

REPORT OF THE MEETING OF THE OIE BIOLOGICAL STANDARDS COMMISSION

Paris, 26–28 January 2010

The OIE Biological Standards Commission met at the OIE Headquarters from 26 to 28 January 2010. Dr Kazuaki Miyagishima, Head of the OIE Scientific and Technical Department, welcomed the Members of the Commission, Prof. Vincenzo Caporale, President, Dr Beverly Schmitt, Vice-President, Dr Mehdi El Harrak, Secretary General, and Dr Alejandro Schudel, Dr Chen Hualan, Dr Paul Townsend, members of the Commission, and as well as the expert participants: Dr Adama Diallo from FAO/IAEA¹ and Dr Peter Wright, Canada.

Dr Bernard Vallat, Director General of the OIE, joined the Group on 27 January 2010. He welcomed the members and answered several questions.

The Agenda and List of Participants are given at Appendices I and II, respectively.

1. OIE Reference Laboratories and Collaborating Centres

1.1. Review of the mandate of OIE Reference Laboratories and Collaborating Centres

Given the evolving role and activities of the OIE Reference Laboratories, the Commission agreed that the current mandate would need to be revised. In particular, OIE Laboratories could be involved more in validation of diagnostic tests. Once more than one OIE Reference Laboratory has been designated for a disease, the laboratories should form a network with each other to ensure equivalence of test results and services. Whether the OIE Reference Laboratories should charge for their services is a matter of policy that should be again analysed and decided upon formally by the OIE. The Commission expressed the view that it is preferable that the designated expert be a qualified veterinarian as Reference Laboratories have as a mandate, among other items “to function as a centre of expertise and standardisation for a designated disease(s)”. The Director General of the OIE pointed out that this would be difficult to impose as many of the experts on aquatic animal diseases were biologists rather than veterinarians and that inter-disciplinary collaboration was encouraged increasingly in the control of zoonoses. He suggested adding to the guidelines for applicants a paragraph asking that the expert provide assurance that his/her knowledge and competence will allow him/her to provide this expertise to OIE Members. Curricula vitae of suggested experts would be examined on a case-by-case basis by the Commission. The Commission stressed that only one OIE Reference Laboratory can be designated for a named disease or topic in any given OIE Member. The Commission will add this topic to its workplan for 2010.

1 FAO/IAEA: Food and Agriculture Organization of the United Nations/International Atomic Energy Agency

1.2 Review of new and pending applications for OIE Reference Laboratories and Collaborating Centres

OIE Collaborating Centre for Diagnosis and Control of Prioritised Animal Diseases and Related Veterinary Product Assessment in Asia

National Institute of Animal Health (NIAH), 3-1-5, Kannondai, Tsukuba, Ibaraki, 305-0856, and National Veterinary Assay Laboratory (NVAL), 1-15-1, Tokura, Kokubunji, Tokyo, 185-8511, JAPAN
Tel: (+81-42) 321-1441; Fax: (+81-42) 325-5122; E-mail: skenichi@affrc.go.jp;

Contact point: Dr Kenichi Sakamoto, Team Leader, Research Team of Exotic Diseases, NIAH. The Commission accepted this submission on condition that the word “prioritised” be removed from the title as it is difficult to define prioritised diseases. The Centre would thus be: *OIE Collaborating Centre for Diagnosis and Control of Animal Diseases and Related Veterinary Product Assessment in Asia*

The Commission recommended acceptance of the following two applications for OIE Reference Laboratory status:

OIE Reference Laboratory for Newcastle disease

National Veterinary Research & Quarantine Service, MIFAFF, 335 Joongang-ro, Manan-gu, Anyang, Gyeonggi 430-757, KOREA (Rep. of)

Tel: (+82-31) 467 1821; Fax: (+82-31) 467 1814; E-mail: kchoi0608@korea.kr, choiks@nvrqs.go.kr

Designated Reference Expert: Dr Kang-Seuk Choi.

OIE Reference Laboratory for West Nile fever

Istituto Zooprofilattico Sperimentale dell’Abruzzo e del Molise “G. Caporale” (IZS A&M), Campo Boario, 64100 Teramo, ITALY

Tel: (+ 39 0861) 332230, Fax: (+39 0861) 332251; E-mail: r.elli@izs.it

Designated Reference Expert: Dr Rossella Lelli.

An application had been received jointly from one laboratory in Belgium and another in France for a cross-country OIE Reference Laboratory for dourine. Although two cross-country OIE Collaborating Centres had been designated in the past, this approach would not work for Reference Laboratories as OIE Members would be confused over which laboratory to send samples, etc. The Commission recommended that the laboratories reach an agreement between them on how to organise their activities and re-submit the application as a single laboratory, which should have the most experience in the disease.

One laboratory in the USA had sent an application for an OIE Reference Laboratory for Porcine Cysticercosis. The Commission agreed that the expert, though not a veterinarian, had a great deal of experience in this disease and that the laboratory was extremely active and competent. Regarding the OIE policy on OIE Reference Laboratories in institutes with a primary vocation for public health, the Director General clarified that the OIE was willing to extend its collaboration with such institutes to effectively combat zoonoses and pointed out that there were already OIE designated Reference Laboratories in public health-oriented institutions, for example in France and the USA. The President of the Commission postponed a decision on this application to give the Commission members more time to consider the issues raised.

One laboratory in Italy had submitted applications for OIE Reference Laboratories for Bovine Anaplasmosis and Piroplasmiasis (Bovine Babesiosis). The Commission pointed out that according to the list of publications provided, the institute was first author of only three papers, only one of which was published in a peer-reviewed journal, and that as the proposed expert was not a veterinarian, she could not assure the function of a centre of expertise and standardisation for the diseases. The Commission agreed to postpone its decision on this application until its next session.

The OIE Reference Laboratories for Avian influenza and for Newcastle disease at the Istituto Zooprofilattico Sperimentale delle Venezie, Padova, Italy had submitted a request to change its title to Avian and Mammalian influenzas (and Newcastle disease). The Commission felt that the word “mammalian” was very broad and covered a large number of species, for some of which the laboratory might not have experience of diagnosing influenzas. The Commission favoured an approach whereby that the applicant be invited to submit a complete dossier for designation as an OIE Reference Laboratory for influenza in an individual named species, e.g. swine influenza, if it intends to enlarge the scope of its designation.

In light of the recent outbreak of Q fever in the Netherlands, the Commission determined that there was a need for an OIE Reference Laboratory and invites proposals for this disease from OIE Delegates. The Commission also noted that a revised chapter for the OIE *Manual of Tests and Vaccines for Terrestrial Animals (Terrestrial Manual)* would shortly be circulated to Members for comment.

The Commission also determined that there was a need for OIE Reference Laboratories for swine influenza though the disease is not an OIE-listed one; proposals OIE Reference Laboratories for this disease from OIE Delegates would be welcome.

1.3. Annual reports of Reference Laboratory/Collaborating Centre activities for 2009

Reports had been received from 152 out of 156 Reference Laboratories and from 30 out of 34 Collaborating Centres for terrestrial animal diseases or topics. The Commission expressed its ongoing appreciation of the enthusiastic support and expert advice given to OIE by the Reference Laboratories and Collaborating Centres. The full set of reports for 2009 would be supplied to Members and to all the Reference Laboratories and Collaborating Centres on a CD-ROM. The international activities relevant to the work of the OIE are summarised in the table:

Reference Laboratories		
General activities		Percentage of Laboratories carrying out these activities
1	Test(s) in use/or available for the specified disease	99%
2	Production and distribution of diagnostic reagents	88%
Specific OIE activities		
3	International harmonisation/standardisation of methods	68%
4	Preparation and supply of international reference standards	49%
5	Research and development of new procedures	87%
6	Collection, analysis and dissemination of epizootiological data	45%
7	Provision of consultant expertise	71%
8	Provision of scientific and technical training	67%
9	Provision of diagnostic testing facilities	49%
10	Organisation of international scientific meetings	32%
11	Participation in international scientific collaborative studies	72%
12	Presentations and publications	80%
13	Inscription of diagnostic kits on the OIE Register	3%
Collaborating Centres		
General activities		Percentage of Collaborating Centres carrying out these activities
1	Activities as a centre of research, expertise, standardisation and dissemination of techniques	88%
2	Proposal or development of any procedure that will facilitate harmonisation of international regulations applicable to the surveillance and control of animal diseases, food safety or animal welfare	69%
3	Placement of expert consultants at the disposal of the OIE	77%
Specific OIE activities		
4	Provision of scientific and technical training within to personnel from OIE Member Countries and Territories	77%
5	Organisation of scientific meetings on behalf of the OIE	54%
6	Coordination of scientific and technical studies in collaboration with other laboratories or organisations	77%
7	Publication and dissemination of any information that may be useful to OIE Member Countries and Territories	73%

1.4. Follow-up: Reference Laboratories that provided a poor Annual report for 2008

The Commission reviewed the responses from five OIE Reference Laboratories that had been judged to have submitted poor annual reports for 2008. Three of these five laboratories had now satisfied the Commission that they fulfilled the mandate for OIE Reference Laboratories. The Commission proposed that the Laboratory for Equine influenza in Munich, Germany, be removed from the list as it provided no evidence of activity. A letter would also be sent to the Director of the Animal Health Trust, Suffolk, United Kingdom requesting clarification of the situation regarding the expert at the OIE Reference Laboratories for equine influenza and equine rhinopneumonitis.

1.5. Changes of experts in the List of Reference Laboratories and Collaborating Centres

The OIE had been notified of the following changes of experts at OIE Reference Laboratories. The Commission recommended their acceptance:

Equine rhinopneumonitis

Dr Peter Timoney to replace Dr George Allen at the Maxwell H. Gluck Equine Research Center, Department of Veterinary Science, University of Kentucky, Lexington, Kentucky, USA

Caprine arthritis/encephalitis and Maedi-visna

Dr Stephen Valas to replace Dr Gérard Perrin at the Laboratoire d'étude et de recherches caprines, Afssa Niort, France.

1.6. Introducing the new Twinning Guide

Dr Keith Hamilton presented the updated Twinning Guide, and explained that the text had been amended to address concerns expressed at the last meeting of the Commission in September 2009. The Commission suggested as a further change, that the section title "Plurilateral twinning projects" be removed and that text be added emphasising that the existing twinning relationship between the parent laboratory and the candidate laboratory would remain the principal structure of twinning although training courses could be expanded to include other laboratories.

The Commission noted that often there was a long delay between the moment when the technical aspects of a twinning project was reviewed by the Commission and the time when that project actually started. The Commission was keen to see acceleration of the process.

It was also noted that it would be advisable to request that parent laboratories assure their support of and collaboration with the candidate laboratories following the ending of the financial support of the OIE project. This would assure that twinings become long-term projects aimed at creating good work relationships within the OIE laboratory network worldwide.

1.7. Review of new and pending applications for laboratory twinning

The Commission recalled that since launching the OIE twinning initiative in 2007, 1 twinning project was completed and 14 projects between OIE Reference Laboratories or OIE Collaborating Centres and candidate laboratories in developing or in-transitional countries had been approved and signed off, and were underway:

1. Italy and Russia	Avian influenza and Newcastle disease (project completed)
2. USA and Brazil	Avian influenza and Newcastle disease
3. Germany and Egypt	Avian influenza and Newcastle disease
4. Italy and Cuba	Avian influenza and Newcastle disease
5. UK and South Africa	Avian influenza and Newcastle disease
6. UK and Botswana	Avian influenza and Newcastle disease
7. Australia and Malaysia	Avian influenza and Newcastle disease
8. Canada and Colombia	Avian influenza and Newcastle disease
9. UK and China (PR of)	Classical swine fever and rabies
10. Italy and Eritrea	Brucellosis
11. UK and Turkey	Brucellosis
12. Italy and Cuba	Epidemiology
13. Italy and Botswana	Contagious bovine pleuropneumonia
14. UK and Morocco	Bluetongue and African horse sickness
15. Germany and Turkey	Rabies

Seven further projects had received the favourable opinion of the Biological Standards Commission and were due to commence:

1. Italy and Qatar	Avian influenza and Newcastle disease
2. France and Thailand	Brucellosis
3. UK and Sudan:	Brucellosis
4. Japan and India	Equine piroplasmosis
5. Argentina and Paraguay	Foot and mouth disease
6. South Africa and Nigeria	Rabies
7. France and Senegal	Veterinary Medicinal Products

In accordance with its decision during its September 2009 meeting, the Commission reviewed, by correspondence, six twinning project proposals. As a result of this review, the Commission formulated a favourable opinion on the technical aspects of proposals for the following five twinning projects:

1. Italy and Botswana for **Trichinellosis**;
2. Germany and Cuba for **Classical swine fever**;
3. UK and Uganda for **Enhancement of diagnostic capacity and epidemiosurveillance of veterinary diseases in Uganda**;
4. USA and China for **Epidemiology**;
5. USA and Chile for **Avian influenza**.

1.8. Role, mandates, function and operation of OIE Laboratory Networks

The future *ad hoc* Group on the Definition of OIE Networking (see Item 2.4 below), would be tasked with defining the mandate, responsibilities, role, function and operation of OIE networks. Prof. Caporale stated that he intends to participate in the meeting of this *ad hoc* Group. The Director General would be consulted on this matter and would take a decision on the creation and membership of this *ad hoc* Group.

2. Ad hoc Groups

2.1. Past *ad hoc* Group meetings: reports for adoption:

2.1.1. Report of the second Meeting of the *ad hoc* Group Vaccines in Relation to New and Emerging Technologies

The Commission adopted the Report of this *ad hoc* Group (Appendix III), including the Report of the OIE/FAO/WHO Meeting on the Assessment of Food Safety Related to the Use of Recombinant Vaccines in Food-Producing Animals (Appendix V of the report of the *ad hoc* Group meeting). The OIE/FAO/WHO Meeting had recommended that a new appendix should be added to Chapter 1.1.8 Principles of Veterinary Vaccine Production of the OIE *Terrestrial Manual* on benefit–risk assessment of veterinary vaccines, including genetically engineered (GE) vaccines, with a specific section on safety including food safety. The Commission believed that this issue could be addressed by adding text on food safety to the introduction to the existing Appendix 1.1.8.2 Risk Analysis for Veterinary Vaccines, rather than creating a new appendix.

2.1.2. Report of the third Meeting of the *ad hoc* Group on Validation of Diagnostic Assays

Dr Peter Wright briefed the Commission on the outcome of the meeting of the *ad hoc* Group. At the previous meeting in February 2009, the Group had combined the two introductory chapters on validation from the *Terrestrial Manual* into one single redrafted chapter, and undertook to develop a number of appendices that would eventually be added to this chapter. At this meeting, the Group finalised these appendices. They covered: development and optimisation of antibody detection assays; development and optimisation of antigen detection assays by immunological means; development and optimisation of nucleic acid detection tests; measurement of uncertainty;

statistical approaches to validation; equivalency; and selection and use of reference panels. As these appendices were longer than expected and their content was rather detailed, the Commission suggested, following the proposition of the *ad hoc* Group, that it would be better to include them in a stand-alone booklet, and refer to them in the chapter rather than to include them in the *Terrestrial Manual*. The content of the new appendices would also be made available on the OIE website, independently of the *Terrestrial Manual*. The Commission also noted that the booklet and website could also include an updated glossary of terms used commonly by validation experts.

The Group would shortly finalise the guidelines to support the dossier used for the OIE Procedure for validation and certification of diagnostic assays.

Prof. Caporale stressed the importance of having standard reference reagents, preferably validated by the OIE, to ensure quality of the performance of diagnostic tests.

The Commission adopted the report of this *ad hoc* Group ([Appendix IV](#)).

2.2. Planned *ad hoc* Group meetings

2.2.1. Second meeting of the *ad hoc* Group on Diseases of Camelids

The Commission reviewed and accepted the proposed Terms of Reference for a second meeting of the OIE *ad hoc* Group on Diseases of Camelids (see [Appendix V](#)) to be proposed to the Director General.

2.3. Proposed *ad hoc* Groups: prioritisation of work and draft Terms of Reference

The Commission identified three priority areas: networking; quality, biosafety and biosecurity of veterinary laboratories; and quality of vaccines and performance of diagnostic tests. It recommended convening three *ad hoc* Groups to address these issues as follows:

2.3.1. *Ad hoc* Group on the Mandate of the OIE Reference Laboratories and their Networking

This *ad hoc* Group would be tasked with defining the mandate, responsibilities, role, function and operation of OIE networks of laboratories. Prof. Caporale requested clarity on the status of OIE networks: whether the networks work with the Biological Standards Commission and decide their activities and work programme in agreement with the Commission and report to the Commission, or work independently of the Commission while regularly informing the Commission of their activities and achievements.

2.3.2. *Ad hoc* Group on the Quality of Vaccines and the Performance of Diagnostic Tests

The President of the Commission proposed that this would be an evolving *ad hoc* Group tasked initially with providing guidelines on vaccine quality in general. The Group would then revise the vaccine component (safety, potency, and purity) of each disease-specific chapter of the *Terrestrial Manual* starting with foot and mouth disease (FMD), classical swine fever (CSF), and rabies in collaboration with an epidemiologist, a specialist on test use and performance and an expert on the disease in question.

2.3.3. *Ad hoc* Group on Quality, Biosafety and Biosecurity of Veterinary Laboratories

The Commission felt that this was a crucial issue for OIE Reference Laboratories, many of which require guidance on how to implement a quality management system so that they can achieve the quality assurance standards outlined in the *Terrestrial Manual*. The OIE Quality Standard must also be coherent with ISO 17025, and the OIE should enter into dialogue with ISO to get agreement to differ on certain issues such as uncertainty of measurement. The Group would also be asked to better define biosafety and biosecurity so that their guidelines would be fit for purpose for veterinary laboratories within the framework of a quality management system. It would be asked to revise the chapter in the *Terrestrial Manual* in this way. The Commission expressed the opinion that OIE Reference Laboratories should be required to work according to the OIE Standard for laboratory quality, biosafety and biosecurity.

3 International Standardisation/Harmonisation

3.1. Vaccines

3.1.1. Review the correspondence and question regarding the anthrax vaccine

The Commission reviewed the correspondence regarding the correct anthrax vaccine dose as recommended in the OIE *Terrestrial Manual* and proposed that the author of the chapter be requested to provide a final answer.

3.2. Diagnostic tests

3.2.1. Progress on the ongoing standardisation programmes for reagents (for the harmonisation of diagnostic testing)

The Commission noted written reports on the following standardisation programmes:

- Highly pathogenic avian influenza (HPAI), project to prepare standard sera for use in the AGID² test; Coordinator: Dr P. Selleck, Australian Animal Health Laboratory (AAHL), Geelong, Victoria, Australia;
- Rabies, project to produce weak positive and negative OIE Standard Sera for the FAVN³ test for rabies; Coordinator Dr A. Fooks, VLA Weybridge, UK;
- Enzootic bovine leukosis (EBL), project to develop to develop a standard PCR⁴ protocol; Coordinator: Dr T. Vahlenkamp, Friedrich Loeffler Institute, Greifswald-Insel, Germany;
- Porcine brucellosis, project to develop International Reference Standards for use in the competitive ELISA⁵, the RBT⁶, the modified RBT and the FPA⁷; Coordinator: Dr K. Nielsen, Canadian Food Inspection Agency, Nepean, Canada;

The Commission noted that no progress had been made with the project to develop internationally validated standard sera for dourine testing and, given that there was no clear definition of what defines an isolate of *Trypanosoma equiperdum* as distinct from other strains, a recognised standard strain that is representative of currently circulating isolates. Prof. Caporale informed the Commission that he would contact Dr Filip Claes to resolve this impasse.

3.2.2. Review of the list of Prescribed and Alternative Tests, including new application

The Commission had received a dossier from the Canadian Food Inspection Agency, Ottawa Laboratory, entitled Application for Certification of a Monoclonal Antibody-Based Antigen Capture ELISA for Detection of *Campylobacter fetus* in Preputial Washings and Other Diagnostic Samples. The dossier included the validation data in support of the designation of this test as a screening test for the presence of *Campylobacter fetus* spp. in preputial washings of bulls or in other diagnostic samples. It is anticipated that the ELISA would not eliminate the need for culture but would allow for a more effective use of diagnostic resources. The dossier was forwarded to a validation expert and the OIE Designated disease expert for their opinion.

3.2.3. OIE Register of diagnostic tests. Review of the current applications and information on current contacts for future applications; rabies

Dr François Diaz informed the Commission that two kits were under review and that there was not yet final report for review and approval.

2 AGID: agar gel immunodiffusion
3 FAVN: fluorescent antibody virus neutralisation
4 PCR: polymerase chain reaction
5 ELISA: Enzyme-linked immunosorbent assay
6 RBT: Rose Bengal plate agglutination test
7 FPA: Fluorescence polarisation assay

The results of an inter-laboratory study on the rabies kit that is on the OIE Register had been reported to the OIE. The results questioned the fitness for purposes for which the kit was certified. The experts on the review panel who had assessed the application submitted to the OIE would be consulted for advice to resolve this issue. The Commission agreed that if these results were confirmed, it would discuss the possible ways to improve the OIE Procedure for the validation and certification of diagnostic assays at its next meeting in September 2010.

3.2.4. Diagnostic tests in wild animals

The President of the Commission requested that the Director General convene an OIE *ad hoc* Group to address the issue of standardisation of the performance of diagnostic tests for OIE-listed diseases in wild animals. The OIE Working Group on Wildlife Diseases, which is convened under the authority of the Scientific Commission for Animal Diseases, could address the issue and provide input that could be made available to the Biological Standards Commission. It was noted that the *ad hoc* Group on Validation of Diagnostic Tests had also provided an opinion on this issue at its last meeting (see [Appendix IV](#))

4. **Manual of Diagnostic Tests and Vaccines for Terrestrial Animals**

For this agenda item, the Commission was joined by the Consultant Editor of the *Terrestrial Manual*, Prof. Steven Edwards.

4.1. **Review of chapters proposed for adoption in May 2010 before they are sent to Members**

The Commission considered the revised drafts of those chapters that would be proposed for adoption in May 2010. These chapters would shortly be sent to Members for comment. As agreed by the World Assembly of Delegates to the OIE, revised chapters approved at the General Session would be updated on the website

4.2. **Selection of chapters for proposal in May 2011**

The Commission selected the following chapters for revision with a view to possible adoption in May 2011:

- 1.1.3. Quality management in veterinary testing laboratories
- 1.1.6. Laboratory methodologies for bacterial antimicrobial susceptibility testing
- New chapter on Epizootic haemorrhagic disease
- 2.1.8. Leishmaniosis
- 2.1.16. Trichinellosis
- 2.1.20. West Nile fever
- 2.2.1. Acarapisosis of honey bees
- 2.2.2. American foulbrood of honey bees
- 2.2.7. Varroosis of honey bees
- 2.3.1. Avian chlamydiosis
- 2.3.2. Avian infectious bronchitis
- 2.3.7. Duck virus enteritis
- 2.3.9. Fowl cholera
- 2.3.10. Fowl pox
- 2.3.14. Newcastle disease
- 2.4.1. Bovine anaplasmosis
- 2.4.9. Contagious bovine pleuropneumonia
- 2.4.11. Enzootic bovine leukosis
- 2.4.12. Haemorrhagic septicaemia
- 2.4.16. Theileriosis
- 2.4.17. Trichomonosis
- 2.4.18. Trypanosomosis (tsetse-transmitted)
- 2.5.7. Equine influenza
- 2.5.8. Equine piroplasmosis
- 2.5.9. Equine rhinopneumonitis
- 2.5.10. Equine viral arteritis
- 2.5.1. African horse sickness
- 2.7.5. Contagious agalactia

- 2.7.6. Contagious caprine pleuropneumonia
- 2.8.9. Swine vesicular disease
- 2.8.1. African swine fever
- 2.9.1. Bunyaviral diseases of animals (excluding Rift Valley fever): to add section on Crimean–Congo haemorrhagic fever
- 2.9.7. *Listeria monocytogenes*
- 2.9.8. Mange

The authors would be contacted shortly. The draft updated chapters would be sent to Members and Reviewers early in 2011 such that the comments can be considered and the chapters proposed for adoption in May 2011.

4.3. Review of author/reviewer list

The Commission reviewed the list of authors and reviewers for those chapters for identified in Item 4.2.

5. Resolutions

5.1. Follow-up of the Resolution No. 27 on Rinderpest, adopted May 2009

The Commission reviewed the document entitled “Global Rinderpest Eradication: Guidelines for Rinderpest Virus Sequestration” that had been drafted by the OIE *ad hoc* Group on Evaluation of Rinderpest Disease Status of Members, with the assistance of some invited experts. The Commission endorsed the draft guidelines with suggested amendments and agreed that the document be forwarded to the Joint FAO/OIE Committee on Rinderpest Eradication.

5.2. Review of Resolutions to be presented in May 2010

The Commission noted that the following resolutions would be proposed for adoption at the General Session in May 2010:

- A resolution urging OIE Members to agree on and adopt international guidelines on rinderpest virus sequestration in time for a global declaration of rinderpest eradication foreseen in May 2011;
- A resolution proposing the adoption of draft chapters for the *Terrestrial Manual*; and possibly
- A resolution proposing the addition of kits to the OIE Register.

6. Conferences, Workshops, Meetings

6.1. Second Global Conference for OIE Reference Laboratories and Collaborating Centres, 21–23 June 2010, OIE Headquarters

The Commission approved the programme and speakers for the Global Conference and designated members to chair each session. The Headquarters would send the draft questionnaire on the transport of infectious goods to all members of the Commission and the expert participants for comment before it is sent to OIE laboratory experts.

7. Liaison with other Commissions

There were no issues for the Biological Standards Commission from any of the other three specialist Commissions.

8. Matters of Interest for Information

8.1. VICH⁸ Conference

Dr Elisabeth Erlacher-Vindel, Deputy Head of the OIE Scientific and Technical Department, updated Commission on this conference, which would be held just after the Second Global Conference for OIE Reference Laboratories and Collaborating Centres.

8.2. Participation of the OIE in the WHO/OIE/FAO/IAEA questionnaire on Laboratory Quality Standards and External Quality Assessment Schemes (EQAS)

As the analysis of the questionnaire had not yet been received from the WHO office in Lyon, France, this item could not be addressed.

8.3. Report of the Meeting of the Expert Surveillance Panel on Equine influenza

The Commission noted the report of the Expert Panel. The recommendations concerning which virus strains to include in the vaccine would be published in the OIE *Bulletin*.

8.4 Update on OFFLU

Prof. Edwards and Dr Keith Hamilton provided an update about OFFLU. OFFLU continues to make progress and to raise its international profile. Several of the OFFLU Technical Activities have provided outputs, including global biosafety guidance, and guidance on pandemic H1N1 detection in pigs. The OFFLU applied epidemiology group is developing a strategy for influenza surveillance in animals.

Dr Peter Daniels replaces Ilaria Capua on the OFFLU Executive Committee; Drs Hualan Chen and Kristien Van Reeth join Keith Hamilton and Gwenaelle Dauphin as members of the OFFLU Executive Committee. A recent strategy meeting set the strategic direction of OFFLU. Another technical meeting for active members of the network is being planned, probably in November 2010.

9. Any Other Business

9.1. Workplan

The Commission had addressed this when it identified its priorities (see Item 2).

9.2. OIE training of laboratory personnel as a new future activity

The Commission was informed that Prof. Caporale, in his capacity as contact point for the OIE Collaborating Centre for Veterinary Training, Epidemiology, Food Safety and Animal Welfare would host a workshop to discuss the scope of this suggested new activity (training of laboratory personnel). A document would be available at the next meeting of the Commission in September 2010 so that a plan of action could be drawn up.

9.3. Dates of the next Biological Standards Commission meeting

The Members of the Commission would meet informally for one half-day on 23 June following the Second Global Conference of OIE Reference Laboratories and Collaborating Centres, to discuss outcomes of both the General Session and the Conference.

The Commission noted the dates for its next meeting: 14–16 September 2010.

.../Appendices

8 VICH: International Co-operation on Harmonisation of Technical Requirements for Registration of Veterinary Medicinal Products

MEETING OF THE OIE BIOLOGICAL STANDARDS COMMISSION

Paris, 26–28 January 2010

Agenda

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- 1.5. Changes of experts in the List of Reference Laboratories and Collaborating Centres
- 1.6. Introducing the new Twinning Guide
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3. International Standardisation/Harmonisation

- 3.1. Vaccines
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4. *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals*

- 4.1. Review of chapters proposed for adoption in May 2010 before they are sent to Members
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- 4.3. Review of author/reviewer list

5. Resolutions

- 5.1. Follow-up of the Resolution No. 27 on Rinderpest, adopted May 2009
- 5.2. Review of Resolutions to be presented in May 2010

6. Conferences, Workshops, Meetings

- 6.1. Second Global Conference for OIE Reference Laboratories and Collaborating Centres, 21–23 June 2010, OIE Headquarters

7. Liaison with other Commissions

8. Matters of Interest for Information

- 8.1. VICH Conference
- 8.2. Participation of the OIE in the WHO/OIE/FAO/IAEA questionnaire on Laboratory Quality Standards and External Quality Assessment Schemes (EQAS)
- 8.3. Report of the Meeting of the Expert Surveillance Panel on Equine influenza
- 8.4. Update on OFFLU

9. Any Other Business

- 9.1. Workplan
- 9.2. OIE training of laboratory personnel as a new future activity
- 9.3. Dates of the next Biological Standards Commission meeting

MEETING OF THE OIE BIOLOGICAL STANDARDS COMMISSION
Paris, 26–28 January 2010

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**REPORT OF THE MEETING OF THE OIE *AD HOC* GROUP ON VACCINES
IN RELATION TO NEW AND EMERGING TECHNOLOGIES
Paris, 17–19 November 2009**

A meeting of the OIE *ad hoc* Group on Vaccines in Relation to New and Emerging Technologies was held at the OIE Headquarters in Paris from 17 to 19 November 2009. The meeting was chaired by Dr David Mackay. Dr Cyril Gay accepted to act as rapporteur. The Agenda and List of Participants are given at Appendices I and II, respectively.

1. Introduction

The *ad hoc* Group on Vaccines in Relation to New and Emerging Technologies was welcomed by Dr Elisabeth Erlacher-Vindel (Deputy Head, OIE Scientific and Technical Department), on behalf of Dr Bernard Vallat, Director General of the OIE.

Dr Erlacher-Vindel provided an update on OIE activities related to the *ad hoc* Group since its last meeting in November 2008.

Dr Erlacher-Vindel also reported that the OIE had sent a letter to the IFAH¹ to invite it to send experts who could participate during the second day of this meeting of the *ad hoc* Group for the purpose of providing technical input on the work of the Group.

2. Adoption of the agenda

The *ad hoc* Group accepted the agenda as proposed by OIE.

3. Review and finalise the new introductory chapter on the application of biotechnology to the development of veterinary vaccines, drafted by Dr Andrew Potter and colleagues (Dr V. Gerdt, Dr G. Mutwiri, Dr S. Tikoo & Dr S. van Drunen Littel-van den Hurk), Vaccine and Infectious Disease Organization, Saskatoon, Canada

The chapter, which would be proposed for adoption as an introductory chapter to the OIE *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals* (the *Terrestrial Manual*) was reviewed and the *ad hoc* Group provided the following general comments:

- The chapter was considered an excellent review of the current state-of-the-art and the Group extended its thanks to Dr Potter and colleagues for their work.
- The Introduction needs to reference the first recombinant technologies used to develop vaccines for disease control and eradication, e.g. Aujeszky's disease virus and rabies virus.
- The Introduction should reference the need for risk assessments to ensure the safety of recombinant vaccines.
- The technologies described in the chapter must represent technologies that have advanced the development of vaccines designed specifically for the control and, if feasible, the eradication of livestock and poultry diseases.

¹ IFAH: International Federation for Animal Health

- Specific technologies described in the chapter should include and reference technologies described in the disease-specific chapters.
- Poultry vaccines are under-represented and appropriate references should be provided to ensure breakthrough technologies for poultry vaccines are identified.

The *ad hoc* Group reviewed and discussed the draft chapter prepared by Dr Potter and colleagues and made the decision to revise the document electronically to address the above comments. In addition, comments were provided that the Group would like the authors to address. The draft document is provided at [Appendix III](#).

The *ad hoc* Group considered that the risk assessment framework specifically for vaccine containing or consisting of genetically modified microorganisms should be revised (Appendix 1.1.8.2 of chapter 1.1.8 of the *Terrestrial Manual* on principles of veterinary vaccine production) and that the glossary should be updated.

4. Review and finalise the four disease-specific chapters identified at the February 2009 meeting by the Biological Standards Commission for priority update based on new vaccine technology (foot and mouth disease, Newcastle disease, Hendra and Nipah virus diseases and classical swine fever,) and revised by scientific teams led by Dr Cyril Gay

Dr Cyril Gay reviewed the process used to prepare the disease-specific chapters. Four scientific teams were assembled by Dr Gay. Team leaders were selected as follows:

- Nipah virus: Hana Weingartl, Canadian Food Inspection Agency, Canada, and James Roth, Iowa State University, USA
- Classical swine fever: Manuel Borca, Plum Island, and Cyril Gay, Beltsville, United States Department of Agriculture-Agricultural Research Service (USDA-ARS), United States of America (USA)
- Newcastle disease: Claudio Afonso, Southeast Poultry Research Laboratory, Athens, USDA-ARS, USA
- Foot and mouth disease: Marvin Grubman and Elizabeth Rieder, Plum Island, USDA-ARS, USA

The following objectives were given to the expert groups:

1. Revise and update the vaccine section of the selected chapters following the new template endorsed at the May 2009 General Session of the OIE (see [Appendix IV](#))
2. Prepare a new section on biotechnological vaccines (Section C.3) and i) identify advantages of new technology, and ii) provide requirements for at least one new technology, paying particular attention to the following: a) efficacy requirement for disease control; b) animal safety; c) food safety.

Dr Gay provided an update of what has been achieved to date (see items 4.1 to 4.4 below) and the OIE *ad hoc* Group reviewed the documents provided by the expert groups.

After reviewing the documents, and with input from the President of the OIE Biological Standards Commission, Prof. Vincenzo Caporale, the decision was made that the expert groups should only focus on Objective 2 above, consistent with the Terms of Reference of the *ad hoc* Group.

4.1. Foot and mouth disease

The *ad hoc* Group reviewed the document prepared by the Foot and mouth disease (FMD) scientific team. Revisions were incorporated directly into the document, including comments to further guide the FMD scientific team in the completion of this document.

The following comments were provided:

1. The document was considered excellent and the Group extended its thanks to the FMD scientific team for their work.

2. The two examples of new technologies provided by the FMD scientific team were deemed excellent choices and appropriate: i) the live human adenovirus-vectored FMD vaccine platform and ii) the reverse genetics inactivated leaderless FMD vaccine platform.
3. A brief introductory paragraph that illustrates some of the new technologies being explored for next generation vaccines for the control and eradication of FMD should be provided.
4. A few sentences should be added explaining why the two biotechnologies i) and ii) above were selected.
5. In regard to the live human adenovirus-vectored FMD vaccine:
 - i) This section should be abbreviated and follow the template format provided in Appendix IV: (Section 3.a. Vaccines available and their advantages, and Section 3.b. Special requirements for the biotechnological vaccines); thus, only special requirements that need to be considered for this technology should be included.
 - ii) Need to add a few references to the improvements made since this technology was derived, if any.
 - iii) Delete any information regarding the adjuvantation of this vaccine. If needed, include this information as a general statement in the introduction to identify that immunogenicity (such as cross-protection and duration of immunity) may be improved for certain FMDV serotypes/subtypes if the live vaccine formulation is adjuvanted.
 - iv) Regarding potency, only describe specific issues that need to be considered for this technology.
6. In regard to the inactivated leaderless FMD vaccine:
 - i) Identify in a general statement that with the exception of the master seed characterisation (or any other category of special requirement) all requirements are as for conventional vaccines as outlined in Section C.2. (of Appendix IV).
 - ii) Identify that proof of concept studies have been completed in target host animals (pigs and/or cattle). Provide references, if available.
 - iii) Provide the rationale for negative *versus* positive markers. DIVA (differentiating infected from vaccinated animals) strategies are implemented through the deletion of antigenic epitopes.

4.2. Newcastle disease

The *ad hoc* Group reviewed the document prepared by the Newcastle disease virus (NDV) scientific team. Revisions were incorporated directly into the document, including comments to further guide the NDV scientific team in the completion of this document.

The following comments were provided:

1. The document was considered an excellent review of the current state-of-the-art and the *ad hoc* Group extended its thanks to the NDV scientific team for their work.
2. A great number of references were provided that comprehensively cover all the vaccine research conducted in the past decade; however, the *ad hoc* Group requests that the references be limited and used solely for the purpose of providing information on the most relevant technologies that address specific problems associated with NDV vaccination.

3. An introductory paragraph that describes why these technologies were used and selected is needed. The technologies that are described should have passed the proof-of-concept stage and, if possible, describe what improvements relative to NDV control are addressed.
4. Identify whether any of these technologies have been licensed or whether they are promising new technologies that actually address problems relative to NDV control. Vaccinia-vectored NDV vaccines may not be a good example if it is unlikely to be licensed for poultry.
5. Trade names should not be used in the OIE *Terrestrial Manual*.
6. A few sections provide opinions on costs and safety of vaccines and should be removed.
7. Reference to the European Union- or USA-specific standards or any other region should be removed. Only standards that are specific for a new technology should be provided.
8. Describe at least one new technology that is the most promising in terms of solving problems associated with NDV control.

4.3. Hendra and Nipah virus diseases

The document was considered an excellent review of the current state-of-the-art and the *ad hoc* Group extended its thanks to the Nipah/Hendra virus scientific team for their work. The *ad hoc* Group accepted the document as is with no further changes or comments.

4.4. Classical swine fever

Dr Gay reported that the work of the Classical swine fever (CSF) scientific team was well underway but that the revised chapter was still a work in progress and was not ready in time for the meeting of the *ad hoc* Group. It is anticipated a first draft should be ready for review by the end of January 2010.

The *ad hoc* Group requests that the FMD, NDV, and CSF scientific teams finalise their chapters by March 2010.

5. Draft Terms of Reference for the meeting that will be devoted to the assessment of food safety in relation to the use of recombinant vaccines and to which experts from WHO and FAO will be invited

Dr Erlacher-Vindel provided information from members of the WHO/FAO/OIE Tripartite Annual Coordination and Executive Meeting that was held at the OIE Headquarters from 3 to 4 February 2009, on the request to address the assessment of food safety related to the use of recombinant vaccines in food-producing animals. The preliminary date for the meeting on this topic was proposed for the 18 January 2010.

The *ad hoc* Group discussed and proposed an agenda that would enable a substantive discussion on the issue of food safety related to the use of recombinant vaccines in food-producing animals.

General comments from the *ad hoc* Group:

- Regulatory requirements and specific procedures are in place to address the issue of public health in the risk assessment process in place in North America, Europe, and many OIE Members (see Chapter 1.1.8 of the *Terrestrial Manual*, Section on Release of live rDNA products [page 100]).
- To prepare for the meeting, provide a list of live recombinant vaccines that have been licensed in OIE Members for food-producing animals.

- Clarify what constitutes a vaccine containing or consisting of genetically modified microorganisms (GMM) for the purpose of food safety.
- Specific risk assessments conducted for live recombinant vaccines selected from the above list could be provided as examples of how public health concerns are addressed through regulations and guidelines. Examples should cover the different categories of recombinant vaccines, such as live recombinant vectored vaccines and gene-deleted vaccines. Specific issues to cover as examples include: shedding and transmission, tissue tropism, recovery of vaccine agent, genetic stability, removal of genes used for selection during cloning procedures if needed, and host-range specificity.
- The *ad hoc* Group proposes at least one representative from a regulatory agency and one from industry who have experience with the specific live recombinant vaccine risk assessments selected as examples.

The report of this meeting is at [Appendix V](#).

6. Meeting with IFAH, Representing the Animal Health Industry

The *ad hoc* Group met with IFAH representative, Dr Michel Bublot, Merial, Lyon, Gerland Laboratory, Virology Department, Discovery Research.

The Chair of the OIE *ad hoc* Group asked Dr Bublot to review his experiences with the registration of GMM vaccines relative to food safety. The following points were made:

1. Dr Bublot discussed the use of live recombinant vector technologies to produce vaccines for poultry and companion animals. The two main categories of existing licensed products are poxvirus vectors (e.g. fowlpox and canarypox) and Herpesvirus vectors (e.g. Herpesvirus of turkeys – HVT). A new category includes the Newcastle disease virus vectors used in Mexico and China (the People's Rep. of).
2. The fowlpoxvirus vectors are usually used in 1-day-old chicks. They induce an immune response in poultry that results in the elimination of the virus after 10–14 days; thus, this eliminates food safety concerns as poultry are slaughtered at least after vector disappearance.
3. The parental HTV virus has been widely used as a Marek's disease vaccine since the early 1970s a long record of safety—in poultry. These vaccines provide life long protection against Marek's disease. The vaccine virus does not replicate in mammals, hence accidental inoculation to people during vaccine administration has occurred with no consequence on human health.
4. An environmental risk assessment and risk/benefit analysis is requested for the licensing of all live vaccines (including vector vaccines). The food safety risk is part of this risk assessment.
5. The use of antibiotic resistance and/or marker genes for selection of viral or bacterial vector vaccine strains was discussed. Dr Bublot stated that in the USA, marker genes such as LacZ may be kept in the final construct but they must establish the stability of the insert. No "antibiotic resistance" genes are currently kept in any of the viral-vectored vaccines marketed by his company.
6. The *ad hoc* Group Chair requested key information and themes relative to food safety that industry could share with the OIE. Also, the *ad hoc* Group requested that IFAH consider sharing risk assessments that are already in the public domain; in particular, IFAH could select a few categories of products that are licensed as examples of the process used to address the food safety aspects of live recombinant vaccines. Dr Bublot agreed to communicate this request to IFAH.

The *ad hoc* Group Chair requested from Dr Bublot comments on the new introductory chapter on the application of biotechnology. Dr Bublot provided a number of comments, which were taken into account by the *ad hoc* Group.

7. Any other business

None.

.../Appendices

Appendix I

**MEETING OF THE OIE AD HOC GROUP ON VACCINES
IN RELATION TO NEW AND EMERGING TECHNOLOGIES
Paris, 17–19 November 2009**

Agenda

1. Introduction
 2. Adoption of the agenda
 3. Review and finalise the new introductory chapter on the application of biotechnology to the development of veterinary vaccines, drafted by Dr Andrew Potter and colleagues (Dr V. Gerdt, Dr G. Mutwiri, Dr S. Tikoo & Dr S. van Drunen Littel-van den Hurk), Vaccine and Infectious Disease Organization, Saskatoon, Canada
 4. Review and finalise the four disease-specific chapters identified at the February 2009 meeting by the Biological Standards Commission for priority update based on new vaccine technology (foot and mouth disease, Newcastle disease, Hendra and Nipah virus diseases and classical swine fever,) and revised by scientific teams led by Dr Cyril Gay
 5. Draft Terms of Reference for the 1-day meeting that will be devoted to the assessment of food safety in relation to the use of recombinant vaccines and to which experts from WHO and FAO will be invited
 6. Meeting with IFAH, Representing the Animal Health Industry
 7. Any other business
-

Appendix II

**MEETING OF THE OIE AD HOC GROUP ON VACCINES
IN RELATION TO NEW AND EMERGING TECHNOLOGIES**

Paris, 17–19 November 2009

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CHAPTER 1.1.7.

THE APPLICATION OF BIOTECHNOLOGY TO THE DEVELOPMENT OF VETERINARY VACCINES

INTRODUCTION

The practice of vaccination for the prevention of animal disease has been used for centuries and has proven to be a powerful tool for the alleviation of animal suffering as well as the economic well being of producers of animal products. Up until 15–20 years ago, vaccines had changed little from those originally pioneered by Jenner and Pasteur. Since that time there have been significant changes in the types of vaccines available owing to a number of factors, including compatibility with eradication programmes and international trade policies as well as cost-effectiveness of production. The first recombinant vaccines were introduced in the late 1980s to control Aujeszky's disease and rabies in wild-life (Pastoret et al., 1988) and are the forerunners of similar products that will be available in the future.

The approaches used in the development of vaccines have expanded rapidly as the result of increased knowledge of the mechanisms by which protective immunity is induced, and the explosion of genomic data on both pathogens and their hosts. The associated evolution of new technology in the field of molecular biology and immunology has furthermore had a large impact on the development of new vaccine strategies and the quality of the products that are produced. It has enabled the design of vaccines targeted for the control and eradication of specific pathogens within the framework of regional, national and international requirements. Use of recombinant technologies bring with it the need for the application of a risk–benefit assessment that takes into account the specific aspects that need to be considered, particularly with respect to safety (see Appendix 1.1.8.1 Risk analysis for biologicals for veterinary use, to Chapter 1.1.8 Principles of veterinary vaccine production of this Terrestrial Manual).

This chapter describes a range of technologies that are used to produce vaccines engineered for a specific purpose. The categorisation is aimed to assist the reader to understand the technologies employed, but it should be recognised that the categories are not mutually exclusive (i.e. reverse genetics may be used to produce a chimeric vaccine). In principle, the technologies can be used to change the target pathogen itself to alter its properties by deletion, insertion, other genetic modifications, or they can be used to modify the isolated genes of pathogens to produce specific immunogens associated with protective immunity.

A. REVERSE GENETICS

The development of a reverse genetics system for a range of different RNA viruses has revolutionised the field of virology by making it possible to introduce designed mutations, insertions and deletions into the viral genome of live viruses. It has by now been used in a range of applications that include the attenuation of viruses, the modification of host specificity and the generation of replication-deficient viruses. These strategies have also been applied to the development of new vaccine strategies and are widely used in the characterisation of the structure and function of individual viral genes.

The technology of reverse genetics involves the generation of a cloned copy of complementary DNA (cDNA) from RNA by reverse transcription *in vitro*, manipulating DNA *in vitro* followed by generating the modified live virus by transfection of permissive cells with the cloned DNA(s). The technology was first demonstrated using the bacteriophage Q-Beta, a positive-strand RNA virus (Taniguchi, 1978). Subsequently, a large number of positive-strand RNA viruses including severe acute respiratory syndrome (SARS) coronavirus, with large genomes have

been rescued, which has helped in the study of the biology of these viruses and the development of new live attenuated viral vaccines. For example, reverse genetics was used to develop an infectious clone of transmissible gastroenteritis virus (TGEV), which induced lactogenic immunity in immunised pigs (Sola *et al.*, 2003). This novel technique has also been used to develop a modified porcine respiratory and reproductive syndrome virus, which can be used as a DIVA (differentiating infected and vaccinated animals) vaccine to help differentiate between vaccinated and infected pigs (de Lima *et al.*, 2008).

Owing to the inherent characteristics of negative-strand RNA viruses, it took years of work before this technique could be developed and used for generating engineered viruses containing negative-strand RNA genomes. Reverse genetics was first developed for influenza virus, a segmented negative-strand RNA virus. Since then, this technique has been successfully used for the generation of a number of RNA viruses containing either unsegmented or segmented negative-strand genomes. For example, the use of this technique has led to the development of a vaccine for avian influenza virus in which the engineered virus contained a haemagglutinin (HA) gene from an H5N1 virus and a neuraminidase (NA) gene from a H2N3 virus, using a H1N1 backbone (Meeusen *et al.*, 2007). The resultant inactivated H5N3 virus vaccine induced complete protection in birds against highly pathogenic H5N1 challenge. A reverse genetics strategy has also been used in the development of Foot and mouth disease, Classical swine fever, and Newcastle disease vaccines (see OIE Chapters 2.1.5, 2.8.3 and 2.3.14, respectively). More recently, reverse genetics systems have been developed for segmented double-stranded RNA viruses including bluetongue virus (BTV) introducing the possibility of new vaccine development strategies for these viruses (Boyce *et al.*, 2008).

B. RECOMBINANT VECTOR TECHNOLOGY

Advances in reverse genetics, genomics, and proteomics have facilitated the identification of mechanisms of virulence, host-pathogen interactions, and protective antigens from many pathogenic microorganisms and also the development of suitable vehicles/vectors for delivery of these antigens to the host. The availability of bacterial and viral genome sequences has facilitated the rapid construction of defined deletions in the genomes of a wide variety of pathogens, which not only results in attenuation, but also creates space for the insertion of foreign genes coding for antigens from heterologous microbes. In general, live bacterial or viral vectors share several characteristics including ease and economy of production, non-integration into the host genome, stability and a reasonable capacity to insert genes coding for heterologous antigens. In addition, as with any live vaccine, the vector should be avirulent and the impact of immunity to the vector should be evaluated

a) Bacterial vectors

In general bacterial vectors are attenuated by deletion of genes required for key metabolic processes or genes associated for virulence. Although they are not used routinely in animals, rapid progress is being made in developing and evaluating different bacteria as vectors. For several years, BCG (Bacillus Calmette–Guerin) and *Salmonella* have been developed as vectors for delivering vaccine antigens to animals and the latter has been used for the generation of live vaccine strains for poultry. There are currently a number of other bacterial vectors being developed based on commensal microorganisms (*Lactococcus*, *Streptococcus*, *Lactobacillus* and *Staphylococcus*) or attenuated pathogenic organisms (*Shigella*, *Bacillus*, *Yersinia*, *Vibrio*, *Cornibacteria*, and *Bordetella*), all of which are being evaluated for their ability to induce protective immunity.

b) Viral vectors

Most viral vectors are developed using viruses that are associated with mild or no disease or using viruses that are pathogenic but attenuated by deletion of virulence genes. Replication competent virus vectors, which can produce progeny virus, as well as replication-defective virus vectors, which do not produce progeny virus, have been developed and evaluated as vaccine delivery vehicles. A number of commercial vaccines based on DNA virus vectors, including poxviruses and herpesviruses, have been successfully licensed for use in veterinary medicine (reviewed in Gerdtts *et al.*, 2006). These include vectors based on vaccinia virus, canarypox virus, fowlpox virus and turkey herpesvirus. A number of viral vectors have been developed or are in the process of being developed, improved and evaluated. These include RNA viruses such as Venezuelan equine encephalitis virus, Newcastle disease virus and feline foamy virus as well as DNA viruses such as adenoviruses, herpesviruses and pox viruses, Fowl pox and canary pox vectors have been used in a wide range of applications (MacLachlan *et al.* 2007; Swayne, 2009) whereas replication-deficient human adenovirus vectors have been used very successfully in the development of FMDV vaccines (Rodriguez & Grubman, 2009). Licensed canary pox vaccines include vaccines against equine influenza (<http://www.emea.europa.eu/vetdocs/PDFs/EPAR/proteqflu/V-073-en6.pdf>) and feline leukaemia (<http://www.emea.europa.eu/vetdocs/PDFs/EPAR/PurevaxFeLV/026600en6.pdf>). Other licensed vector vaccines include the herpesvirus of turkeys vectored with an infectious bursal disease insert (<http://www.emea.europa.eu/vetdocs/PDFs/EPAR/Vaxxitek/075502en6.pdf>).

C. GENE-DELETED VACCINES

The knowledge of specific virulence factor(s) of a pathogen and the availability of recombinant DNA technology has facilitated the creation of specific gene-deleted pathogens for use as live vaccines. The approach of creating and testing defined gene deletions ultimately aids in reducing the pathogenicity/virulence of the organism without affecting the immunogenicity. Such gene-deleted organisms can be used as vaccines as they retain the immunogenic features of the wild-type organism but cannot cause disease. However, to be effective as viable vaccine(s), these organisms should be genetically stable, easy to grow and easy to administer. So far, genes involved either in determining virulence or regulating key metabolic pathways of the organism(s) have been targeted for such deletions.

This approach has been successfully used to create several live attenuated vaccine strains of bacterial pathogens that are genetically stable, safe to use and induce better protection than killed vaccines. Gene-deleted *Salmonella enterica* serovar *typhimurium* and serovar *enteritidis* vaccines have been licensed for use in poultry (Babu *et al.*, 2004; Meesun *et al.*, 2007) and similarly, an *aroA* gene-deleted *Streptococcus equi* vaccine has been licensed for use in horses (Jakobs *et al.*, 2000; Meesun *et al.*, 2007).

This technology has also been successfully used to create live attenuated vaccine strains of viral pathogens that are genetically stable and can be used as marker vaccines to differentiate between vaccinated and infected animals. A double gene (gE and TK) deleted pseudorabies virus marker vaccine has been licensed for use in pigs (Ferrari *et al.*, 2000; Meesun *et al.*, 2007) and similarly, gE deleted a bovine herpesvirus-1 marker vaccine has been licensed for use in cattle (Meesun *et al.*, 2007; Van Oirschot *et al.*, 1996).

D. CHIMERIC VIRUSES

Chimeric viruses are defined as recombinant viruses that may contain parts of two closely related viral genomes. For example, a chimeric virus could be one that contains structural genes of one viral serotype and nonstructural genes of another serotype of the same virus. Alternatively, a chimeric virus would be one that contains part of the genome from different members belonging to the same virus family. In principle, chimeric viruses display the biological characteristics of both the parent viruses. One of the main advantages of this approach is that a single dose of chimeric virus delivers the complete repertoire of antigens closely resembling the pathogen(s), which can induce protective immune response against multiple viral pathogens belonging to or different serotypes of the same viral pathogen.

The availability of infectious full-length complementary DNA (cDNA) clones of different RNA viruses using reverse genetics technologies has led to novel vaccine development strategies. Chimeric pestiviruses have been constructed using an infectious cDNA clone containing the classical swine fever virus (CSFV) genome or the bovine viral diarrhoea virus (BVDV) genome backbones. In one instance, a chimeric pestivirus was constructed by replacing the BVDV E2 coding sequence in the infectious DNA copy of BVDV strain CP7 with the corresponding E2 coding sequence of CSFV strain Alfort 187 (Reimann *et al.*, 2004). Another chimeric virus was constructed by replacing the CSFV E2 coding sequence in the infectious DNA copy of CSFV vaccine strain C with the corresponding E2 coding sequence from BVDV (44). These chimeric viruses appeared to be attenuated in pigs, induced complete protection against CSFV challenge and helped to discriminate between vaccinated and infected pigs (Reimann *et al.*, 2004; van Gennip *et al.*, 2000).

In another application, chimeric porcine circoviruses (PCVs) have been isolated using infectious cDNA clones of porcine circovirus PCV1 in which the capsid protein from pathogenic PCV2 was used to replace the corresponding gene in the nonpathogenic PCV1 strain (PCV1-2). Likewise, the capsid gene in PCV2 has been replaced with the gene from PCV1 (PCV2-1). The chimeric PCV1-2 virus appeared to be attenuated in pigs and induced protective immunity against wildtype PCV2 challenge in pigs (Fenaux *et al.* 2004).

This platform technology has also been used to generate chimeric flaviviruses. In one example, a chimeric virus was generated by replacing the structural genes of yellow fever YF-17D virus with structural genes of West Nile virus (WNV). A single dose of this chimeric flavivirus vaccine induced both cell-mediated and humoral immune responses in horses, and provided protection against WNV challenge without causing any clinical illness (Meeusen *et al.*, 2007). The same platform technology has also been used to develop human vaccines for Japanese encephalitis virus, WNV and Dengue virus. Although chimeric flavivirus vaccines have shown satisfactory safety profiles and protective efficacies, caution should be used in evaluating chimeric viruses for the change in virulence.

E. SUBUNIT VACCINES

Subunit vaccines composed of semi-pure or purified proteins have been commercially available since the early 1980s, with subunit components produced by recombinant DNA technology available since the 1990s (Cohen, 1993;

Rhodes et al., 1994; Ulmer et al., 1993; 1995). The latter have attracted growing interest and activity since that time. Subunit vaccines do not include live recombinant vector technologies, which provide the delivery of recombinant proteins *in vivo*. The field of genomics and related areas has revolutionised the manner in which microbial antigens are identified. Since the first bacterial genome was sequenced in 1995, there has been a huge increase in the number of bacterial, viral, and parasite genomes for which genome sequences are available. Indeed, virtually all pathogens of animals are represented and those pathogens that are not can readily be obtained in less than a day. More importantly, the development of the bioinformatics resources and tools that are required to analyse these genomes has proceeded in parallel and it is now relatively easy to identify surface-exposed antigens, specific B- and T-cell epitopes, etc. There is no requirement to have the ability to grow the organism in culture: for example subunit vaccines for *Piscirickettsia salmonis*, a salmonid pathogen, have been developed even though the organism could not be readily grown (Kuzyk et al., 2001).

The production of subunit antigens can be achieved by both conventional biochemical (e.g. <http://www.emea.europa.eu/vetdocs/PDFs/EPAR/euricanherpes/V-059-en6.pdf>) or recombinant DNA technologies. The latter involves a range of prokaryotic and eukaryotic expression systems including yeast, insect cell and plants (Chichester & Yusibov, 2007) by means of a variety of integrated or transient expression strategies. Biochemical techniques remain useful in some cases where recombinant expression is not appropriate, such as antigens requiring complex assembly (e.g. fimbriae), or when post-translational modification is necessary. For example, *Campylobacter jejuni* is one bacterial species that glycosylates many surface proteins and, as such, they are best produced in *C. jejuni* rather than heterologous expression systems, although *Escherichia coli* strains have been engineered to carry out the same function (Wacker et al., 2002). An excellent example of a subunit vaccine composed of an authentic antigen that retained three dimensional structure was the original *E. coli* K99 vaccine for calf scours, which was tested three decades ago (Acres et al., 1979). This product was based on the K99 fimbrial antigen, which could readily be extracted from cells by heat treatment, thus retaining the three dimensional fimbrial structure. Another example includes a baculovirus expressed vaccine against porcine circovirus type 2 (Fachinger et al., 2008). In a number of cases the expressed subunit vaccine protein spontaneously assembles into well defined particles that may resemble virus particles. These virus-like particles (VLPs) are a sub-class of subunit vaccines (Roy & Noad, 2008) and their application in vaccine development is reviewed in section F. Vaccines containing ORF 2 protein of PCV-2 expressed in baculovirus has been commercialised (<http://www.emea.europa.eu/vetdocs/PDFs/EPAR/PorcilisPCV/V-135-en6.pdf>; <http://www.emea.europa.eu/vetdocs/PDFs/EPAR/ingelvacCircoFLEX/V-126-en6.pdf>).

Subunit vaccines could have some advantages over live attenuated and inactivated vaccines, including the ability to induce strong humoral and cell-mediated immune response. The vaccines furthermore have an excellent safety profile, and can be used in combination with other subunit vaccines. However, efficacy is dependent on the protective immunity being induced by inoculation of a single or set of defined recombinant proteins. Experience has shown this may be affected by the gene expression system used. In addition, subunits vaccines may be expensive to produce for some glycoproteins and may require the use of adjuvants to enhance immune responses.

One of the biggest advantages of subunit vaccines is that they are generally compatible with DIVA strategies as long as the antigen is not being used as a marker. In the case of bovine herpesvirus, glycoprotein gD has been successfully used in subunit vaccine formulations. Although immunisation with gD has proven to be protective at an individual animal level (Harland et al., 1992; van Drunen Littel-van den Hurk et al., 1994), it has not reduced the prevalence of the virus in the field, thus limiting its use. Subunit vaccines against a variety of other respiratory and enteric viruses, including BVDV, BRSV, PI3, rotavirus, and coronavirus have been successfully tested, although none of these is used on a commercial basis. Bacterial subunits have arguably proven more successful than their viral counterparts. This is because of the cost-effectiveness of growth of both conventional and recombinant organisms and a general requirement for a Th₂-biased immune response in many cases. Recombinant vaccines are commercially available for respiratory pathogens such as *Mannheimia haemolytica* and *Actinobacillus pleuropneumoniae* based upon the leukotoxins produced by these organisms, as well as transferrin-binding proteins. *Actinobacillus pleuropneumoniae* is an excellent example of a vaccine composed of subunits selected on cross-serotype reactivity, thus providing broad-spectrum protection against disease. Likewise a vaccine against atrophic rhinitis containing a non-toxic derivative of *Pasteurella multocida* dermonecrotic toxin produced by a genetically modified *Escherichia coli* strain together with a conventional *B. bronchiseptica* bacterin has been commercialised: (<http://www.emea.europa.eu/vetdocs/PDFs/EPAR/Porcilisartdf/V-055-en6.pdf>).

Vaccines against CSF demonstrate well the need to target recombinant technology to a particular purpose. Conventional, live attenuated CSF vaccines have a rapid onset of immunity and are effective at preventing transmission of infection (Van Oirschot, 2003) but have the disadvantage that it is not possible to differentiate infected pigs from those that have merely been vaccinated. Commercial E₂ subunit vaccines (<http://www.emea.europa.eu/vetdocs/PDFs/EPAR/Porcilispesti/041200en6.pdf>) have a slower onset of immunity and reduce but do not prevent viral shedding. However, they enable a DIVA strategy to be followed thereby facilitating a vaccination to live' strategy. Their use is therefore likely to be of particular benefit in high value breeder pigs where the vaccine can be used to limit the clinical impact of the disease whilst allowing individual infected pigs to be identified and eliminated.

F. VIRUS-LIKE PARTICLES

Virus-like particles (VLPs) are supra-molecular structures composed of one or more recombinant proteins. The particles form through self-assembly and typically range from 20 to 100 nm in size. Depending on the origin they can be icosahedral or rod-like in structure (reviewed in Jennings & Bachmann, 2008). VLPs offer the advantage of formulating the vaccine antigen in a particulate structure, thereby increasing the immunogenicity of the vaccine. VLPs can be used as either vaccine itself or as carrier for genetically fused (chimeric), incorporated or covalently linked antigens. VLPs have been extensively studied for the past 20 years, with human vaccines against hepatitis B virus (Zuckerman, 2006) and human papillomavirus (Stanley, 2008) commercially available and several vaccines for veterinary application in development. These include vaccine for bluetongue virus, rota- and parvovirus.

VLPs offer several advantages for the use as vaccine including a high safety profile, the similarity to viral and bacterial structures, the ability for large-scale production and the possibility of combining the VLPs with other adjuvants. Typically, immunisation with VLP induces rapid and strong antibody responses. Similar to viruses and bacteria multiple copies of the vaccine antigens are displayed in a highly repetitive and ordered, quasi crystalline-structure (Bachmann & Zinkernagel, 1996), which can cross-link the B cell receptor resulting in activation of the B cell and subsequent induction of T-independent IgM responses (Thyagarajan *et al.*, 2003). Furthermore, this enables interaction with the complement system resulting in increased phagocytosis. Moreover, the particulate structure of VLPs also enhances uptake by dendritic cells and subsequent cross-presentation of the antigen. It was demonstrated by Lenz *et al.* (2001) that cross-presentation of particulate antigens was more effective than presentations of soluble antigens. However, the induction of T cell responses overall is still not as effective as those induced by live vaccines. To overcome this, VLPs have been successfully combined with molecular adjuvants such as CpG ODN and single-stranded RNA. Other VLPs have been demonstrated to directly stimulate dendritic cells (DC.) For example, L1 protein-VLPs of papillomavirus have been shown to directly activate DC.

VLPs can either be used as vaccine itself or be used as carrier for recombinant antigens, either incorporated, directly, genetically fused or covalently linked. For example, the bovine rotavirus virus protein 6 (VP6) forms VLPs that are highly immunogenic and already confer protection against challenge infection (Redmond *et al.*, 1993). However, using the VP4 and VP7, other antigens can be covalently linked to the VP6 particles and used for immunisation (Redmond *et al.*, 1993). Other prominent examples include the hepatitis B surface antigen VLPs (HBsAg-VLP), human immunodeficiency virus 1, dengue virus VLPs, norovirus VLPs, and influenza A VLPs. Examples of VLPs used as carriers include the well characterised hepatitis B core antigen VLPs (HBcAg VLPs; [Blanchet & Sureau, 2006; Pumpens & Grens, 2001]) as carrier for the influenza A M2 protein (M2-HBcAg [Jegerlehner *et al.*, 2002]), or malaria B- and T-cell epitopes (Nardin *et al.*, 2004). While mostly administered systemically, some VLPs-based vaccines already have been tested for mucosal administration.

G. DNA VACCINES

Immunisation with DNA represents a relatively new vaccination strategy that is based on a simple concept. DNA vaccines can be defined as antigen-encoding bacterial plasmids that are capable of inducing specific immune responses upon inoculation into a suitable host. Immunisation is accomplished by the uptake of purified plasmid in the host cells, where it persists extrachromosomally in the nuclei. Subsequent expression of protein results in the presentation of normally processed or modified forms of the protein to the immune system. In the host, native forms of the proteins have access to presentation pathways by class I major histocompatibility (MHC) antigens in addition to class II MHC presentation, which results in a balanced immune response. The use of pure plasmid DNA offers many advantages over other vaccine delivery vehicles. One of the greatest advantages is the ability of DNA vaccines to induce both humoral and cell-mediated immune responses, which is critical for protection from many diseases. There is also evidence that DNA vaccines can induce long-term immunity, which is a further requirement for vaccine efficacy. As the vector itself does not induce immune responses, DNA vaccines can be repeatedly administered without the interference of antibodies. From a technical viewpoint, DNA vaccines are easy to engineer, produce and purify, so new DNA vaccines can be constructed and evaluated in animal models within months. DNA vaccines are very stable and therefore have a long shelf life and can be transported without a cold chain. The safety of DNA vaccines has been established in various trials in several species including humans (Bagarazzi *et al.*, 1998; Kim *et al.*, 2001).

As soon as the concept of DNA immunisation began to be explored, this technology was found to be very effective in rodents, but initially did not perform as well in larger species. However, recent progress has resulted in the development of DNA vaccines in a number of outbred target species (Carvalho *et al.*, 2009; Redding & Weiner, 2009). Currently, four veterinary DNA vaccines have been licensed, against growth hormone releasing hormone for swine in Australia, infectious haematopoietic necrosis virus for salmon in Canada, WNV for horses and melanoma for dogs in the USA (Kutzler & Weiner, 2008). To achieve higher efficacy in large animal species, optimisation at various levels has been required, including (i) vector modifications; (ii) protein engineering to modify subcellular localisation; (iii) improvements in DNA delivery routes and methods; (iv) inclusion of adjuvants, as a gene or co-administered agent, and (v) antigen targeting to antigen-presenting cells (APCs). It is likely that the often unsatisfactory efficacy of DNA vaccines in large animals was caused by inefficient transfection, as well as

'immunological blandness', of the administered plasmids. The use of a needle-free vaccine delivery device was shown to reduce the effective dose of an experimental polyvalent DNA vaccine for avian influenza, and to rapidly deliver repeated injections in poultry (Rao *et al.*, 2009).

H. ANTIGEN DELIVERY AND MOLECULAR ADJUVANTS

Adjuvants are substances that enhance immune responses when co-administered with antigens. They are a critical component of killed (recombinant and subunit) vaccines, which are often poorly immunogenic. Adjuvants can be classified into two broad categories based on their presumed mechanism of action: i) delivery systems, and ii) immunostimulatory adjuvants. Delivery systems include many conventional adjuvants and numerous particulate adjuvants and will be discussed separately below.

Despite the importance of adjuvants in vaccines, their mechanisms of action remains poorly understood. Recent advances in the understanding of innate immunity have provided important clues on the molecular mechanisms of action of immunostimulatory adjuvants. In this regard, immune cells express a variety of receptors, collectively termed pattern recognition receptors (PRR) that broadly detect conserved microbial components referred to as pathogen-associated molecular patterns (PAMPs). A number of PRR have been described including Toll-like receptors; e.g. TLR 9 recognises bacterial CpG nucleic acid motifs, natural agonist of TLR7/8, single-stranded viral RNA (oligoribonucleotides, ORN) strongly activate innate immune responses in mice, humans and are particularly potent in large animals; TLR4 agonist such as lipopolysaccharide (LPS) that is known for its powerful immunostimulatory and adjuvant properties, but unfortunately this molecule is highly toxic; nucleotide oligomerisation domain (NOD)-like receptor (NLL), retinoic acid inducible gene (RIG)-like receptors (RLL), and C-type lectin receptors (CLR), all of which detect microbial components. Engagement of these receptors by their agonists leads to a cascade of molecular and cellular events that result in activation of innate immunity, which directs antigen-specific adaptive immunity. Of these receptors, TLR agonists are most widely explored and have shown great promise as adjuvants. Interestingly, the live-attenuated yellow fever vaccine 17D (YF-17D), one of the most successful vaccines available, activates TLR2, 7, 8 and 9 (Querec *et al.*, 2006), suggesting that the success of at least some of the live vaccines may be the result of their ability to activate TLRs. This has generated a great deal of interest in TLR agonist as adjuvants.

The existing paradigm in the veterinary vaccine industry of "one adjuvant-one vaccine," is driven partly by costs associated with including more than one adjuvant in a vaccine; however, it may severely limit the efficacy of potentially safe vaccine candidates, and may explain, at least in part, why some vaccines or adjuvants have only achieved suboptimal efficacy. Evidence is slowly accumulating to indicate that multiple adjuvants may offer more than can be achieved with a single adjuvant. For example, although CpG ODNs are a good adjuvant, they can have even greater adjuvant activity if formulated or coadministered with other compounds, such as particulates, mineral salts, saponins, liposomes, cationic peptides, polysaccharides and bacterial toxins and the synthetic polymers, polyphosphazenes (Wack *et al.*, 2008).

The adjuvant effect of microparticles has been known for some time and has been previously reviewed (Mutwiri *et al.*, 2005). Particulate delivery systems are thought to promote trapping and retention of antigens in local lymph nodes. In addition, microparticles facilitate antigen presentation by APCs via both MHC class I and MHC class II restricted processing and presentation pathways. One of the main advantages of microparticles for targeted antigen delivery is that they can be a flexible delivery platform that can be used to deliver both antigens and immunostimulatory molecules.

Other potential antigen delivery systems include polyphosphazenes, a class of synthetic polymers consisting of a backbone with alternating phosphorus and nitrogen atoms and organic side groups attached to each phosphorus (Mutwiri *et al.*, 2007). Immune stimulating complex (ISCOM), which is a small 40 nm nanoparticle composed of saponin (adjuvant), lipids and antigen, and has been described as an antigen delivery system because it not only has adjuvant activity and but also the ability to target APC (Morein *et al.*, 2004). A commercial ISCOM-based vaccine against equine influenza has been licensed for many years (Heldens *et al.*, 2009).

I. VACCINE DELIVERY

Vaccine delivery comprises a diverse range of approaches with the overall goal of providing vaccines for mass vaccination during disease outbreaks and delivery of vaccines to wildlife. Oral vaccines used in rabies vaccination of wildlife such as foxes were initially based on attenuated rabies vaccine viruses such as the ERA strain, but concerns that these vaccines could rarely cause rabies (Fehlner-Gardiner *et al.*, 2008) have largely led to their replacement (http://ec.europa.eu/food/animal/diseases/eradication/rabies_pres_19.pdf). In Canada, a live adenovirus-vectored rabies vaccine with a good safety profile (Knowles *et al.*, 2009) is currently being used in rabies vaccination campaigns directed at controlling skunk and raccoon rabies (Rosatte *et al.*, 2009). The live oral vaccinia-rabies glycoprotein (V-RG) vaccine is widely used elsewhere, and attempts are being made to optimise the vaccine baits for efficacy for other species including dogs (Cliquet *et al.*, 2008). Rabies infection in stray dogs and wildlife

represents a serious problem for humans globally, and research for safer, more stable and efficacious live oral rabies vaccines continue (Faber *et al.*, 2009). Other possibilities for mass vaccination using edible plant-made vaccines have been actively investigated, but in spite of biotechnological advances in plant expression of vaccine antigens, no commercial products for oral use have been identified to date (Rice *et al.*, 2005).

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Appendix IV

New template for the vaccine section of disease-specific chapters of the OIE *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals*, adopted May 2009

C. REQUIREMENTS FOR VACCINES

1. Background

a) Rationale and intended use of the product

2. Outline of production and minimum requirements for conventional vaccines

a) Characteristics of the seed

- i) Biological characteristics
- ii) Quality criteria (sterility, purity, freedom from extraneous agents)

b) Method of manufacture

- i) Procedure
- ii) Requirements for substrates and media
- iii) In process controls
- iv) Final product batch tests
 - Sterility/purity
 - Safety
 - Batch potency

c) Requirements for authorisation

- i) Safety requirements
 - Target and non-target animal safety
 - Reversion-to-virulence for attenuated/live vaccines
 - Environmental consideration
- ii) Efficacy requirements
 - For animal production
 - For control and eradication
- iii) Stability

3. Vaccines based on biotechnology

a) Vaccines available and their advantages

b) Special requirements for biotechnological vaccines, if any

Appendix VOriginal: English
January 2010**REPORT OF THE OIE/FAO/WHO MEETING ON THE ASSESSMENT OF FOOD SAFETY
RELATED TO THE USE OF RECOMBINANT VACCINES IN FOOD-PRODUCING ANIMALS****Paris, 18–19 January 2010**

An OIE/FAO/WHO meeting on the Assessment of Food Safety Related to the Use of Recombinant Vaccines in Food-Producing Animals was held at the OIE Headquarters in Paris from 18 to 19 January 2010. The meeting was chaired by Dr David Mackay. Dr Donna Hutchings accepted to act as rapporteur. The Agenda and List of Participants are given at Appendices I and II, respectively.

1. Introduction

The Group was welcomed by Dr Elisabeth Erlacher-Vindel (Deputy Head, OIE Scientific and Technical Department), on behalf of Dr Bernard Vallat, Director General of the OIE.

Dr Erlacher-Vindel provided an update on OIE activities related to the *ad hoc* Group on Vaccines in Relation to New and Emerging Technologies, which met in November 2008 and again in November 2009.

2. Adoption of the agenda

The Group accepted the agenda as proposed by OIE. It was noted that observers from industry would be available in the afternoon to provide information upon request.

3. Clarification of the scope of the discussion on the use of recombinant vaccines in food-producing animals

The Group's discussion was focused on non-heritable constructs, as heritable constructs were not considered to fall within the generally understood definition of vaccines, including that used within the OIE *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (Terrestrial Manual)*.

While noting that there was some variation in the use of generic terms such as “genetically modified (GM)”, “genetically engineered (GE)” and “recombinant vaccines”, the Group chose to use the term GE vaccine as the standard term for this discussion, thus including all common types.

The Group noted that vaccination with GE vaccines, as with conventional vaccines, that result in persistency or latency of the vaccine organism (e.g. Herpesvirus, DNA vaccines) require specific evaluation of safety including food safety, but that animals vaccinated with GE vaccines should not be considered as genetically modified animals.

The Group agreed that the scope of the current discussion was on safety of GE vaccines in food animals, and not on vaccines to reduce shedding of organisms relevant to food safety.

4. Outline of the approach to assessing the safety of recombinant vaccines with examples

The Group discussed international requirements for licensing of GE vaccines for animals, in particular safety requirements, and noted that regional requirements were not well known. This point was confirmed by the observers from industry.

The Group noted that the classification of pathogens with respect to their inherent pathogenicity for humans and animals was important for the safety evaluation of vaccines derived from them, e.g. Class 1 organisms present less of an inherent safety risk than those in Classes 2 or 3, however classification might vary between regions. The Group agreed that the use of an internationally standardised classification of veterinary pathogens was desirable to help in the assessment of safety.

The Group concluded that only general safety guidance could be provided, as the possible situations were so variable that case-by-case evaluation was needed. The way of demonstrating safety was different amongst various products; for example, replication of some vaccine vectors was abortive, and most animal vaccine strains were not zoonotic.

The Group agreed that the general advice and requirements for safety evaluations on all vaccines should also apply to GE vaccines, and that licensing authorities should ensure that food safety is an element included in risk assessments. The type of vaccine, the target species, novelty, dose and route of administration (individual or mass vaccination) might determine the complexity of the safety evaluation, in particular the use of zoonotic agents. If relevant, a withdrawal period for the different food products (e.g. meat, eggs, milk) would be established. MRL (maximum residue limits or tolerance limits) were required to be set for pharmacologically active ingredients, including adjuvants and excipients in vaccines. This could apply to toxins used to enhance immunity to vaccines, e.g. lectins and cholera toxin.

The European Medicines Agency provided two examples of risk assessments for the discussion, including live canarypox vectored vaccines in horses and live Marek's disease vectored vaccines in chickens. It was noted that CFIA (Canadian Food Inspection Agency) had similar conclusions regarding safety of the same products: <http://www.inspection.gc.ca/english/anima/vetbio/eae/eae.html>. In both of these cases, and in many other cases of which the Group was aware, the starting point for producing the GE vaccine strain was a well characterised vaccine strain with a long history of safe use. Indeed some such strains had been investigated for their potential for use in humans and therefore much was known about their safety with respect to human exposure. In such cases it was possible for regulators to adopt an approach similar to that described in the Codex Alimentarius Guidelines for the Conduct of Food Safety Risk Assessments of Foods Produced Using Recombinant-DNA Microorganisms (CAC/GL 46-2003) whereby knowledge of the extensive safe use of the parental organism is fully taken into account when considering the potential impact of introduced genetic changes. The Group recognised that more information would be required in situations where the vector organism had not previously been licensed for use in food-producing species.

The Group considered that DNA vaccines (i.e. vaccines consisting of purified recombinant DNA) constituted a special case. However, very few such vaccines had been commercialised for food-producing species and the Group considered that economic and other factors make it unlikely that their use would become widespread. In general, these vaccines were not considered to pose a significant food safety risk, as only low amounts of administered DNA were likely to be present in vaccinated animals at the time of slaughter and any such DNA would rapidly be degraded through digestion.

The Group considered it was important to differentiate clearly between genetically modified foods and the use of GE vaccines. In the case of food, the intention is to introduce a new trait into a food that will still be present when the food is eaten by the consumer. In the case of GE vaccines, the intention is generally to induce a protective immune response by means of an immunogen that is often no longer itself present at the time the animal is slaughtered. The Group recognised that this was a generalisation to which there are exceptions and it was therefore necessary to assess risk on a case-by-case basis. Nevertheless this difference in principle was considered important to emphasise, particularly to those not familiar with the principles of vaccinology.

5. Review of relevant guidance in the OIE *Terrestrial Manual*

The Group reviewed the existing relevant chapters from the OIE *Terrestrial Manual*, including the text of Chapter 1.1.8 Principles of Veterinary Vaccine Production, and recommended addition of a new Appendix to Chapter 1.1.8 on benefit–risk assessment of veterinary vaccines, including GE vaccines, with a specific section on safety including food safety.

It was noted that a paper, available in an OIE publication, could be used as the basis for the safety part of the Appendix on benefit–risk evaluation (Safe use of vaccines and vaccine compliance with food safety requirements, K. Grein *et al.*, *Rev. sci. tech. Off. int. Epiz.* [2007]. **26**, 339–350):

http://www.oie.int/boutique/index.php?page=ficprod&id_prec=119&id_produit=200&lang=en&fichrech=1&PHPSESSID=f16022cedb6ce265c164795d31c9a6cb

6. Conclusions

Considering the following factors:

- Licensed GE vaccines have a history of safe use in animals for about 20 years, as the safety of GE vaccines is currently well defined and predictable due to knowledge of the genetics of these vaccines, and is ensured through a system of thorough review with licensing;
- Types of technology available for GE vaccines are continually evolving, yet those with established safety profiles remain preferred in practice. Current types of technology for GE vaccines are described in the draft *Terrestrial Manual* Chapter 1.1.7 The Application of Biotechnology to the Development of Veterinary Vaccines;
- Food safety aspects of GE vaccines other than the active ingredient (e.g. excipients, adjuvants) are not different from conventional vaccines, and are taken into account during licensing, establishing residue limits and withdrawal periods when necessary;
- An evaluation of food safety is already considered an integral part of the overall benefit–risk assessment that forms the basis of any decision by a regulatory authority to licence (authorise) a vaccine, GE or not;

The Group recommended that:

- a) Food safety, as part of overall human safety assessment, should not be separated from overall safety for users and consumers; the review of food safety should therefore be kept in context.
- b) The standard of safety should not be different between GE vaccines and conventional vaccines.
- c) Animals vaccinated with GE vaccines should not be considered GM animals.
- d) At present, no need was identified for new international guidance on risk assessments for GE vaccines other than an update to the OIE *Terrestrial Manual*.
- e) A new Appendix should be added to the OIE *Terrestrial Manual* Chapter 1.1.8. on benefit–risk assessment of veterinary vaccines, including GE vaccines, with a specific section on safety including food safety.

The Group further noted that:

- f) Although DNA vaccines are not currently used in terrestrial food animals, this type of GE vaccine may require closer scrutiny on a case by case basis as there is currently little experience on which to base an assessment of their safety under normal conditions of use.

- g) Concerning allergenicity, GE vaccine strains may not be an issue if the vector and inserted genes are naturally occurring in animal pathogens that may be present in food. The allergenicity of other ingredients, including excipients, adjuvants and preservatives, should be monitored through pharmacovigilance as for any other type of vaccine.

7. Any other business

None

8. Adoption of the report of the meeting

The Group adopted the report of the meeting.

.../Appendices

Appendix I

**MEETING ON THE ASSESSMENT OF FOOD SAFETY
RELATED TO THE USE OF RECOMBINANT VACCINES IN FOOD-PRODUCING ANIMALS**

Paris, 18–19 January 2010

Agenda

1. Introduction and welcome
 2. Agreement on proposed ToRs and draft agenda
 3. Clarification of the scope of the discussion on the use of recombinant vaccines in food-producing animals
 4. Outline of the approach to assessing the safety of recombinant vaccines with examples
 5. Review of relevant guidance in the OIE *Terrestrial Manual*
 6. Conclusions
 7. Any other business
 8. Adoption of the report of the meeting
-

Appendix II

**MEETING ON THE ASSESSMENT OF FOOD SAFETY
RELATED TO THE USE OF RECOMBINANT VACCINES IN FOOD-PRODUCING ANIMALS**

Paris, 18–19 January 2010

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Appendix III

**MEETING ON THE ASSESSMENT OF FOOD SAFETY
RELATED TO THE USE OF RECOMBINANT VACCINES IN FOOD-PRODUCING ANIMALS**

Paris, 18–19 January 2010

Terms of Reference (ToRs)

1. To review available scientific information on aspects of food safety that might be related to the use of recombinant vaccines in food-producing animals and provide relevant scientific advice on this issue as necessary.
 2. To review the current guidance in the OIE *Terrestrial Manual* and the proposed revisions of the *ad hoc* Group on Vaccines in Relation of New and Emerging Technologies, as it relates to assessing the safety of vaccines, in particular those derived using recombinant technology.
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**REPORT OF THE THIRD MEETING OF THE OIE AD HOC GROUP
ON VALIDATION OF DIAGNOSTIC ASSAYS
Paris, 20–22 January 2010**

The third meeting of the OIE *ad hoc* Group on Validation of Diagnostic Assays was held at the OIE Headquarters in Paris from 20 to 22 January 2010.

The meeting was chaired by Dr Rich Jacobson and Dr Axel Colling acted as rapporteur. The Agenda and the list of participants are presented as Appendices I and II of this report, respectively.

1. Introduction

Dr Elisabeth Erlacher-Vindel, Deputy Head of the Scientific and Technical Department, welcomed the members of the Group on behalf of the OIE Director General, Dr Bernard Vallat and thanked all of them once more for the documents they drafted in preparation of this meeting. Prof. Vincenzo Caporale, President of the OIE Biological Standards Commission, presented to the members the future activities of the Biological Standards Commission in the area of the validation of diagnostic assays. He pointed out the importance of international reference reagents and their availability for the diagnosis of animal diseases of interest for the OIE and he confirmed that the Commission would keep on working in collaboration with the OIE Reference Laboratories to reach this objective. He also mentioned the need to take into account the use of diagnostic tests for mass screening and to reflect this potential use and the interpretation of data in the guidelines for validation. He finally suggested to the Group that the essential points on validation need to be accessible for the user of the *Terrestrial Manual* who is looking for general guidance.

2. Discussion and finalisation of the drafts of the seven appendices for the introductory chapter on “Principles and methods of validation of diagnostic assays for infectious diseases”

The Group recalled that the second meeting of the Group, in February 2009, had identified the authors for the seven different appendices on “Best Practices” among the participants of the Group. A first version of each of these appendices was drafted and circulated for comments within the Group at the end of 2009. The present meeting of the Group had as a main objective to discuss and finalise the relevant documents as far as possible.

The Group first discussed the length of the appendices, pointing out the difficulty of being both concise and complete at the same time, considering the issues addressed. The appendices would certainly be around 10-15 pages each and therefore this might be too long to be attached as a group of appendices to the introductory chapter on validation.

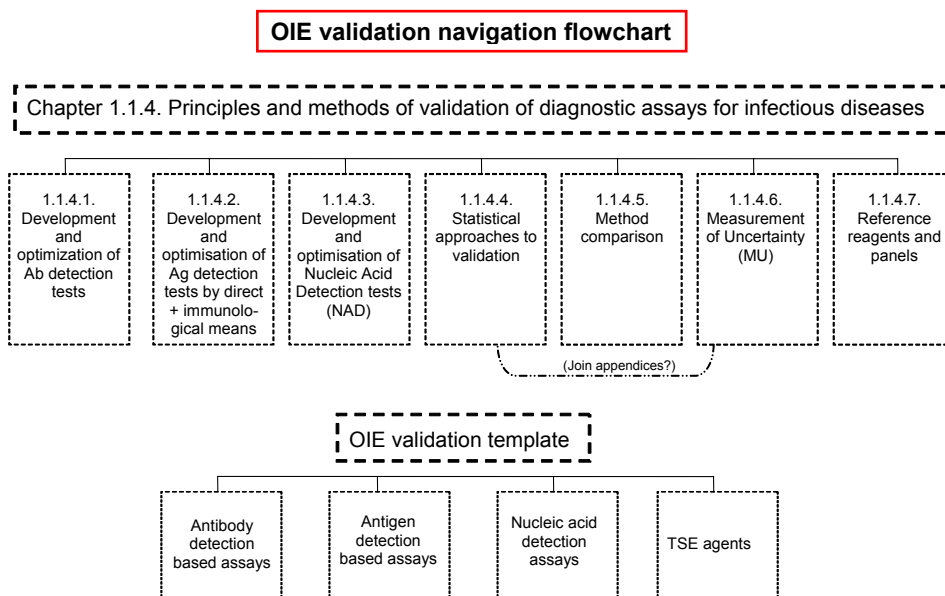
The Group agreed to propose to the Biological Standards Commission two options:

1. To have the introductory chapter with all the appendices in the *Terrestrial Manual*, or
2. To have the introductory chapter stand alone in the *Terrestrial Manual* and publish, in a parallel publication, the introductory chapter with all the appendices (in the same format, for example, as the OIE Quality Standard and Guidelines for Veterinary Laboratories).

The Group reviewed and discussed all the draft appendices and made substantial improvements. For ease of cross referencing, a common general template, following the outline of the chapter, was adopted for the three appendices on the development and optimisation of assays (antibody detection, antigen detection and nucleic acid detection).

Due to the lack of time, the Group could not finalise all of the appendices and agreed to complete the work by email communications so that final versions would be provided to the Biological Standards Commission by 31 May 2010.

The Group agreed that other appendices on modern topics such as microarrays might be necessary in the future to address the validation of novel diagnostic test technologies and recommended that the option to add additional appendices be left open. The current OIE validation navigation flowchart is summarised below:



3. Discussion of the final modifications to the chapter on “Principles and methods of validation of diagnostic assays for infectious diseases”, as informed by the writing of the best practices

Following the discussions on the different appendices, the Group was now harmonising the terminology and definitions used in the introductory chapter on “Principles and methods of validation of diagnostic assays for infectious diseases” with those of the appendices in order to be consistent throughout. The Group was also updating the glossary of the *Terrestrial Manual* and the “OIE Quality Standard and Guidelines for Veterinary Laboratories” to cover the specific terminology used in the chapter and the appendices. The Group agreed that the final version of the Chapter would be sent to the Biological Standards Commission by 31 March 2010.

4. Discussion of Dr Ingrid Bergman’s request regarding reference panels

The Group congratulated Dr. Bergmann for her efforts in assembling and characterising the bovine evaluation serum panel for non-structural protein tests for foot and mouth disease diagnosis and providing draft guidelines on use of this reference panel. The Group has reviewed the draft with special attention to “... how such a panel might be used in evaluating other tests” and considered it a very useful resource document.

The Group was currently drafting best practices on reference reagents and panels for use in development and validation of assays and this would appear as the seventh appendix of the validation chapter. The Group agreed not to comment on Dr Bergmann’s suggestions until the best practices on reference panels had been completed (May 2010), due to the many factors under consideration that must be addressed in development of the best practices document.

5. Evaluation of the need for guidelines (for trade) in cases where a validated diagnostic test is not available for a disease and/or a species (e.g. wildlife, certain domestic camelids)

The Group noted that there might be tests or procedures that were not species-specific for agent detection (e.g. culture, NAD detection, antigen detection) that could be used as a starting point. The tests or procedures should be verified using reference material, if available, from OIE Reference Laboratories. For antibody detection, where tests might exist for one species for that particular disease, the question then was to know whether the test was valid in another species. The first consideration should be species specificity and the second should be the infection/disease threshold for the pathogen of interest. Finally, the test type, e.g. competitive ELISA, might make a difference since some antibody tests had broader species applicability. If the test appeared to be able to be used in trade, it should go through at least a Stage-1 (analytical) validation before being used. In the absence of full validation, it would be wise to add a disclaimer that indicates that the test in question had only been partially validated for that disease and host species as specified in the OIE validation template. Another *ad hoc* Group might be necessary to provide detailed guidance on this issue.

6. Discussion on the advancement on the guidelines to accompany the template used in the framework of the OIE Procedure for validation and certification of diagnostic assays

Due to time limitation, the Group did not address this issue but noted that the development of these guidelines would be finalised by the authors identified, once the introductory chapter and appendices had been finalised, so that the overall flow would be consistent and harmonised (see also OIE validation navigation flowchart above).

.../Appendices

Appendix I

**MEETING OF THE OIE AD HOC GROUP ON VALIDATION OF DIAGNOSTIC ASSAYS
Paris, 20–22 January 2010**

Agenda

1. Introduction, Announcements and Approval of Agenda
 2. Discussion and finalisation of the drafts of the 7 appendices for the introductory chapter on “Principles and methods of validation of diagnostic assays for infectious diseases”:
 - a. Development and optimisation of NAD tests: Drs Belak and Webster
 - b. Development and optimisation of antibody detection assays: Dr. Colling
 - c. Development and optimisation of antigen detection by immunological means: Dr. Colling
 - d. Statistical approaches to validation: Dr. Gardner
 - e. Methods to assess comparability (Equivalency) of assays: Dr. Gardner
 - f. Measurements of uncertainty: Dr. Colling
 - g. Selection and use of reference panels: Drs. Wright and Jacobson
 3. Discussion of the final modifications to the introductory chapter on “Principles and methods of validation of diagnostic assays for infectious diseases”, as informed by the writing of the best practices.
 4. Discussion of Dr Ingrid Bergman’s request regarding reference panels.
 5. Request from the Biological Standards Commission addressed to the *ad hoc* Group: Evaluation of the need for guidelines (for trade) in cases where a validated diagnostic test is not available for a disease and/or a species (e.g. wildlife, certain domestic camelids).
 6. Discussion on the advancement on the guidelines to accompany the template used in the framework of the OIE Procedure for validation and certification of diagnostic assays.
 - a. Antibody detection based assays: (Drs. Wright and Gardner)
 - b. Antigen detection based assays: (Drs. Wright and Gardner)
 - c. Nucleic acid detection assays: (Drs Webster, Belak and Gardner)
 - d. TSE agents (Drs Webster and Gardner)
 7. Any other business
-

Appendix II

MEETING OF THE OIE AD HOC GROUP ON VALIDATION OF DIAGNOSTIC ASSAYS

Paris, 20 – 22 January 2010

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MEETING OF THE OIE AD HOC GROUP ON DISEASES OF CAMELIDS

Paris, 2010

Draft of Terms of Reference

- Update the list of diseases of camelids proposed by the *ad hoc* Group during its first meeting, taking into account any recent publications and the ISOCARD Conference held in Djerba, Tunisia, in March 2009;
 - Determine the diseases of priority for validation of diagnostic assays and the need for research in this area;
 - Review the disease-specific chapters of the OIE *Manual of Diagnostic Tests and Vaccines for Terrestrial Animals* for the diseases that concern Camelids in view of adding, if necessary, specific requirements for diagnostic assays and vaccine production, and propose an outline for these revisions and a timetable to draft them;
 - Provide an update on the current disease situation with regards to camelids worldwide;
 - Consider setting up an OIE laboratory network on diseases of camelids linked with already existing networks on this topic.
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