Diagnostic Laboratories including National, Regional and International networking

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Introduction

Role of the veterinary laboratory

Current diagnostic tests

Laboratory networking

Conclusion
Introduction

Control and prevention of peste des petits ruminants depends on:

- Capacity to sample, detect and confirm disease at the earliest possible time for the design of appropriate response measures.

- Diagnostic measures in place at national level should ideally combine with networks to allow for a regional approach in PPR management.
Role of the Veterinary Laboratory

- Establish diagnosis to complete observations of clinical symptoms;

- Implement quality diagnosis with standardised methods to deliver reliable PPR diagnosis results;

- Share with epidemiology units sampling workplans for:
  - Serosurvey
  - Virus studies.
Laboratory confirmation of clinical cases of PPR is compulsory

PPR can be easily confused with other diseases such as:

- Bluetongue, CCPP, Pasteurellosis,
- Definitive diagnosis of PPR is demonstrated when combined with clinical observations and epidemiological data.
- Essential that diagnosis rely on validated, sensitive and specific tools either for virology, serology, molecular biology.
Current laboratory tests

- **Virology tests**: ANTIGEN and GENE detection
  - Antigen Capture Elisa; Lateral Flow Device (LFD; field test)
  - Conventional RT-PCR
  - Real-time RT-PCR
  - LAMP PCR (field test)
  - VIRUS
  - Isolation on Vero cells
  - Isolation on Vero Slam cells
  - Characterization by sequencing

- **Serology tests**: ANTIBODY detection
  - VNT (OIE prescribed test for international trade)
  - c-Elisa,
Virology tests

- It is crucial to provide laboratories with efficient tools allowing the early detection of PPR emergence/re-emergence and to conclude on the origin of the virus.

- Conventional RT-PCR, now widely implemented in labs, allows direct sequencing and thus for the genotyping of strains.

- With recent technical breakthroughs on NGS, diversity of field strains is established, facilitating source tracking and understanding disease diffusion pathway.
Virology tests

High capacity labs: different steps:
Real time RT-PCR can be used as a screening tests and RT-PCR in association with viral isolation allows for strain genotyping.

Ref Labs

Samples

Strain GENOTYPING:
500 samples/25 days

RNA extraction
50 minutes of 88 samples

QRT-PCR Screening

P2 Lab P3 Lab

Viral isolation

Sequencing

RT-PCR amplification
Virology tests

Oppositely, in many laboratories or resource limited, simple, rapid and robust assays can be adopted as routine techniques, able to detect viral:

- **Antigen**: such as Antigen Capture Elisa,
  
  Sensitivity = Q-RTPCR

- **Pen-side tests**: LFD

- **Gene**: LAMP-PCR: RT- loop-mediated isothermal amplification at 63°C obtained 60min, observed by the naked eye
  
  Sensitivity = Q-RTPCR = 10-fold higher than conventional RT-PCR

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**Bst**: *Bacillus stearothermophilus*
Serology tests

ELISA (developed 30 years ago)

- These tests are able to promptly detect new outbreaks of PPRV and to produce data on the prevalence in infected areas.

- A set of ELISAs were developed.

  ✓ Competitive ELISA (C-ELISA) are H or N-Mab-based, high degree of correlation to the VNT, the gold standard assay.

Well adapted (96 wells format) to large scale serology studies
### OIE manual: Purpose of the methods

<table>
<thead>
<tr>
<th>Method</th>
<th>Target</th>
<th>Confirmation of clinical cases</th>
<th>Population freedom from infection</th>
<th>Individual freedom from infection</th>
<th>Prevalence of infection - Surveillance</th>
<th>Immune status in individual animals - Vaccination</th>
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<tbody>
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</tbody>
</table>

**Key:**  
+++ = recommended method;  
++ = suitable method;  

**Source:** Last version Chapter 2.7.11. – Peste des petits ruminants
Main challenges/needs for Veterinary Laboratories

- Overall technical level has increased but still...
- Number/quality of samples?
- Reliability of lab results?
- Continuity in reagents availability?
- Staff: competence, future generations?
- National priorities versus global priorities?
- Solution: Break the isolation of national teams
  Build synergy and efficiency
- Networking implies:
  - close collaboration
  - coordination of activities between countries in a given region.
Veterinary laboratory networking in Africa: Lesson learned from rinderpest.

- PARC and PACE programs.
- Capacity building for diagnosis of animal infectious diseases was reinforced.
- Collaboration and communication between veterinary laboratories.
- Perfect setting to create a network in order to adopt the ELISA method (technical support of FAO/IAEA) for serological surveillance.
- Concept of regional laboratories was introduced (KARI, Muguga; ISRA Dakar and LANADA, Bingerville).

PARC and PACE programs aimed and succeeded to eradicate rinderpest and strengthen surveillance of other epizootic diseases.
AU-PANVAC inherited from these projects.
Veterinary laboratory networking in Africa: Concerns about avian influenza (HPAI).

- In Africa, laboratory networking is present and currently operational.
  - Build synergy and efficiency in terms of laboratory and epidemiological expertise etc...

  - Central to detecting and reporting outbreaks, for tracking the virus.

Motivated and re-enforced many initiatives concerning training and capacity building at veterinary laboratory level and improved sustainable networks. Focused the attention of the international community.
Veterinary laboratory networking in Africa: Main activities of these networks. Exemple of RESOLAB

Laboratories are encouraged to collaborate notably:

- Diagnostic techniques harmonization,
- Implementation of quality assurance, disease reporting,
- Link with epidemiological surveillance networks to increase the number of samples collected and analyzed by:
  - Sharing with the epidemiological unit sampling workplans
  - Improving sample logistics and cold chain

- Specific network on PPR is being built
- Involved in the USAID-funded IDENTIFY
  - Technical training in field pathology, diagnostic techniques
  - Support for pathogen sequencing, quality assurance and biosafety/biosecurity practices
The Reference laboratory (3 at present in the world for PPR) plays an important role in assisting and supporting the training and diagnostic activities within regional networks, giving them opportunities to:

- Participate to ring trials,
- Involved in OIE twinning projects,
- Providing training relating to PPR, supplying reagents, scientific and technical knowledge.

A major issue for the laboratory networks is the close involvement of international reference laboratories. Parent - candidate twinning consists in strengthened diagnostic / quality assurance (including metrology), in strengthened disease surveillance capabilities adapted to the specific epidemiological situation in the country.
Conclusion 1/2

- It is crucial that laboratories implement efficient diagnostics allowing the early detection of PPR.
- All these tests will allow to appreciate at national level:
  - the presence/spreading of the disease into new areas or to certify freedom from the disease.
Conclusion 2/2

- It is important to integrate national/regional laboratories activities and epidemiological surveillance networks. Benefit from FAO and OIE capacity building projects, support from reference laboratories:

- Allow to:

  ✓ Clarify the regional situation of peste des petits ruminants and understand PPRV diffusion pathway,

  ✓ Map the health risk areas to improve the coordination of prevention and control measures.

![Map showing regions with different lineages of PPRV](imageurl)
Merci de votre attention