not pathognomonic for a single disease; however, they may narrow the range of possible diagnoses.

6.2.1. Definition of suspect case in clinically affected animals

The presence of infection with *M. refringens* shall be suspected if at least one of the following criteria is met:

i) Positive result by wet mounts

ii) Positive result by tissue imprints

iii) Positive result by histopathology

iv) Positive result by real-time PCR

v) Positive result by conventional PCR

Reference	Member	Comment	Aquatic Animals Commission response
2.4.4_6.2.1_1	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *M. refringens* shall be suspected if at least one of the following criteria is met: i) Positive result by wet mounts ii) Positive result by tissue imprints iii) Positive result by histopathology iv) Positive result by real-time PCR v) Positive result by conventional PCR vi) Positive result by *in-situ* hybridisation	Disagreed: ISH is not used for screening populations. Histopathology would be the screening method for a suspect case with ISH as a confirmatory method provided it has suitable specificity.

		Rationale: We recommend including the *in-situ* hybridisation method as an applicable approach for suspected cases to make this *Manual* more comprehensive. **Supporting evidence:** not relevant	

6.2.2. Definition of confirmed case in clinically affected animals

The presence of infection with *M. refringens* is considered to be confirmed if at least one of the following criteria is met:

i) Positive result by tissue imprints, histology or *in-situ* hybridisation and ~~real-time~~ positive result by PCR ~~and conventional PCR followed by sequence analysis~~ combined with sequencing

ii) Positive results by ~~species-specific ISH and conventional~~ two PCRs ~~followed by sequence analysis~~ targeting different parts of the genome combined with sequencing

~~iii) Positive result of real-time PCR followed by species-specific ISH~~

Reference	Member	Comment	Aquatic Animals Commission response
2.4.4_6.2.2_1	**China (People's Rep. of)**	**Category:** change **Proposed amended text:** The presence of infection with *M. refringens* is considered to be confirmed if at least one of the following critera is met: i) Positive result by tissue imprints, histology or *in-situ* hybridisation and ~~real-time~~ positive result by species-specific PCR ~~and conventional PCR followed by sequence analysis~~ combined with sequencing ii) Positive results by ~~species-specific ISH and conventional~~ two species-specific PCRs ~~followed by sequence analysis~~ targeting different parts of the genome combined with sequencing ~~iii) Positive result of real-time PCR followed by species-specific ISH~~ **Rationale:** One out of three real-time quantitative PCR tests and all three conventional PCR tests are not species-specific. Therefore, species-specific PCR should be emphasised when confirming cases. **Supporting evidence:** not relevant	Disagreed: sequence analysis adds the required level of specificity to these assays.
2.4.4_6.2.2_2	**Australia**	**Category:** general **Proposed amended text:** ii) Positive result~~s~~ by ~~two~~ conventional PCR~~s~~ ~~targeting different parts of the genome~~ combined with sequencing **Rationale:** What evidence is there to justify the case definition at 6.2.2.ii)? There is a comment earlier in this report (Page 24, Item 8.2.1, notes for the September 2025 meeting) regarding *Bonamia* sp., where the Reference Laboratory expert "clarified that for these diseases, molecular methods may identify endogenous DNA and therefore should either be combined with another diagnostic method or two species-specific molecular methods targeting non-overlapping regions of the pathogen genome should be used". This same logic	Partially agreed, did not agree to include "conventional" before PCR because there are other available PCR methods. Agreed to remove "targeting different parts of the genome", this information is already included in the text.

		has not been stated anywhere for *Marteilia sp.* Further, what is the evidence for this? **Supporting evidence:** N/A	

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