KEYNOTE: DRIVING UP GLOBAL STANDARDS FOR FMD DIAGNOSTIC: A KEY ROLE FOR PROFICIENCY TESTING AND INTERNATIONAL ORGANISATIONS.

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ABSTRACT

Laboratory diagnostics are used for foot-and-mouth disease (FMD) outbreak confirmation, post-outbreak and post-vaccination surveillance, vaccine matching, and import/export control. Participation in a national quality assurance (QA) system and international proficiency testing (PT) schemes is essential to maintain confidence in results between different laboratories. A workshop on PT, with the National Reference Laboratories (NRL) for FMD and classical swine fever in the European Union, recommended: the yearly organisation of PT, the use of the ISO 17043/43 standard to guide the development of proficiency testing schemes for the organisers and participants; the primary goal of PT schemes to be the evaluation of laboratories with well defined criteria for lab conformity prior to the start; the establishment of an Advisory Board; a clear feedback with a draft report prior to the annual meeting including results, conclusions on non-conformities and recommendations; corrective actions to address non-conformities agreed between Community Reference Laboratory and NRL and communicated to DG SANCO. Regional Reference Laboratories organising PTs should also be evaluated via a PT schemes supervised by international organisations such as FAO or OIE. The availability of reference standards is fundamental in a QA system but the production and evaluation through collaborative studies is highly expensive, time consuming and requires a serious international financial investment. Commercial ELISAs and (real time) RT-PCR kits are currently available and should be fully validated and licensed. Recently the OIE implemented a system with different levels of assay validation giving private companies the opportunity to submit a dossier for international recognition. However, the question remains what to do if an assay without this international recognition is used to support a country’s international disease status recognition. Moreover, an assay recognition or marketing authorisation does not provide a sufficient guarantee for the quality of the test kit batches and therefore serial release testing on each serial of FMD kits that will be used in routine testing is highly recommended.

1. INTRODUCTION

International trade in animals and animal products is liberated and confidence in this global trade can increase only if appropriate control measures are applied. As foot-and-mouth disease (FMD) diagnostics play an essential role in this respect, the Food and Agriculture Organisation European Commission for the Control of Foot-and-Mouth Disease (EUFMD) co-ordinates, in collaboration with the European Commission, several programmes to increase the quality of FMD diagnostics. A quality assurance (QA) system is deemed essential for laboratories using FMD diagnostics aiming to (i) confirm an outbreak; (ii) perform a post-outbreak (sero)surveillance; (iii) test vaccine efficacy; (iv) perform a post-vaccination surveillance; (v) check vaccine matching; control import/ export.

The performance of laboratories participating in a National Accreditation System must be evaluated at least yearly by an Independent EQC body. The latter should be integrated in an international system such as the Organisation for Economic Co-operation and Development (OECD). Key elements in a world-wide quality system are first of all the possibility for all Veterinary Services to have access to FMD diagnosis through a National Reference Laboratory (NRL) or through an agreement with an OIE-FAO Regional Reference Laboratories (RRLs). Moreover, this is a prerequisite with a quality system for Veterinary Services. Furthermore, NRLs should implement an accreditation system with External Quality Control (EQC) and should take part in a Proficiency Testing (PT) scheme. A World PT should also be organised for OIE/FAO RRLs in which International
The availability of International or regional reference material is essential for controlling and comparing test results. The series of devastating epizootics in the last decade increased the commercialisation of FMD diagnostics and therefore procedures for kit and batch control should be developed.

2. PROFICIENCY TESTING TO EVALUATE THE LABORATORY PERFORMANCE

As there is a considerable variation in the organisation of PTs, a workshop was organised for the European NRLs for FMD and CSF with the participation of EC, FAO and OIE. The objective of the Workshop on Proficiency Testing was to make recommendations for Good Laboratory Practice concerning proficiency testing, in order to improve QA/QC of laboratory tests for FMD/SVD and CSF.

Representatives of FMD/SVD and CSF National Reference Laboratories from the following countries were invited and have participated: Austria, Belgium, Bulgaria, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Poland, Portugal, Romania, Slovakia, Slovenia, Spain, Sweden, United Kingdom (the EU member states) and Bosnia Herzegovina, Iceland, Norway, Israel, Turkey, Switzerland, Croatia, Albania, FYR of Macedonia, Serbia, Montenegro (non-EU but EUFMD member states).

The workshop consisted of a number of presentations outlining the problems with regard to the organisation, participation, reporting, follow-up and feedback of proficiency testing followed by a discussion session and a conclusions and recommendations session.

Following recommendations were formulated:

- Need for Guidelines:
  1. The Community Reference Laboratories for CSF and FMD/SVD will use the proposed ISO 17043/43 standard to guide the development of proficiency testing schemes for the organisers and participants.
  2. Use Guide 43 in conjunction with ILAC G13, until ISO17043 is formally published.
  3. The Community Reference Laboratories for CSF and FMD/SVD will develop a Standard Operating Procedure that outlines the proficiency testing scheme. This will not be too prescriptive and they will try to harmonise between FMD and CSF in first instance.

- Evaluation of Lab or Method:
  1. The primary goal of Proficiency Testing schemes is to evaluate lab (individual test and/or test system) performance against assigned values (qualitative or quantitative);
  2. Secondary purposes must be clearly separated in design and reporting;
  3. The scope and purpose of the Proficiency Testing exercises need to be clearly stated in advance.

- Scope of Proficiency Test:
  1. The scope of the Proficiency Test needs to be fit for purpose and the purpose should be clearly defined in advance;
  2. The scope of the Proficiency Test (outbreak, surveillance, etc.) should be agreed and clearly communicated to the National Reference Laboratories prior to the start of the Proficiency Test.

- Statistical analysis:
  1. Statistical guidelines need to be followed where relevant;
  2. The design of PTS should be done so as to maximize the power of statistical analysis;
  3. Both trueness and precision should, if possible, be addressed.

- Criteria for acceptance:
  1. Criteria for lab conformity must be set PRIOR to the start of the Proficiency Test;
  2. The scope will define whether the individual results and/or the complete test system is evaluated;
  3. DG-Sanco should provide a list of contacts for the National Reference Laboratories.

- Advisory board:
  1. An Advisory Board should be established at the beginning of each Proficiency Testing exercise
  2. Membership (~4 persons) – but this should not be too prescriptive:
     • Has to be appointed at the beginning of the Proficiency Test and communicated to all participants;
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- Representatives should mainly be from participating labs;
- Statistical representation of participants;
- The Community Reference Laboratory should represent the EU opinion; A representative from regulatory bodies is not essential.

- Reporting:
  1. Feedback:
     - The Community Reference Laboratories should provide a draft report prior to the annual meeting;
     - The final report should include feedback from Proficiency Test participants (questionnaire);
     - The report should include details of the scope, criteria, statistical approaches, findings, conclusions on non-conformities and recommendations.
  2. Confidentiality:
     - The consensus from the workshop was to maintain confidentiality between participants;
     - Decoded results will be provided to DG Sanco (for EU Member States).
  3. Follow up:
     - Corrective actions to address non-conformities will be agreed between Community Reference Laboratory and National Reference Laboratory and communicated to DG SANCO.
  4. To whom?
     - To all the Proficiency Test participants and DG Sanco;
     - Proficiency Test reports could be placed on a restricted access website.

3. PRODUCTION AND VALIDATION OF REFERENCE MATERIAL/REFERENCE STANDARDS

It is clear that the key element in the validation of tests and in the comparison of test results among laboratories used for the control of transboundary diseases or for trade purposes is the availability of reference standards. At present, only 3 sera from cattle against FMD are internationally recognised by the OIE as reference standards. Weak positive and strong positive reference standards, as well serological as virological, against all serotypes (O, A, C, Asia1, SAT1, SAT2, SAT3) and within the serotypes against some important topotypes are urgently needed. Therefore several standards must be produced, tested by different laboratories and a final selection must be made. This is a huge task, which can only be performed by a network of laboratories through a ‘Collaborative Study’ with the specific aim of establishing international standards. Trying to get agreements on standards as a secondary goal of proficiency tests is not recommended. The standards agreed upon in the Collaborative study can then be considered as primary standards and distributed among other laboratories to calibrate their in house tests. Based on this calibrated test the laboratories should make their own secondary standards to use in routine diagnosis. The main difficulty in this respect is the fact that a lot of laboratories have not the animal facilities to produce secondary standards. Especially producing standards from infected animals to serve as secondary standards in e.g. the NSP tests is impossible for most European laboratories. Thus large quantities of secondary standards should be produced and made available to FMD laboratories. As well standards from vaccinated as from infected animals must be produced. To be recognised as international reference standards the production must fulfil international recognised criteria and specific information must be available as specified by the OIE.

The standards should be similar in nature to those routinely tested by participants (fit for purpose). The importance of one species (cattle, pigs, sheep, goats) versus another depends on the region. For some laboratories it will be very important to have secondary standards from pigs while others need sheep sera. The development of standards from different species has, therefore, to be considered. Special attention is needed in providing evidence of the homogeneity and stability of the newly produced standards. Adequate packaging and transport methods to protect the stability and characteristics are essential taking into account the biosecurity hazards.

4. VALIDATION AND CERTIFICATION OF DIAGNOSTIC ASSAYS

Recently, the OIE established an evaluation process for the Validation and Certification of Diagnostic Assays. A Standard Operating Procedure (SOP) was developed and is available to all kit producers. Producers have to enter their application form and the application will be subjected to formal evaluation by an expert panel appointed by the OIE, as detailed in the SOP. The applicant is highly recommended to consult the SOP and the following chapters in the OIE Manual (2008):
Chapter 1.1.3., "Principles of validation of diagnostic assays for infectious diseases" and Chapter 1.1.4., "Validation and quality control of polymerase chain reaction methods used for the diagnosis of infectious diseases". Although this OIE procedure is a major step forward in the harmonisation of a world-wide quality control system, some serious questions remain without answer such. Is the OIE certificate now internationally recognised or will individual countries maintain their own system of kit certification? If producers will have to certify their kits in all countries then there is no interest for them to go centrally to OIE. What will happen if a manufacturer does not enter an application to OIE for the certification of its kits and those kits are used for the recognition of a change in disease status? Is a validation by a NRL or RRL sufficient? In any case an independent evaluation is essential!

5. BATCH CONTROL

In some parts of the world large difference in quality between batches of the same FMD diagnostic kit could occur. To guarantee a high grade of reliability, it is necessary to test (if possible partly by the producer) each serial (lot - batch) and to describe its capacities with regard of sensitivity and specificity. As a consequence, some countries established governmental regulations for batch quality control and require manufacturers to submit a report of the test results for each batch or lot of kits produced. Batches with satisfactory test results are approved for marketing. This licensing authority, however, may select random samples for confirmatory testing. In other countries, marketing is allowed upon certification that a batch was produced and monitored in accordance with a 'good manufacturing practice' marketing authorisation. Animal Health authorities may require additional testing of kits used in disease control programmes sponsored by the government. Generally, the purpose of such testing is to ensure that the performance of the kit is appropriate for the proposed use. Most countries require retention of samples from each batch of kits for future examination should problems arise.