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ANNEX 3

EU COMMENTS

ON THE PROPOSED CHANGES TO THE

OIE MANUAL OF DIAGNOSTIC TESTS AND VACCINES FOR TERRESTRIAL ANIMALS

PRESENTED FOR COMMENTS IN OCTOBER 2016

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

SUMMARY:

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

Glossary

General comments

The EU can support the proposed changes to the glossary.

Specific comments

None.

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CHAPTER 1.1.5.: Quality management in veterinary testing laboratories

General comments

The EU can in general support this revised chapter and has a few specific comments.
In addition, we would like to point out that ISO 17011 and 17025 standards are currently being revised and new versions are expected to be published in 2017. This should preferably be taken into account before adopting this revised chapter.

Specific comments

LINES 32-40: While in general supporting the addition of the examples in points i) to vi), there might be misunderstandings as to the role of the laboratory in e.g. declaration of disease freedom the way these are drafted. Perhaps it would be better to insert the word "for" after the word "e.g." in points ii), v) and vi).
Furthermore, in **LINE 36**, the EU suggests expanding the example for the level of risk, e.g. vaccination vs. culling or slaughter.

LINES 47-48: We suggest mentioning that equivalent quality management systems would also be acceptable for OIE Reference Laboratories, e.g. by inserting the words "or equivalent" after "use of ISO/IEC 17025 (ISO/IEC, 2005)".

LINE 68: Please insert "(ISO/IEC, 2012)" after "such as ISO 15189", and include a reference to that ISA standard in the reference section.

LINES 115: The EU questions the pertinence of including veterinary practices in this list of veterinary testing facilities, as these would usually not be accredited to ISI/IEC 17025.

LINES 116-117: The statement "*If new testing methods are introduced they must be assessed and accredited before they can be aded to the scope*" is not quite correct. Indeed, most ISO 17025 systems have an option of flexible scope, whereby the laboratory is assessed as competent to add tests to the scope, and then they are formally added at the next accreditation visit.

LINE 149: Please delete ":2010" after "17043", as this is not necessary.

LINE 166: We suggest inserting the words "that have been updated in recent years" after "Disease-specific chapters", as not all these Manual chapters do already contain the fitness for purpose table.

LINE 266: Please insert "17025" in the parentheseis, for it to read as follows: "(ISO/IEC 17025, 2005)".

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CHAPTER 2.1.6.: Echinococcosis/hydatidosis (infection with *Echinococcus granulosus* and with *E. multilocularis*)

General comments

The EU can in general support this revised chapter and has a few specific comments.
However, as the chapter was written around 30 years ago and was amended only a few times, it in general seems rather outdated. It would thus benefit from a more thorough review in the near future. The EU would be pleased to offer the technical expertise of its Reference Laboratory experts to assist in this task.

Specific comments

LINE 2: Please replace the title of the chapter as follows, which reflects better the disease and pathogen names:
"CYSTIC AND ALVEOLAR ECHINOCOCCOSIS ~~/ HYDATIDOSIS~~ (INFECTION WITH *ECHINOCOCCUS GRANULOSUS* SENSU LATO AND WITH *E. MULTILOCULARIS*)"

LINE 6: Please replace "demonstration" by "detection", which is the correct term.

LINE 16: Please delete "in the" which are repeated in **LINE 17** (editorial).

LINES 11-23: We note that the part of the introduction describing the taxonomy of the genus *Echinococcus* now only mentions *E. granulosus*, whereas *E. multilocularis* is not mentioned any more at all, which is somewhat strange as the chapter covers both.

LINES 29-30: The EU invites the OIE to include molecular detection methods in this chapter (i.e., real time PCR, magnetic capture techniques).

LINE 34: The mention of Arecoline clearly shows the need for a deep review of this chapter. Indeed, that molecule is unacceptable nowadays because of potential lethal adverse reactions in dogs. It has been replaced more than 30 years ago by Praziquantel.

LINE 41: The mention of cats should be deleted, as this is not correct.

LINE 44: The word "specialised" should be deleted, as PCR nowadays is performed in most laboratories.

LINES 46-49: Serological tests should be discouraged. The main limitations are sensitivity in low worm burden and specificity for cross reactions.

LINE 82: Table 1 should be completed with all species/genotypes currently identified.

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LINE 96: The text is a bit unclear, as it seems to suggest that bladders of up to 30 cm in size can be found in the lungs, which is not correct.

LINES 98-107: The geographical distribution should be revised completely, as it is outdated and thus does not reflect the current situation.

LINES 109-110: *V. ferrilata* and *Canis latrans* should be deleted, as these are not wild definite hosts.

LINE 121: The word "sporadically" should be replaced by "mainly", as the Northern Hemisphere is indeed where the parasite is mainly found.

LINE 125: There is also a rare possibility that dogs may also be infected with the larval (metacestode) stage of *E. multilocularis* with lesions in internal organs. This may be simultaneous with adult parasite infection within the gastro-intestinal tract (Deplazes & Eckert, 2001).
Reference:
DEPLAZES P., and ECKERT J. (2001). Veterinary aspects of alveolar echinococcosis – a zoonosis of public health significance. Vet. Parasitol., 98, 65-87.

LINE 178: In the context of the first sentence of this paragraph, meat inspection and post-mortem are the same. Therefore, the words "or post-mortem" should be deleted.

LINE 346: The EU queries why the section 1.4. on arecoline is maintained at all, if its use as control agent has been superseded by praziquantel (as stated in the second sentence).

LINE 520: Table 3 should be updated with newer primers, for instance Boubaker 2013, Boufana 2013, Dinkel 2010, Knapp 2014, Isaksson 2014.

LINE 523: It is suggested to delete "*E. multilocularis*" before "eggs", for the sentence to read: "[...] PCR-amplified DNA from eggs present in faeces".

LINE 538 To reflect the development of new methods, we suggest adding a new method which is being used by several member countries for screening purposes, as follows:
"In practice, it is recommended to screen definitive hosts (e.g. foxes) using the coproantigen test and confirm with the PCR DNA test. For E. multilocularis, a semi-automated magnetic capture probe DNA extraction method and real time hydrolysis PCR (Isaksson *et al*., 2014) is also being used for screening definitive hosts."
Reference:
ISAKSSON M., HAGSTRÖM Å., ARMUA-FERNANDEZ M.T., WAHLSTRÖM H., ÅGREN E.O., MILLER A., HOLMBERG A., LUKACS M., CASULLI A., DEPLAZES P., and JUREMALM M. (2014). A semi-automated magnetic capture probe based DNA extraction and real-time PCR method applied in the Swedish surveillance of Echinococcus multilocularis in red fox (Vulpes vulpes) faecal samples. Parasites & Vectors, 7:583.

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LINE 539: Section 2.2.2. should be updated with new methodologies currently available.

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CHAPTER 2.1.8.: Foot and mouth disease (Infection with Foot and mouth disease virus)

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 3: The EU suggests aligning the spelling of the name of the virus with the one used by the International Committee on Taxonomy of Viruses (ICTV), i.e. "*Foot-and-mouth disease virus*".

LINES 28-29: The wording "appropriate level of bio-containment, determined by risk analysis" is rather vague and can open the possibility to work with FMD in laboratory/vaccine production facilities with a very low standard. It is thus suggested to amend as follows:
"[…] appropriate level of bio-containment, determined by risk analysis in accordance with Chapter 1.1.4.". (The same comment is valid for **LINES 66-67**.)

LINE 123: It seems necessary to be more precise as regards zoonotic risk of FMD. While it is true that the zoonotic risk from FMD is not very high, human infections are possible, even if extremely rare (usually in the laboratory setting) and usually benign. Therefore, it is suggested to amend the sentence as follows:
"FMD virus is ~~not~~ considered a very low zoonotic risk. Human infections are extremely rare and benign.".

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CHAPTER 2.1.19.: Rinderpest (Infection with Rinderpest virus)

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 47: Also research laboratories handling live RPV need to be approved by FAO/OIE, not only vaccine manufacturing laboratories. Therefore, the word "approved" should be moved before the word "research".

LINE 92: As the process has now been established, we propose amending the sentence as follows:
"[…] FAO and OIE have collaboratively established the principle of […]"

LINE 196: Please replace "registration of the laboratory with" by "approval of the laboratory by", as this is the appropriate terminology (as used in line 200 and elsewhere).

LINE 312: We note that a new paragraph break was introduced compared to the current version of the chapter. This separates the old method for RNA extraction from the new one (column purification). These changes should be accompanied by a repeated reference to the article of Forsyth & Barrett, 1995. Indeed, in the present state, it is unclear which primer sets should be used for performing the RT-PCR assay.

Line 333: A reference for the real-time RT-PCR assay for RPV should be added.

LINES 377 and **392**: There is a contradiction between "A competitive ELISA is no longer commercially available" and "Kits will continue to be available commercially". It is suggested to delete the latter sentence.

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CHAPTER 2.1.20.: Trichinellosis (Infection with *Trichinella* spp.)

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 159: Please replace the word "baggage" by "luggage", as this is the appropriate term.

LINE 74: The time period referred here should be "1 week or longer" instead of "3 weeks or longer". Indeed, in highly infected animals or humans, seroconversion is observed 3 weeks after the infection, i.e. 1 week after muscle larvae become infective in pigs.

LINE 126: The reference (Pozio & Zarlenga, 2013) should be moved to the end of the sentence, i.e. after the word "freezing".

LINE 134: Information on *T. pseudospiralis* should be inserted here (i.e., the deleted sentence in lines 120-123 should be inserted after "Non-encapsulated taxa include the following:").

LINE 137: ", South Africa" should be inserted after "Zimbabwe"; the word "and" before "in monitor lizards" should be deleted; and ", and in lions of South Africa" should be added at the end of the sentence (i.e., after "of Zimbabwe").

LINE 140: It is suggested to add the relevant species, for the sentence to read as follows:
"Most species (*T. spiralis*, *T. nativa*, *T. britovi*, *T. pseudospiralis*, *T. murrelli*, *T. nelsoni*, and *T. papuae*) and the *Trichinella* T6 genotypes have been detected in humans […]".

LINE 143: Please insert ", *T. nelson*" after "*T. britovi*".

LINE 175: The title of table 1 should be modified as follows, since most of the information available is only derived from pigs:
"Test methods available for *Trichinella* infections in pigs ~~trichinellosis~~ and their purpose ".

LINE 247: The following should be added at the end of the sentence:
", but the sensitivity is lower than that of the gold standard method since the serine-protease works at 60 °C killing the larvae. Dead larvae are more difficult to be recognised in the sediment than live larvae, which are more easily recognized due to their movements."

LINE 384: Please replace "trichinellosis" by "*Trichinella* infections" (consistency).

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LINE 385: Please add the following at the end of the sentence (after the reference to Gamble *et al*., 2004):
", but until now only ELISA and Western blotting have been validated according to the OIE guidelines (Gomez Morales *et al*., 2012; 2014; 2015; 2016).".
Consequently, the following sentence should be deleted (**LINES 385-388**):
"Methods include immunofluorescence assay (IFA), immuno-electrotransfer blot (IEBT), Western blot, enzyme immunohistochemical assays, and enzyme-linked immunosorbent assays (ELISA). Except for the ELISA and Western blot (Gomez Morales *et al*., 2012; 2014; 2015; 2016), these tests have not been standardised."

LINES 392-393: The last part of the paragraph should be amended as follows:
"; it is not reliable for assuring food safety of individual animals (pigs, horses or other animals)."

LINE 406: Given the addition of false positive results as an additional disadvantage, the words "A disadvantage" should be replaced by "Disadvantages", and "is" by "are".

LINE 409: We suggest replacing the sentence starting with "This is primarily" by the following:
"The false-negative results are due to the lag time of the immune response following the ingestion of infective larvae; whereas, false-positive results are due to the cross-reactions with other microorganisms."

LINE 430: The words "the quality of" should be deleted, as it is not only the quality that matters here.

LINE 440: Please insert the word "all" before "*Trichinella*-infected animals".

LINE 441: This sentence should be moved down to the end of the paragraph, and slightly modified to read as follows:
"ES antigen preparations have been developed that provide a high degree of specificity for *Trichinella* infection in pigs (Gamble *et al*., 1988)."

LINE 450: Please replace the word "However" by "Moreover".

LINE 462: Please replace "18-20 hours" by "not more than 18 hours", in order to be in line with what is recommended in the following paragraph.

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CHAPTER 2.2.5.: Infestation with *Aethina tumida* (small hive beetle)

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

Line 25: Please delete the word "Murray" after "*Aethina tumida*", as this is not necessary.

LINE 34: Please add a reference mentioning other SHB cases, and change the reference concerning the Italian SHB cases, as follows:
"Since then it has spread to Canada and a number of countries in South and Central America. *Aethina tumida* has also been found in Egypt, Australia and the Philippines. Cases have been recently identified in the South of Italy (OIE WAHIS Interface, database accessed on 06/10/2016) (Palmeri *et al*. 2015)".
Indeed, a reference is lacking concerning the SHB cases found in Egypt, Australia and the Philippines.
Reference:
Palmeri, V., G. Scirto, A. Malacrino, F. Laudani, and O. Campolo (2015). A scientific note on a new pest for European honeybees: first report of small hive beetle *Aethina tumida* (*Coleoptera*: *Nitidulidae*) in Italy. Apidologie 46(4): 527-529.

LINE 42: References concerning the SHB biology should be updated as new publications have been published. The following references should be added:
De Guzman L.I. and Frake A.M. (2007). Temperature affects *Aethina tumida* (*Coleoptera*: *Nitidulidae*) Development. Journal of Apicultural Research 46(2): 88-93.
Cuthbertson, A. G., M. E. Wakefield, M. E. Powell, G. Marris, H. Anderson, G. Budge, J. J. Mathers, L. F. Blackburn, and M. A. Brown (2013). The small hive beetle Aethina tumida: A review of its biology and control measures. Current Zoology 59:644-653.

LINE 45: It is suggested to clarify where the larvae pupate, e.g. by inserting the words "in the soil" after "to pupate".

LINE 73: This sentence sounds odd. Please replace by "The adults are dark brown to black".

LINE 102: The title of section 1.3. should be specified. We suggest replacing it by "Visual inspection", as this adds clarity.

LINES 105-141: These should be deleted (starting from "Detailed instructions […]"). Indeed, the procedure described is a time-consuming method of investigation which is hard to conduct for routine diagnosis of SHB at the apiary level. Indeed, it can induce disorder and robbing (EFSA, 2015). It should be deleted from the Manual. Complementary details should be added

to complete the description of the visual examination method of the colony (cf. EFSA, 2015, p. 30-31).
Reference:
EFSA AHAW Panel (EFSA Panel on Animal Health and Welfare), 2015. Scientific opinion on the survival, spread and establishment of the small hive beetle (*Aethina tumida*). EFSA Journal 2015;13(12):4328, 77 pp. doi:10.2903/j.efsa.2015.4328.

LINES 143-166: Please add additional details for the use of traps for SHB diagnosis. Indeed, practical information should be added to specify the way traps should be used for SHB detection (e.g., efficacy depending on the climatic conditions) (cf. EURL *A. tumida* surveillance guidelines, EFSA scientific opinion and EFSA scientific report).

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CHAPTER 2.2.6.: Infestation of honey bees with *Tropilaelaps* spp.

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINES 36-55: Information concerning the *Tropilaelaps* life cycle should be updated regarding new publications available on the topic and the below new references need to be added. Indeed, data have been published on the interaction between *Tropilaelaps* and viruses.
Refereneces:
Khongphinitbunjong, K., De Guzman, L.I., Tarver, M.R., Rinderer, T.E., and Chantawannakul, P. (2015). Interactions of *Tropilaelaps mercedesae*, honey bee viruses and immune response in *Apis mellifera*. Journal of Apicultural Research 54, 40-47.
Khongphinitbunjong, K., Neumann, P., Chantawannakul, P., and Williams, G.R. (2016). The ectoparasitic mite *Tropilaelaps mercedesae* reduces western honey bee, *Apis mellifera*, longevity and emergence weight, and promotes *Deformed wing virus* infections. Journal of Invertebrate Pathology 137, 38-42.
Khongphinitbunjong, K., Guzman, L.I., Burgett, M.D., Rinderer, T.E., and Chantawannakul, P. (2012). Behavioral responses underpinning resistance and susceptibility of honeybees to *Tropilaelaps mercedesae*. Apidologie 43, 590-599.

LINE 194: Please delete the repeated word "of" (editorial).

LINES 274-287: We query whether section 3 is relevant in light of the other disease-specific chapters of the Manual, where treatments are not described. Indeed, there is no section "3. Treatment" in other chapters. For reasons of consistency, it should be deleted.

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CHAPTER 2.2.7.: Varroosis of honey bees (infestation of honey bees with *Varroa* spp.)

General comments

The EU cannot support this revised chapter as proposed, as a more thorough revision is needed.
Indeed, the chapter should be globally restructured in order to integrate the combined pathogenic effects of *Varroa* and viruses of bees.
Moreover, throughout the text, a distinction should also be made between the varroosis diagnosis (clinical affection of the colony by *Varroa* and the associated viruses) and the infestation with the mites in absence of clinical signs.
Furthermore, there is also a need to improve the description of the agent for morphological identification, and figures and photographs should be added to improve the quality of the method descriptions.
Finally, there is an apparent inconsistency between the title of the chapter, which includes all *Varroa* species (i.e., "*Varroa* spp.") and the text, which is about *Varroa destructor* only.
Additional specific comments are given below.

Specific comments

LINE 6: The EU suggests removing the text between brackets. Indeed, the shift of hosts of both species (*V. destructor* and *V. jacobsoni*) has evolved and is detailed in the introduction (see also EU comment on LINES 27-29 below).

LINE 11: Please delete the words "especially late in the season, when clinical signs of infestation can first be recognised" and replace by the words "while clinical signs of infestation can usually be recognised when the brood is reduced". Indeed, one can usually observe clinical signs of infestation at the end of winter when the brood is reduced (see also EU comment on LINE 59 below). However, it is still better to link it to the amount of brood than to the season, the designation of the season is rather subjective. Depending on when one decides the season begins, it might be prior to the season or at the beginning of the season or late in the season or otherwise.

LINES 13-14: Please delete "(Note: *Varroa jacobsoni* is a separate species, however and is primarily a parasite of *Apis cerana* [Asian honey bees].)" (see also EU comment on LINE 6 above and on LINES 27-29 below).

LINE 16: Please delete the words "so that diagnosis entails the examination of the hive debris". Indeed, diagnosis can be performed not only on hive debris.

LINES 20-21: Please delete the words "However, this method is less accurate due to the unequal distribution of mites and the usually small sample sizes". Indeed, studies have shown that this method was accurate enough if a sample of 300 bees is taken.

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LINES 27-29: Please replace the sentence ("Until 2000 ~~recently~~ *Varroa* mites that affect *Apis mellifera* world-wide were assumed to be *V. jacobsoni*, however it has now been shown that these mites are *V. destructor.* ~~(~~Figure 1~~)~~ is a general representation of heavy infestation of *Varroa* mites on any female *Apis* species.") by the following:
"*V. destructor* and *V. jacobsoni* are the most common and widespread species of the genus *Varroa*. Both species are morphologically very similar. *V. destructor* exists on *A. cerana* throughout mainland Asia, and only two haplotypes (Korea and Japan) have successfully shifted host to colonize *A. mellifera* around the world. In contrast, *V. jacobsoni* exists on *A. cerana* in the Malaysia–Indonesia region, New Guinea and Solomon Islands and until recently was unable to parasitize *A. mellifera*. In 2008, *V. jacobsoni* was found for the first time reproducing in high numbers on both *A. mellifera* drone and worker brood in Papua New Guinea (Roberts *et al*. 2015)."
Indeed, the host shifts of both species (*V. destructor* and *V. jacobsoni*) has further evolved.
Reference:
Roberts, J. M., D. L. Anderson, and W. T. Tay. (2015). Multiple host shifts by the emerging honeybee parasite, *Varroa jacobsoni*. Molecular Ecology 24:2379-2391.

LINE 38: Please delete the word "generally". Indeed, it is now known that the first egg is male and the subsequent ones are female.

Figure 2 and Figure 3: We suggest replacing the Figures by more recent documents such as the ones in Rosenkranz *et al*. 2010 (figure 4 on page 4). Indeed, it would be appropriate to include updated figures, with more precise information based on recent observations.
Reference:
Rosenkranz, P., P. Aumeier, and B. Ziegelmann (2010). Biology and control of *Varroa destructor*. J.Invertebr.Pathol. 103:S96-S119.

LINES 51-52: Please delete the words "except in some areas, such as tropical Latin America (De Jong, 1997; Ritter *et al*., 1984)". Indeed, this information has not been observed recently and is misleading regarding the harmfulness of varroosis.

LINE 59: Please delete the words "first" and "latter". Indeed, one can observe clinical signs of infestation at the end of winter when the brood is reduced. Therefore depending on when one decides to start the season, they might be called the first of the season or the last. It is thus better to remove such references to time and/or sequence and link it only to the amount of brood.

LINES 60-61: Please delete the sentence "Heavy infestations are usually reached 3–4 years after the primary invasion, but can occur within weeks if infested by bees from nearby colonies that are collapsing.". Indeed, it appears that the course of infestation has changed in the past few years. Heavy infestations are reached much earlier than 3-4 years after the primary invasion. At the same time, it is very difficult to give a threshold to reach heavy infestations, given that it is depending on local conditions of rearing.

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LINES 63-68: Text on viruses should be added, as new data are now available on the interaction between viruses and *Varroa*.
In addition, the wording regarding "clinical signs of European foulbrood" is misleading and should be modified (to "resembling those of").
Finally, the word "infected" should be replaced by "infested", as this is the correct term.
The EU thus suggests the following:
"Essentially, the brood is damaged by the parasitic mites. Bees and their offspring that have been infested during the brood phase by only one parasitic mite show various ill effects, such as a shortened life span, changes in behaviour and an increased disease susceptibility (De Jong & Goncalves, 1982). The parasitism is critical if more than one mite enters the brood cell for reproduction. Only in the lethal stage immediately before the collapse of the colonies do clinical signs, such as shrunken wings and shortened abdomen, appear (Figure 5). A consensus exists in the literature that clinical signs and overwintering colony losses are closely associated with the vectorial transmission of the *Deformed wing virus* (DWV) to pupae through parasitizing mite (deMiranda and Genersch 2010). *V. destructor* acts as a mechanical virus vector supporting replication of virulent strains prior the transmission to honeybees (Martin *et al*. 2012, Ryabov *et al*. 2014). *Acute bee paralysis virus* (ABPV), *Kashmir bee paralysis virus* (KBV), and *Israeli acute bee paralysis virus* (IAPV) belonging to the same virus-complex could be excessively virulent when transmitted by the mite to developing pupae. The lower virulence of DWV was found to be in favour of a rapid displacement of the ABPV-complex viruses with DWV during the first 2-3 years of *V. destructor* establishment in regions (Mondet *et al*. 2014). If the brood dies shortly before or after sealing, clinical signs resembling those of European foulbrood appear without the presence of the specific agent *Melissococcus pluton*. If the brood survives, the emerging bees show various behavioural changes and their life span is considerably shortened (De Jong & De Jong, 1983; Ritter, 1996)."
References:
deMiranda, J. R., and E. Genersch (2010). *Deformed wing virus*. J.Invertebr.Pathol. 103 Suppl 1:S48-S61.
Martin, S. J., A. C. Highfield, L. Brettell, E. M. Villalobos, G. E. Budge, M. Powell, S. Nikaido, and D. C. Schroeder (2012). Global honey bee viral landscape altered by a parasitic mite. Science 336:1304-1306.
Mondet, F., J. R. de Miranda, A. Kretzschmar, Y. Le Conte, and A. R. Mercer (2014). On the front line: quantitative virus dynamics in honeybee (*Apis mellifera* L.) colonies along a new expansion front of the parasite *Varroa destructor*. PLoS Pathogens 10:e1004323.
Ryabov, E. V., G. R. Wood, J. M. Fannon, J. D. Moore, J. C. Bull, D. Chandler, A. Mead, N. Burroughs, and D. J. Evans (2014). A Virulent Strain of *Deformed Wing Virus* (DWV) of Honeybees (*Apis mellifera*) Prevails after *Varroa destructor*-Mediated, or In Vitro, Transmission. PLoS Pathogens 10:e1004230.

LINE 74: It is suggested to replace the word "attacked" by "infested", as this is the correct term.

LINES 78-79: The morphological description of the agent should be improved for differential diagnosis with other parasites or commensal agents of

honeybee hives, as new data are available. The EU will be happy to propose specific text at a later stage.

LINE 81: Please replace the words "the diagnosis of varroosis" by "to dectect the presence of Varroa" (see explanations above on the difference between *Varroa* mites carriage and varroosis).

LINE 82: Please replace the sentence "An insert covered with screen mesh is placed on the floor of the hive." by the following:
"Various devices can be used to monitor the mite falls: screened bottom boards or inserts covered with mesh.".
Indeed, the use of screened bottom boards has been widely advertised in order to monitor *Varroa* mite falls. Therefore they are available and used in many countries.

LINE 85: Please add the following before the start of the paragraph:
"This device can be used to assess

- The natural fall of *Varroa* mites;
- The fall of *Varroa* mites induced by a treatment applied to the colony."

LINES 85-90: Please replace the sentences ("The debris produced within a few days in the late season usually contains little other than visible mites (Fries *et al*., 1994; Ritter, 1996). The debris collected in winter, however, must be examined in the laboratory. An insert is placed in the hive as before, but an effective medication is used to cause the mites to fall off the bees, so that after a given time, a number of mites may be observed on the floor insert.") by the following:
"In case of monitoring natural fall, the debris produced within a few days in the late season or the debris collected in winter can be examined. When a treatment against *Varroa* is applied to the colony, a number of mites may be observed on the floor insert. Depending on the situation, mites can be directly counted on the insert or by using the flotation procedure."
Rationale: this method of screened mesh floors has been widely used in both cases (natural fall and using a treatment against *Varroa*).

LINE 102: Section 1.2.1. should be revised in accordance with new data indicating the different stages of *Varroa* present in honeybee cells.

LINE 103: Please replace the word "knife" by "sharp tool", as any sharp tool can be used to uncap honeybee brood cells.

LINE 111: The word "infected" should be replaced by "infested", as this is the correct term (this should be done throughout the text).

LINE 115: Section 1.3. should be improved in the light of new data such as Lee *et al*. 2010.
Reference:
Lee, K. V., R. D. Moon, E. C. Burkness, W. D. Hutchison, and M. Spivak (2010). Practical sampling plans for *Varroa destructor* (*Acari*: *Varroidae*) in *Apis mellifera* (*Hymenoptera*: *Apidae*) colonies and apiaries. J.Econ.Entomol. 103:1039-1050.

LINE 121: It is unclear whether this is the same method as the one described in LINES 19-20 ("Bees are washed in petroleum spirit, alcohol or detergent solution.").

LINE 130: Please replace the word "diagnostics" by "diagnosis". Indeed, "diagnostics" is an adjective and not a noun.

Figure 6: It is suggested to add *Tropilaelaps* mites in the figure, as like *Varroa*, *Tropilaelaps* is an ectoparasitic mite of honeybees. It is visible to the naked eye and thus can be confused with *Varroa*.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.3.8.: Duck virus hepatitis

General comments

The EU can support this revised chapter.

Specific comments

None.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.3.13.: Marek's disease

General comments

The EU can support this revised chapter.

Specific comments

None.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.4.4.: Bovine genital campylobacteriosis

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 6: After "subsp."the"'v" of "venerealis" should be lower case.

LINE 19: "flying seagull silhouette shapes" would better describes the appearance. Furthermore, please add "using selective media" after "cultured". Finally, please replace the word "for" by "after".

LINES 28- 30: Please delete the word "much", and replace "completed" by "conducted" in LINE 30.

LINE 31: Please replace the word "testing" by "evaluating".

LINE 71: Please replace the first "and" by "whereas".

LINE 78: MALDI-TOF is usually written in capital letters.

LINE 81: Please remove the first "the".

LINE 82: Please replace "reliable" by "reliably".

LINE 89: Please insert "Another PCR assay is described for *C. fetus venerealis* identification with a sensitivity and specificity of 98.7% and 99.8% respectively (McGoldrick *et al*. 2013)".

LINE 100: MALDI-TOF is usually written in capital letters.

LINE 105: Please delete the words "the fluids of inoculated and".

LINES 122 and 130: Please replace "sown" by "inoculated", and add "into" before "transport".

LINE 126: Please delete the words "with a tissue paper", as other types of wipes may be more suitable.

LINE 128: Please replace the word "suctioning" by "suction".

LINE 131: Please add a hyphen between the first two words (i.e., "Cervico-vaginal").

LINE 141: Please add the words "collected aseptically and" before "inoculated"; delete "directly"; and replace "in" by "into".

LINE 147: Please add the following at the end of the sentence:

"and shipped to reach the laboratory as soon as possible, preferably on the same day as sampling".

LINES 303-313: The EU queries whether this section is necessary. Indeed, this phenotype is not listed in Table 2 on differential characteristics.

LINE 386: We suggest inserting the following sentence:
"A recent PCR to identify *C. fetus venerealis* has been published with a sensitivity and specificity of 98.7% and 99.8% respectively (McGoldrick *et al.* 2013).".

LINE 470: Please replace the word "will" by "should".

LINE 471: Please replace the word "keep" by "maintain".

LINE 473: Please replace the word "are" by "should be".

LINE 475: Please replace the word "will be" by "is".

LINE 578: Please insert reference to McGoldrick CF, CFV PCR.
Reference:
McGoldrick A, Chanter J, Gale S, Parr J, Toszeghy M, and Line K (2013). Real Time PCR to detect and differentiate *Campylobacter fetus* subspecies *fetus* and *Campylobacter fetus* subspecies *venerealis*. J. Micro. Meth., 94, 199-204.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.4.12.: Infectious bovine rhinotracheitis/infectious pustular vulvovaginitis

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINES 9-10: Please amend the sentence as follows:
"Several other European countries have control and eradication programmes ongoing, for example Belgium, the Czech Republic, Republic of Ireland, Luxemburg, Scotland and northern regions of Italy."

LINE 23: Please replace the word "Therefore" by the words "Due to this latency phenomenon".

LINE 39: Please add the following sentence at the end of the paragraph:
"gE ELISAs are able to differenciate infected animals from animals vaccinated with a marker vaccine (DIVA strategy).".

LINES 79-80: The EU queries the reference for this assertion. What is the average age limit for the persistence of colostral antibodies, if 9 months is occasional, would it be 6 months?

LINES 87-90: Please delete the sentences on control strategy (starting from "Ideally, […]"), as this is outside the scope of the Manual and should be dealt ith b the Code.

LINE 364: Please insert the words "associated with an amplification curve" before "is regarded as positive".

LINE 418: We suggest defining HTS techniques, as they are relatively new and not everybody will be familiar with them.

LINE 456: Please insert ", although methods using milk concentration protocols have proved to be able to increase the sensitivity of gE ELISA on bulk milk (Schroeder *et al*., 2012)" after "(Wellenberg et al., 1998b)".
Reference:
Schroeder C, Horner S, Burger N, Engemann C, Bange U, Knoop EV, and Gabert J. (2012). Improving the sensitivity of the IBR-gE ELISA for testing IBR marker vaccinated cows from bulk milk. Berl Munch Tierarztl Wochenschr 125(7-8):290-296.

LINE 468: Please amend the sentence as follows:
"by testing individual blood or milk samples from all animals which contributed to the ~~included in a~~ positive bulk milk sample.

LINES 484-486: The EU queries a reference for these assertions.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

LUNES 574-576 : We query whether this procedure (thawing and heating) is recommended only in case of suspicion of non-specific results, or as a systematic procedure. Furthermore, it I unclear which ELISA tests are referred to by "With some ELISAs" - commercial kits, or in-house tests?

LINE 586: It is not clear what is meant by meaning "FCS". This should be explained.

Line 614 : Please amend the sentence as follows:
"Samples, tested with a gE ELISA test, were collected within 4 weeks after vaccination with a marker vaccine (vaccination phenomenon).

Line 660-662: This assertion is problematic. Marker vaccines and DIVA tests also have disadvantages: higher cost, decreased test sensitivity especially in milk, increased number of non-specific reactions. The type of vaccines to be recommended depends on the control and the test strategy, and the disease situation (free and infected zones within the same country), so there cannot be a "one size fits all" recommendation on vaccine type in the Manual.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.4.14.: Lumpy skin disease

General comments

The EU cannot support this revised chapter as proposed. Specific comments are given below, one of which is important and should be taken into account before adoption.

Specific comments

LINE 16: The EU suggests inserting the words "South-East" before "Europe", as this more accurately describes the current limited occurrence in Europe.

LINE 66: Please insert the words "South-East" before "Europe" (same rationale as comment above).

LINE 336: Please replace the word "antibody" by "antibodies".

LINES 605-611: The EU queries the reasons for deleting this paragraph (LINES 605-610).
Indeed, that exact wording has been included in the LSD Manual chapter since 1996, and was not amended in 2000, nor in 2010 or 2016. In fact, a similar wording with essentially the same assertion was included in the LSD Manual chapter as early as 1992 ("*Duration of immunity: The modified live virus vaccine produces antibodies which have been shown to persist for three years. The Kenya sheep and goat pox strain produces antibodies persisting for at least two years. Immunity is likely lifelong*.").
The scientific reference for these statements seems to be Capstick and Coakley 1961, which was already referenced in the 1992 version of this chapter (CAPSTICK P.B. & COAKLEY W. (1961). *Protection of cattle against lumpy skin disease. Trials with a vaccine against Neethling type infection*. Res. Vet. Sci., 2, 362–368).
Another reference which was added in the 1996 version of the chapter (Carn 1993) further supports these statements (CARN V.M. (1993). *Control of capripoxvirus infections*. Vaccine, 11, 1275-1279.). Indeed, the following is extracted from the abstract: "*Recently developed live attenuated vaccines provide good, virtually lifelong, protection, which is dependent on stimulating cell-mediated immunity*."
Furthermore, we do not agree with the new sentence proposed in LINE 611, as this could be misunderstood as saying that the vaccines are essentially ineffective.
As the changes proposed may have serious consequences for vaccination campaigns, the OIE should provide the scientific rationale and evidence substantiating this significant change before it is introduced in the Manual.
The EU therefore cannot support the adoption of this revised chapter as proposed.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.5.1.: African horse sickness (Infection with African horse sickness virus)

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINES 144-145: Please amend the sentence as follows:
"Virus neutralisation (VN) for serotype identification, type-specific RT-PCR or sequencing ~~with sequencing~~ should be performed […]".

LINES 207-209: Please add the reference of this method or primers sequences.

LINES 212-213: Please replace "from conserved sequences of viral segments 5 or 7 (Agüero *et al*. 2008; Fernández-Pinero *et al*., 2009; Rodriguez-Sanchez *et al*., 2008)" by "from conserved sequences of viral segments 3, 5 or 7 (Agüero *et al*. 2008; Fernández-Pinero *et al*., 2009; Rodriguez-Sanchez *et al*., 2008, Bachanek-Bankowska *et al*.,2014)".

LINE 229: Please replace "trialwith" by "trial with" (editorial).

LINES 439-440: The PCR assay from Guthrie has never been defined as "Gold standard". The title "c) Performance characteristics of AHSV gold standard real-time RT-PCR test method" should be modified accordingly.

LINES 642-644: The wording is a bit unclear. We suggest replacing it by the following (taken from the SN test for Border Disease Virus):
"i) From a starting dilution of 1/5, serial twofold dilutions of the test sera are made in a cell-culture grade flat-bottomed 96-well microtitre plate, using cell culture medium as diluent. For each sample two wells are used at each dilution. Control positive and negative sera should also be included in each batch of tests. An equal volume (e.g. 25 µl) of a stock of AHSV containing 100 TCID50 (50% tissue culture infective dose) is added to each well."

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.5.9.: Equine rhinopneumonitis (infection with Equid herpesvirus-1 and -4)

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 537: For consistency with other disease specific chapters, please delete "/registration/licensing".

LINE 539: Please replace the word "registration" by the words "marketing authorisation", as this is the correct term.

LINE 549 and 557: As above, please use the term "marketing authorisation". The terms "registration" or "licensing" have a different meaning in EU legislation and may lead to confusion.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.7.1.: Border disease

General comments

The EU can in general support this revised chapter and has one specific comment.

Specific comments

LINE 146: Please replace the word "perhaps" by "possibly", which seems to be the more appropriate term in this context.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.7.2.: Caprine arthritis/encephalitis & Maedi-visna

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 7: Please delete the word "to" after "known" (editorial).

LINE 22: Please replace "confirming" by "confirm" (language).

LINE 75: Please replace "is" by "was" (language).

LINE 168: Please add the reference "De Regge & Cay, 2013".
Reference:
De Regge N, and Cay B (2013). Development, validation and evaluation of added diagnostic value of a q(RT)-PCR for the detection of genotype A strains of small ruminant lentiviruses. J Virol Methods.194(1-2):250-7.

LINES 327-333 and 350-352: Please consider removing these example, as validation criteria are test specific and the examples could thus cause confusion.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.7.13.: Sheep pox and goat pox

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 419: Please replace the word "proved" by "shown" (style).

LINE 423: Further to the proposed deletion of ", and will probably provide lifelong protection against lethal challenge", please delete the word "Similarly", as "over a year" and "at least 30 months" are hardly similar.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.8.9.: Teschovirus encephalomyelitis

General comments

The EU can support this revised chapter.

Specific comments

None.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.9.3.: Campylobacteriosis

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 14: Please add the word "normally" before "cause disease", as evidence is increasing of adverse consequences to broiler health of Campylobacter infection.

LINE 20: Please add ",or the use of PCR" after "caecal contents".

LINES 24-25: Please amend the sentence as follows:
"Preliminary confirmation of isolates can be made by examining the morphology and motility using a light microscope".

LINES 28-30: Please include polymerase chain reaction (PCR) as follows:
"Biochemical and molecular tests, including PCR (polymerase chain reaction) and MALDI-TOF (matrix assisted laser desorption ionisation–time of flight) mass spectrometry can be used to identify *Campylobacter* strains at species level."

LINES 29, 78 (Table 1) and 83: MALDI-TOF is usually written in capital letters.

LINE 31: Please insert the word "also" as follows:
"PCR assays can also be used for the direct detection and identification of *C. jejuni* and *C. coli*.".

LINE 40: Please move "with enteritis" to **LINE 39** after "young livestock".

LINE 50: Please insert "between 50 and" before "80%".

LINE 52: Please add "but" after "poultry meat"; replace "has" by "have"; and add ", e.g. via environmental contamination" after "transmission route".

LINE 62: Please check the number of species, currently listed as 34.

LINE 63: Please replace "in the upcoming years" with "over time" (language). Furthermore, lease replace "prokaroytic" by "prokaryotic" (typographical).

LINE 78: Please insert the word "for" in the title as follows:
"Test methods available for the diagnosis of *Campylobacter jejuni* and *C. coli* and their purpose".

LINE 78 (Table 1): Please list all the detection methods before the colony confirmation and typing methods – should rapid test kits such as LFDs also be

mentioned – or is this included under antigen detection? There are other PCR genes used for detection e.g. *HIPo*, so please remove CEU and MAP.

LINES 78-79: MALDI-TOF is listed in Table 1. However, no information on this test is given in Section 1.6. Molecular detection of Campylobacter (LINES 280-290), where only the sequencing of 16SrDNA is discussed.
The EU therefore suggests including the following text in the Section 1.6.:
"MALDI-TOF MS has proven invaluable in identifying *C. coli* and C. *jejuni* on a routine basis. Standard extraction protocol for colony analysis using e.g. the HCCA matrix (Bruker Daltonics) gives excellent results. Although not all 27 currently described *Campylobacter* species can be recognized by this technique (19 species listed in the JAN 2015 release of the Bruker Daltonics database), identification of *C. coli* and *C. jejuni* as well as differentiation from the other species is highly trustable."
Reference:
Mandrell RE, Harden LA, Bates A, Miller WG, Haddon WF, and Fagerquist CK (2005). Speciation of *Campylobacter coli*, *C. jejuni*, *C. helveticus*, *C. lari*, *C. sputorum*, and *C. upsaliensis* by matrix-assisted laser desorption ionization-time of flight mass spectrometry. Appl Environ Microbiol 71(10):6292-307.

LINES 85-88: Please amend the text as follows:
"Two ISO (International Organization for Standardization) procedures for detection of *Campylobacter* exist, a Horizontal method for detection and enumeration of thermotolerant *Campylobacter* spp. (ISO 10272) with two parts: part 1 detection method and part 2: colony count technique. Both parts of the ISO are under revision and will be published 2017. The new revised ISO 10272 will include standard methods that also are optimal for isolation of *Campylobacter* from live animals. Another ISO concerns water quality, with detection and enumeration of thermotolerant *Campylobacter* species (ISO 17995, 2005 – last reviewed in 2014)."

LINE 89: Please use the term "*Campylobacter*" instead of "campylobacters" (this also applies in several other places in the chapter).

LINE 93: Please insert a comma before the word "less".

LINE 96: Please replace "via copraphagy" by "via exposure to faecal contamination"

LINE 98: Please replace "72 hours" by "a few days". Indeed, more recent data suggests infection in large modern poultry houses is likely to spread more slowly.

LINE 103: Please list "Cary-Blair" first as it has been shown to be superior to the other transport media.

LINE 105: Please add the following to the end of the line:
"and is normally based on boot-swab samples, faecal/caecal droppings or cloacal swabs".

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

LINE 114: Please list "Cary-Blair" first as it has been shown to be superior to the other transport media.

LINES 118-119: Please replace the words "in a plastic bag or Petri dish" by "in a suitable container". Indeed, there are alternative ways to transport the samples and it should be up to the laboratory themselves to decide the best way to transport the caecae.

LINE 121: Please insert the word "guarded" before "rectal swabs". Indeed, normal rectal swabs should not be used because of cross contamination from the perineal area.

LINE 125: Please delete the word "remarkably'". Indeed, more recent work has shown *Campylobacter* to be more robust than previously thought; it's just difficult to isolate from certain environmental samples without special techniques.

LINE 128: It is stated that samples should be transported and processed within at least 2 days. However, on **LINE 146** it says that samples should be processed no longer than 3 days after collection. There is a need for consistency, and we propose to change 2 days into 3 days on **LINE 128**. Furthermore, please add the words ", extreme temperatures and desiccation" to the end of **LINE 128**.

LINE 132: Please add the words "than 48 hours" after "longer".

LINE 134: Please replace "in swabs" by "on boot-swabs or rectal swabs".

LINE 135: Please insert the words "Cary-Blair or" before "Amies".

LINES 157-158: Please replace the last sentence by the following: "However, enrichment of faecal samples is usually subject to overgrowth by competing bacteria and is not routinely carried out.".

LINE 161: Please add "most commonly" before "recommended", as this depends on the purpose.

LINE 164: Please change "containing" to "based" in two places.

LINE 169: Overgrowth by ESBL-producing *Enterobacteriaceae* is not mentioned. This is very important and is reduced by adding polymyxin B, colistin or B-lactamase inhibitors to the media.

LINE 176: Please change "containing" to "based".

LINE 194: Please delete the additional "the".

LINES 235 and 237 "Microaerobic atmosphere" should be changed into "aerobic atmosphere". Indeed, according to the new revised ISO 10272 method that will be published in 2017, "microaerobic atmosphere" will be changed into "aerobic atmosphere".

LINES 238-240: Section 1.4.5. should be deleted. Indeed, there are *Campylobacter* species that sometimes grow aerobically. In the revised ISO 10272 method that will be published in 2017 this part has been removed.

LINE 256: Please insert a "*" in Table 2 after the "+", indicating hydrolysis of hippurate of *C. jejuni* and include the information "Not all strains".

LINES 258 and 259: As a new Table 1 was added (see LINE 79), this table should become "Table 2" (NB: this comment may be relevant also for the other chapters where a new Table 1 was added).

LINES 250 and 260: "Table 2" should become "Table 3" (see also comments above).

LINE 259 (Table 1): Aerobic growth at 41,5°C should be deleted as there are *Campylobacter* isolates that sometimes grow aerobically. Thus, please change "Microaerobic growth at 25°C" by "Aerobic growth at 25°C".

LINE 275: We suggest amending the sentence as follows:
"MALDI-TOF can be used to rapidly and efficiently identify *Camplyobacter* isolates at the genus and species level".

LINES 276-279: The way Section 1.6 "Molecular detection of *Campylobacter*" is drafted, it also includes identification of *Campylobacter*. We suggest to move some of the text into the last part of section 1.5.2. and to include information about the two PCR methods (Mayer 2010 and Wang *et al*. 2002) that will be included as appendix to ISO 10272, as follows:
"A variety of DNA probes and polymerase chain reaction (PCR)-based identification assays has been described for the identification of *Campylobacter* species (On, 1996; Vandamme, 2000). On & Jordan (2003) evaluated the specificity of 11 PCR-based assays for *C. jejuni* and *C. coli* identification. A fast method to differentiate *C. jejuni* and *C. coli* strains is a duplex real-time PCR, targeting gene *mapA* for *C. jejuni* identification and gene *CeuE* for *C. coli* identification (Best *et al*., 2003). Another real-time PCR method commonly used to identify and differentiate between *C. jejuni, coli* and *lari* is described by Mayer (2010). A gel-based method that is commonly used differentiates between *C. jejuni, C. coli, C. lari and C. upsaliensis (Wang et al., 2002). Campylobacter* isolates can also be molecular identified at species level with 16S rRNA sequencing (Gorkiewicz *et al.,* 2003)."
However, the following line in section 1.6. should be kept for detection:
"Inclusion of positive and negative reference strains is required for all molecular *Campylobacter* detection methods.".

LINE 286: Please remove the word "molecular".

LINE 290: Please add the following after "strains":
"and process controls to detect inhibition of the PCR reaction by the sample matrix are required".

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

LINE 296: Please add to the end of the sentence: ", although they are useful for sero-surveillance of human populations".

LINE 298: A commercial live vector vaccine for prevention of Campylobacter in chicken has been available in recent years but may now no longer be available commercially.

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.9.9.: Toxoplasmosis

General comments

The EU can in general support this revised chapter and has a few specific comments.

Specific comments

LINE 261: A reference should be added at the end of the sentence ("Recently, *H. hammondi*-specific primers have been published").

LINE 136 (Table 1): It would be good to write "DAT/MAT" (instead of "MAT") in the table, as the differences between these two tests are really minor (whether the mercaptoethanol is added to the serum or the antigen suspension before mixing them with each other; and how the titres are expressed).

LINE 263: Sections 1.4.1. and 1.4.2. do not concern the "PCR method", but rather the isolation and concentration of oocysts, and the extraction of DNA. Thus, the sentence "A PCR method is detailed below" is not correct. Furthermore, no information is given as to where this method comes from. We thus recommend adding a reference.

LINE 330: We recommend mentioning why it is important to add mercaptoethanol (i.e., to avoid false positive results due to non-specific IgM). This information could be given before the sentence "Kits are commercially available.".

EU COMMENTS
On the proposed changes to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals

CHAPTER 2.9.11.: Zoonoses transmissible from non-human primates

General comments

The EU can in general support this revised chapter and has one specific comment.

Specific comments

LINE 102 (Table 1): Perhaps the table should not be in 2 columns, as it is somewhat confusing to have different viruses on the same line that have nothing to do with each other.
Furthermore, please remove the comma in "Simian haemorrhagic, fever virus" (typographical).