ANNEX 4

EU COMMENTS

ON THE PROPOSED CHANGES TO THE

OIE MANUAL OF DIAGNOSTIC TESTS AND VACCINES FOR TERRESTRIAL ANIMALS

PRESENTED FOR COMMENTS IN OCTOBER 2014

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 of terms

General comments

The EU can support this revised glossary of terms.

It is noted that the definition of the term "Thermotolerant" is included twice in the glossary of the Manual (on p. vi and vii), the first one of which should be deleted.

Specific comments

None

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CHAPTER 1.1.1.: Management of veterinary laboratories

General comments

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

Specific comments

LINE 3: The EU notes that there currently already is a chapter number 1.1.1. ("*Collection, submission and storage of diagnostic specimens*") and thus presumes that this new chapter will become number 1.1.1.a. (or the current number 1.1.1. will become number 1.1.1.a), to avoid any confusion. Consequently, and in general, references to chapters will have to be updated throughout the Manual when chapter numbers are being amended.

LINES 15-21: A reference to international trade seems to be missing, as national laboratories also have a key role to play for that area.

LINE 100: Please add "and international standards" after "relevant national regulations", as e.g. OIE standards in this area should also be complied with.

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CHAPTER 1.1.6.: Principles of veterinary vaccine production and control

General comments

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

Specific comments

LINES 50-63 (Table 1): The EU suggests including a definition of vaccines in the glossary of the Manual.

LINE 194: As the term "Thermotolerant" is defined in the glossary of the Manual, reference to that definition should be included here (e.g. by adding a parenthesis with the words "see glossary of terms"). In addition, for reasons of consistency, the word "thermo-tolerant" in line 194 should be spelled "thermotolerant", as in the glossary.

LINES 208, 232, 342: It is noted that references to websites containing guidelines of other international organisations (e.g. VICH) are included in the text of the chapter. Whilst recognising that such references are useful for practical reasons, the EU recommends moving the references to specific URLs to the references section of the chapter, as these URLs are beyond OIE's influence and may change in the future.

LINES 216-218: As currently worded this sentence indicates that overdose and repeated dose safety studies are required for all products. However, it is correctly recognised in lines 205-210 that VICH GL 44 only requires overdose testing for live vaccines. Furthermore repeated single dose testing is only required for vaccines that are given more than once in the lifetime of the animal. The following alternative wording for this sentence is therefore suggested:
"Safety studies during development and licensing ~~for all products~~ should include the safety of a single dose for all products, as well as the safety of an overdose in the case of live vaccines and of repeated single doses for vaccines that require more than one dose during the lifetime of the animal."

LINE 335: It seems unclear why this would be the case if there is a significant effect on the human environment, as surely the animal environment and wider environmental issues are equally of concern. It is therefore sugged to delete the word "human".

LINES 402 and 405: Efficacy/immunogenicity tests are not usually carried out using the master seed but using the highest passage level from the master seed that is permitted in the Outline of Production (i.e. potentially the most attenuated passage, see line 267). It is therefore suggested to delete the words "master seed".

LINE 481: In order to avoid being overly prescriptive, it is recommended to delete reference to "court order or decree", as the procedure would depend on

the legal system of the country concerned, and thus these instruments may not be needed.

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CHAPTER 1.1.10.: International standards for vaccine banks

General comments

The EU can in general support this revised chapter, however has some important comments that would need to be addressed before adoption of this modified chapter.
It is noted that the introduction section focuses mainly on explaining vaccination strategies and criteria for their application, which is not the scope of this chapter. Indeed, these vaccination strategies and criteria are part of disease control measures and management, which would be better placed in a horizontal chapter in section 4 "disease prevention and control" of the Terrestrial Code than in the Manual. It is therefore suggested to rewrite the introduction, focusing on vaccine banks, and move the section on types of vaccination to a specific new first section of the chapter, which would merely briefly describe the different vaccination strategies, without elaborating on criteria.
Some further specific comments are included below.

Specific comments

LINES 75-78: The EU questions the need to include "service contracts" as a third type of bank. Indeed, at least the first type (antigen bank) will usually be based on a contract with the vaccine manufacturer, as there is a need to formulate the antigen into a ready-to-use product before deployment. This is exemplified by the use of "and/or" before "(iii) be based on service contracts", and the paragraph in lines 101-112. Service contracts are a specific way of managing a bank (either antigen or vaccine, or both), but should not be regarded as a third "type" of bank besides antigen and vaccine.

LINES 87-89: In line with the comment above, this sentence seems misplaced, and could be merged with the paragraph on antigens (lines 79-84). Indeed, it is unclear what the "sophisticated mechanism" consists of, e.g. when compared with contracts for antigen banks.

LINE 90: Again, the need to refer to "service contracts" separately in this sentence seems questionable.

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CHAPTER 2.1.12.: Q fever

General comments

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

Specific comments

LINES 26 and 221: It is suggested to exclude faeces as a diagnostic sample as research indicated that *Coxiella burnetii* is not actively excreted in the faeces.

LINES 47, 229 and 511: The EU notes that IFA is noch commercially available in veterinary diagnosis.

LINE 163: Please remove the word "recently", as the reference is from 2010.

LINE 196: It is suggested to add the following sentence after the parenthesis referring to Chapter 1.1.3.:
"Precautions must be taken either for phase I or phase II C. burnetii. Indeed, if the phase II bacteria are considered avirulent, phase I bacteria may be present in a phase II preparation."

LINE 197: it is recommended to add the words "class 3" after the words "filtering face piece", and amend the parenthesis to read "(FFP3)", as this is the type of filter normally used.

LINE 201: Please add "with viable C. burnetii" after "aerosols".

LINE 203: It is suggested to add the following sentence at the end of this paragraph:
"In some countries, vaccination is practised for occupationally exposed people, such as abattoir workers, veterinarians and laboratory personnel. Phase I vaccines are effective, but vaccination is contraindicated for individuals who had seroconverted or had been exposed to C. burnetii prior to immunisation."

LINE 206 (Table 1): It should be reconsidered whether only ELISA should be the recommended method ("+++") for serological diagnosis of Q fever. While numerous publications have shown a weak sensitivity of CFT, when compared to the ELISA, on the other hand its very high specificity was also shown (Emery et al., 2012, 2014; and Banazis et al., 2010). Natale et al. (2012) reported fair agreement between CFT and ELISA in cattle but poor agreement in small ruminants. On the other hand, Emery et al. (2012) described surprising opposite results. They showed that sera which had low CFT titres were ELISA-negative. Similar results confirmed that ELISA test lost positive results, were obtained by Polish Reference Laboratory (the data has not been published yet but is in press) and also Kittelberger et al., 2009; Krt, 2003. Moreover, Bötcher et al. (2011) reported that 45% of bovine sera with phase I-negative and phase II positive were negative when the ELISA with

mixed antigen test was used. The incongruent results between CFT and ELISA could appear because phase II antigens are produced in early infection and the antibody response to phase II has been associated with IgM class immunoglobulin. The ELISA is able to detect the IgG antibodies while CFT detects both IgG and IgM antibodies. Emery et al. (2014) suggested that IgM antibodies could produce a positive CFT result and negative ELISA response. Additionally, String et al., 2013 proved that conversion from phase II to phase I antibody dominance occurs earlier in the phase-specific quantitative MONA-ELISA than in the ELISA with the mixed antigen. According to the available literature data currently it is difficult to evaluate which serological method ELISA or CFT gives more reliable results because both of them have some limitations. ELISA should be preferred for evaluation of epidemiological situation while CFT can give more reliable results at the herd level or when individual animals are tested. Conditionally the positives or doubtful CFT results should be confirmed by other serological methods ELISA or IFA. Therefore the diagnostic methods for Q fever as presented in table 1 of the draft revised OIE Manual chapter should be further discussed by experts. The scientific evidence described above shows that there are situations when CFT gives more reliable results than ELISA. Both ELISA and CFT have some limitations, and it is not logical that according to table 1 the first methods is much more suitable than the second.

LINE 212: The abbreviation BTM (bulk tank milk) is given under the table; however it is not used in the table. Thus, either BTM should be mentioned in the table (e.g. ELISA can be used on milk samples) or be deleted.

LINE 222: Please replace the word “unreliable” by “difficult to achieve”.

LINE 225: It is suggested to replace the sentence by the following: “Serological analyses should be now carried out using ELISA, ~~and~~ indirect immunofluorescence assay (IFA) or ~~rather than~~ complement fixation test (CFT)”. Indeed, several papers show that relative sensitivity is lowest for the CFT. However, literature shows that ELISA has some limitations (Emery et al., 2014). Sera which had low CFT titres were ELISA-negative (Kittelberger et al. 2009; Krt, 2003) and bovine sera with phase I negative and phase II positive are negative when the mixed phase ELISA test was used (Bötcher et al., 2011). Thus, ELISA should be preferred for evaluation of the epidemiological situation, while CFT can give more reliable results at the herd level or when individual animals are tested.

LINE 242: please delete the word by before “serologically” (grammar).

LINE 244: Please replace “a serum” by “each tested serum”.

LINE 261: It is suggested to include the following sentence after the one ending with ”[...] or parturition”:
“In the placenta, *Coxiella* is searched in several cotyledons (at least 3) because colonisation can be heterogeneous”.

LINE 295: Please replace “Cultural cells” by “Cell culture”, as this is the commonly used term.

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LINE 387: Please add the words “for example” before “IS1111 [...]” to indicate that also others can be used.

LINES 399-400: Please delete the words “recent” and “new”, as the reference is from 2012. Furthermore, please add the words “for diagnosis of abortions” after “validated”.

LINE 426: Please add the words “and informative values” at the end of this paragraph.

LINES 568-571: It is suggested to move this paragraph to the beginning of section 2.2.3. Test procedure (beginning at line 582), as it is part of the test description.

LINE 602: In line with comments above, it is recommended to add the following sentence at the beginning of this section:
“The complement fixation test (CFT) was largely employed before implementation of methods based on ELISA and IFA kits. In recent years, numerous reports showed a weak sensitivity of CFT compared with other methods (EFSA, 2010; Kittelberger et al., 2009; Rousset et al., 2007; 2009a; Horigan et al., 2011, Niemczuk et al., 2014, Emery et al., 2014).”.

LINE 824: As this paper has already been published, please replace the words “[Epub ahead of print]” by “61, 519-533”.

References:

1. Banazis, M.J., Bestall, A.S., Reid, S.A., Fenwick, S.G., 2010. A survey of Western Australian sheep, cattle and kangaroos to determine the prevalence of Coxiella burnetii. Vet. Microbiol. 143, 337-345.

2. Böttcher, J., Vossen, A., Janowetz, B., Alex, M., Gangl, A., Randt, A., Meier, N., 2011. Insights into the dynamics of endemic Coxiella burnetii infection in cattle by application of phase-specific ELISAs in an infected dairy herd. Vet. Microbiol. 151, 291-300.

3. Emery, M.P., Eileen, N., Ostlund, E.N., Ichou, M.A, Ballin, J.D., McFarling, D., McGonigle, L., 2014. Coxiella burnetii serology assays in goat abortion storm. J. Vet. Diag. Invest. 26, doi: 10.1177/1040638713517233.

4. Emery, M.P., Eileen, N., Ostlund, E.N., Ichou, M.A, Ballin, J.D., McFarling, D., McGonigle, L., 2014. Coxiella burnetii serology assays in goat abortion storm. J. Vet. Diag. Invest. 26, doi: 10.1177/1040638713517233.

5. Kittelberger R., Mars J., Wibberley G., et al., 2009: Comparison of the Q fever complement fixation test and two commercial enzyme-linked immunosorbent assays for the detection of serum antibodies against Coxiella burnetii (Q fever) in ruminants: recommendations for use of serological tests on imported animals in New Zealand. N Z Vet J 57:262-268.

6. Krt B. The influence of Coxiella burnetii phase I and phase II antigens on the serological diagnosis of Q fever in cattle. Slov Vet Res 40: 203-208.

7. Natale, A., Bucci, G., Capello, K., Barberio, A., Tavella, A., Nardelli, S., 2012. Old and new diagnostic approaches for Q fever diagnosis: Correlation among serological (CFT, ELISA) and molecular analyses. Comp. Immunol. Microbiol Infect. Dis. 35, 375-379.

8. String, R., Molz, K., Werner, P., Bothe, F., Runge, M., Ganter, M. 2013. Quantitative real-time PCR and phase specific serology are mutually supportive in Q fever diagnostics in goats. Vet. Microbiol. 167, 600-608.

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CHAPTER 2.1.19.: Vesicular stomatitis

General comments

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

Specific comments

LINE 98: The note in brackets (“Note: glycerol is toxic to virus [...]”) seems inaccurate. Indeed, glycerol is not toxic to virus but to cells – in this context used as cell culture monolayers for isolation and detection of live virus – and should therefore not be used for sample material for this purpose or alternatively should be diluted before contact with cells.

LINE 218: The EU notes that the use of mice for VSV diagnostics is deleted in other parts of this Manual chapter. Reference to tissues of mice” should thus be deleted also here.

LINE 325: The EU is of the opinion that ascites fluid should be replaced in this test, if possible. Indeed, if alternatives are available, these should be favoured for reasons of animal welfare.

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CHAPTER 2.3.9.: Fowl cholera

General comments

The EU can support this revised chapter.

Specific comments

None

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CHAPTER 2.4.1.: Bovine anaplasmosis

General comments

The EU can support this revised chapter.

Specific comments

None

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CHAPTER 2.4.8.: Bovine viral diarrhoea

General comments

The EU can support this revised chapter.

Specific comments

None

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CHAPTER 2.5.9.: Equine rhinopneumonitis

General comments

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

Specific comments

LINES 100-101: It is suggested to replace the sentence by the following: "However, for the purpose of agent identification, international trade or management (biosecurity practice) strain typing is not relevant".

Furthermore, in line with the previous EU comment in the introduction section, it is suggested to delete the sentence on strain typing in **LINES 178-183**.

LINE 119 (Table 1): The CFT is mentioned under "Agent identification" but it is in fact a serological test and should thus be moved to "Detection of immune response" below. Furthermore, the AGID test is listed in the table, however that test is not mentioned anywhere else in the text.

LINES 188-225: A nested PCR such as described is generally not considered state of the art for diagnostic labs not least as it requires handling of material post PCR (first round). While this can remain as it was in the past it seems recommendable not to describe any particular PCR as none of the PCRs resembles a prescribed test for international trade anyhow. Thus, it is suggested to delete the description of the test procedure (lines 188 to 225).

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CHAPTER 2.5.11.: Glanders

General comments

The EU cannot support this revised chapter as it currently stands. Important comments are given below.

Specific comments

LINES 65-66: The EU suggests deleting this sentence, as its meaning and rationale are not very clear. Indeed, glanders can be introduced into free areas via any equid, i.e. movement of live susceptible animals is the main way spreading the disease over long-distances. Furthermore, it is not clear what is meant by "leisure or racing" equids.

LINES 67-83: This new section on "*Diagnostic pathway to confirm a case of glanders*" is unacceptable for the EU and should be deleted without replacement. Indeed, it *de facto* represents a case definition, which as a general principle should not be included in the Manual, but solely in the Terrestrial Code. It is to be noted that the OIE Code Commission has proposed a case definition in the draft revised version of the Terrestrial Code chapter on glanders, which was circulated for member country comment with its September 2014 meeting report (see Annex XX of that report). While that case definition proposed for the Terrestrial Code is similar in structure, it differs significantly in detail from the one proposed here for the Manual. Having two contradicting case definitions for the same disease in two separate OIE standards is unacceptable, as this has created trade problems in the past (cf. example of Newcastle Disease and EU comment at the OIE General Session in May 2013).

Furthermore, this new section seems to suggest that either section i), or section ii), or section ii) would suffice to confirm a case of glanders. Thus, section iii)a) would suggests that a positive serology sequence by CFT and then either western blot or ELISA would be considered to be confirmation of a case of glanders (lines 75-80). This would be wrong and unacceptable, because neither the first nor the second test has perfect specificity. Therefore, it would be possible to get two sequential false positives by the law of averages, which would not be a confirmation of infection. It would thus be likely, especially in countries doing a lot of tests, to have a "confirmed" glanders case based on this case definition within a few years in a horse that was never infected. As regards section iiib) (lines 81-83), false positive results would equally be possible in these species. Therefore, the qualification statement under ii) on line 71 "*or is epidemiologically linked to a confirmed or suspected outbreak of glanders, or is giving cause for suspicion of previous contact with B mallei;*" should cover both sections ii) and iii), i.e. the serology may be considered as evidence of glanders if accompanied by an existing epidemiology or evidence of contact with *B. mallei*, for example in an existing outbreak or known infected area, or a horse with plausible contact with an infected animal or originating from an infected area. This is precisely the case in the proposed case definition in the draft revised Terrestrial Code chapter.

Finally, it is noted that there is no mention of the CFT in case of a mule, hinny or donkey (lines 81-83). That is inconsistent with lines 253-261, where in the title of this section CFT is indicated as a prescribed test for international trade in horses, donkeys, and mules. Furthermore, in the paragraph CFT is considered valid for horses, mules and camels, and in donkeys particular care is needed to avoid misdiagnosis.

In conclusion, the EU would strongly suggest putting this section on hold, until the case definition in the glanders chapter in the Terrestrial Code has been adopted, in order to avoid contradictions between these two OIE Standards.

LINE 263: There is a specific reference to the Dubai 7 strain as *B. mallei* antigen for CFT. In the description under section 2.1 (lines 255-261), it is noted in line 259 that the sensitivity and specificity of the test is critically dependent on the antigen used. However, the sensitivity and specificity of the Dubai 7 strain is not supported in the provided publications of Elschner et al. 2011 or Khan et al. 2011. Furthermore, the Dubai 7 antigen is not readily available for use, as it is not available commercially (most countries do not make their own antigen, but purchase commercially available antigen). Even if it was commercially available, one would need to validate why this strain would have a monopoly on the antigens used. The section should rather exclude any reference to any strain type. The EU therefore suggests deleting the specific strain type, as in the 2008 version of this procedure.

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CHAPTER 2.7.9.: Ovine epididymitis (*Brucella ovis*)

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 2.8.7.: Porcine reproductive and respiratory syndrome

General comments

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

Specific comments

LINE 14: Please add the words “for Type 2 virus” after the words “has also been confirmed”, as aerogenic spread has been demonstrated only for Type 2 virus.

LINE 23: As serum is not a tissue sensu stricto, it is suggested to delete the words “tissues such as”.

LINE 37-38: Please replace by “[...] antibodies to genotype 1 or genotype 2 viruses.” (grammar).

LINE 53: Please replace the word “becoming” by the word “also”, as these vaccines are now available.

LINE 178: 1000g seems very high for washing cells, as cells could be damaged. It is therefore suggested to replace 1000g by a lower speed, e.g. 300g, which is more common for this type of procedure.

LINE 191: It is suggested to add the word titration to the title, as follows: “Virus isolation / titration on alveolar macrophages”. Indeed, this better reflects the content of that section.

LINE 202: It is recommended to add also Glutamine 1% for better survival of the cells. Furthermore, 5% FBS seems a bit low, 10% is preferred for better results.

LINE 203: On the other hand, 10% antibiotic seems excessive, 1 or 2% would be sufficient.

LINE 206: If the purpose is to merely isolate PRRSV virus, the many dilutions described here are not necessary. Working with 1/10 and 1/100 dilutions only for PRRSV isolation has been used successfully.

LINE 262: It would be of interest to also describe the neutralisation assay in the OIE manual.

LINE 289: As above, it is recommended to add 1% Glutamine to the medium, which should be supplemented with 10% FBS instead of 5%.

LINE 290: the concentration of streptomycin should be specified, by adding “per ml” after “streptomycin”.

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LINE 426: It is suggested to add "<u>, mouse</u>" after "rabbit", as this species is also commonly used.

LINE 452: It is suggested to specify the cell concentration for the seeding. Furthermore, PAM could also be used.

LINE 454: It is suggested to also specify the virus concentration.

LINE 488: It is suggested to add the word "virus" after the word "Type 2".

LINE 489: It is suggested to replace the word "high" by the word "highly", as this is the common term used.

LINE 585: Please replace "African green monkey kidney" by "<u>MARC-145</u>", as this is the correct cell line (in line with changes in lines 543-546).

LINE 618: It is suggested to specify the inoculum concentration also here.

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CHAPTER 2.8.9.: Influenza A virus of swine

General comments

The EU can support this revised chapter.

Specific comments

None

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CHAPTER 2.9.6.: Nipah and Hendra virus diseases

General comments

The EU can support this revised chapter.

Specific comments

None

DRAFT EU COMMENTS
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CHAPTER 2.9.12.: Zoonoses transmissible from non-human primates

General comments

The EU can support this revised chapter.

Specific comments

None