

FMD Vaccine Bank & impact on diagnostic assay use

National Animal Health Laboratory
Network (NAHLN) Symposium

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The North American FMD Vaccine Bank is a strategic reserve of antigens for the rapid production of vaccine in case the FMDV is accidentally or intentionally introduced in North America.

North American FMD vaccine Bank (NAFMDVB)

- Created in 1982
- USA, Canada and Mexico
- Stores antigen to be finished as FMD oil vaccines.

Governing Bodies of the NAFMDVB

Governing Bodies:

- The commission
- The Technical Committee

Membership:

Commissioners: The CVO's of the three countries

The Technical Committee: One technical and one regulatory representative from each country.

Administration:

The administration of the NAFMDVB is done by APHIS.

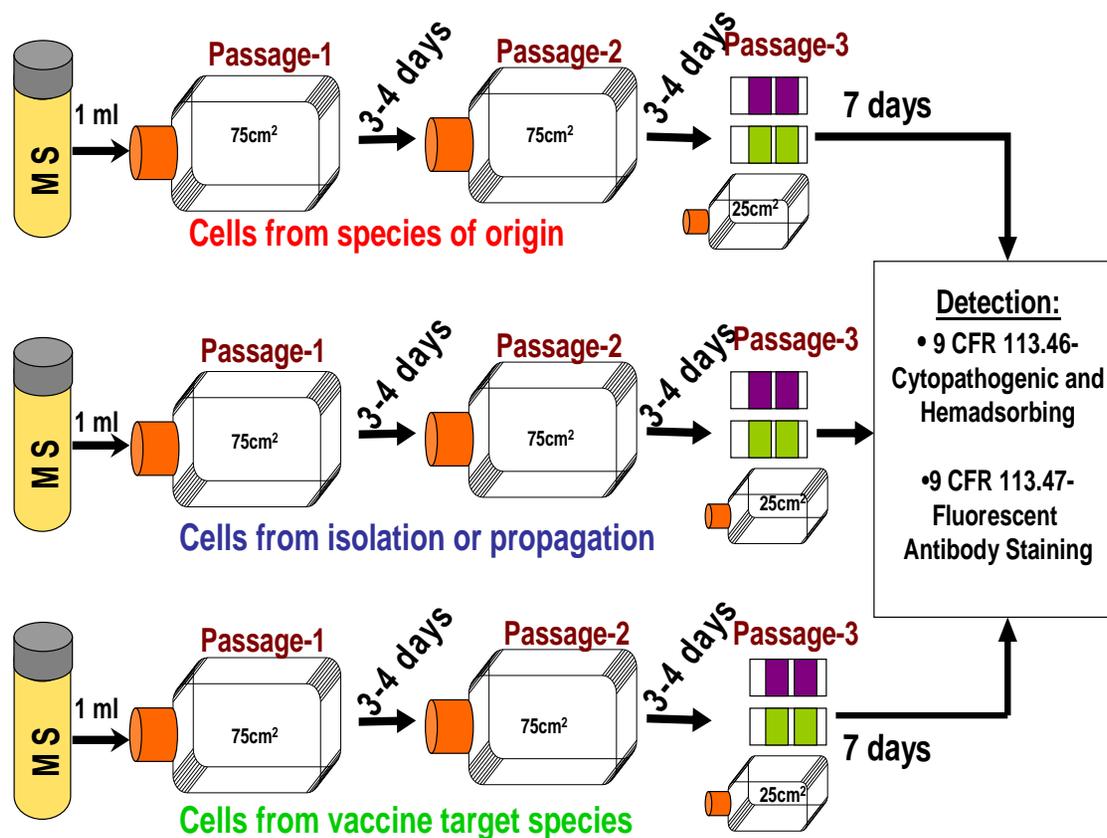
NAFMDVB operation

- Antigens are acquired from foreign manufactures (contracts).
- Master seed viruses, antigens and pilot vaccines are tested before accepting the antigen for stockpile.
- Antigens are stored on LN
- The antigens are tested periodically for stability

Vaccine Testing

- MSVs are tested for the presence of extraneous agents.
- Antigen samples are tested for safety
- Pilot vaccines are tested for potency and efficacy.
- Stability of antigens is tested for potency and efficacy.

Diagram 1. Passage and detection of extraneous agents in MS
(9 CFR 113.55)



Detection of Extraneous Agents in Master seed virus (cont) 9 CFR 113.55

Virus detection with Fluorescent Antibodies (FA)

Bovine adenovirus 1 and 5
Bovine Virus diarrhea I and II
Bluetongue virus
Bovine parvovirus
Bovine respiratory syncytial virus

Porcine adenovirus
Porcine parvovirus
Transmissible gastroenteritis virus
Hemagglutinating encephalitis virus
Reovirus
Rabies virus

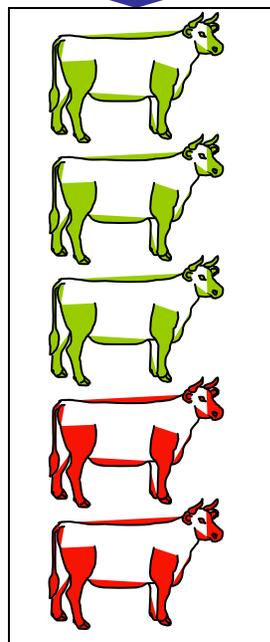
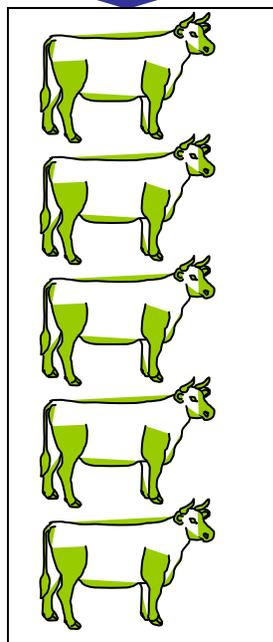
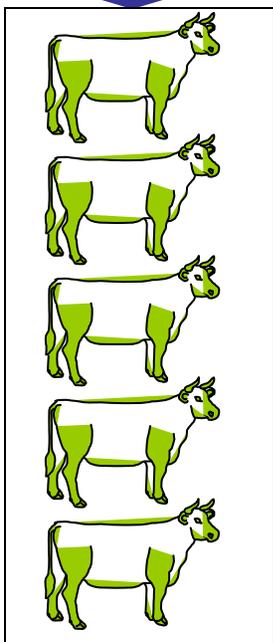
Potency test PD50

3 groups of 5 vaccinated cattle

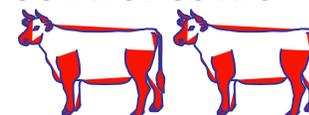
2.0 ml
1 ds

0.5 ml
1/4 ds

0.125 ml
1/16 ds



2 unvaccinated control cattle



Vaccinated and control cattle are challenged with virulent virus 21 days after vaccination

Animals are inspected for 8 days for FMD lesions in tongue and feet

Valid test: both control cattle have to show signs of FMDV in feet

A vaccine serial is satisfactory if Contains more than 6 PD50

■ Protected

■ Not protected

18.1 PD50

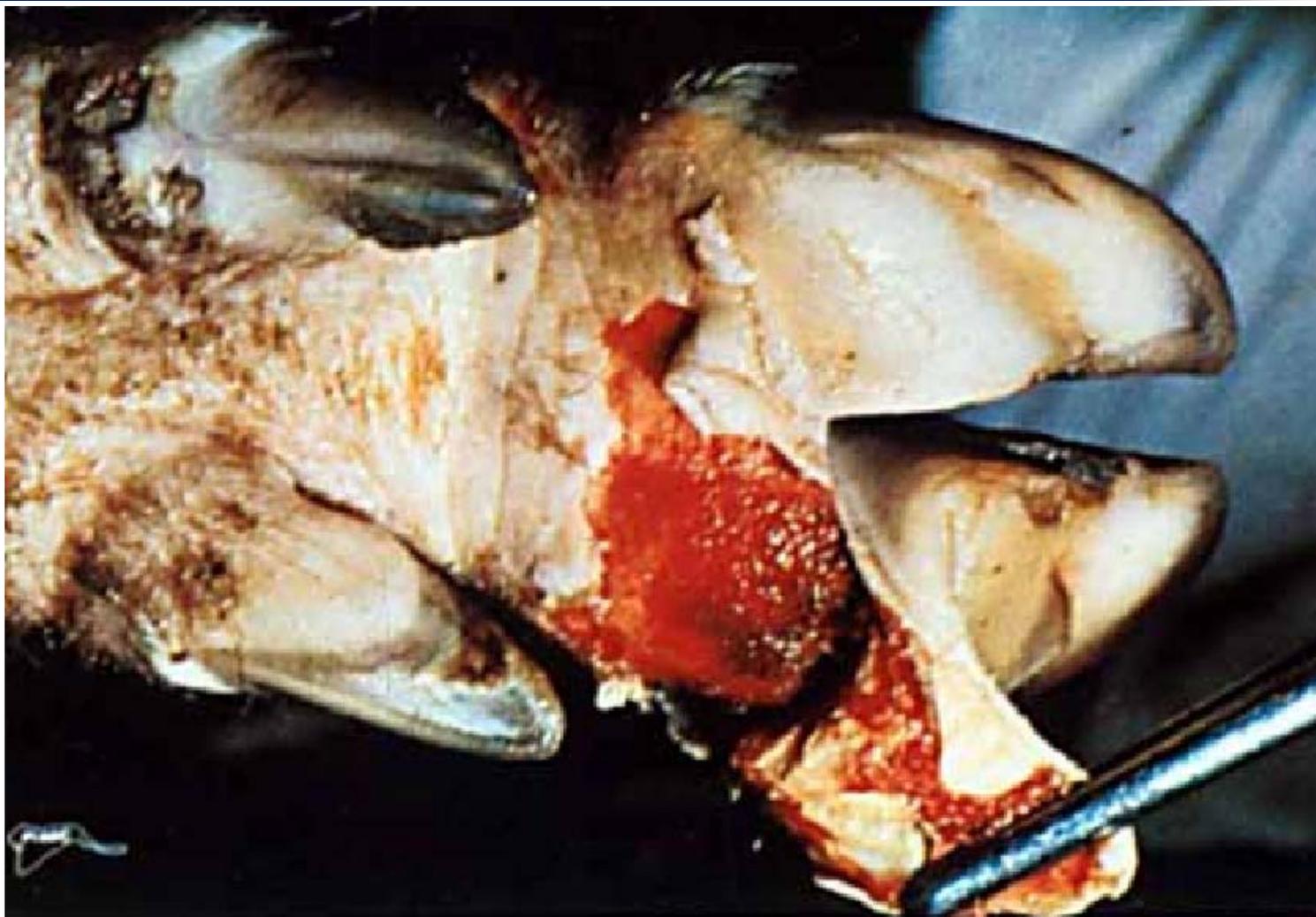
Safeguarding Animal Health

The Disease

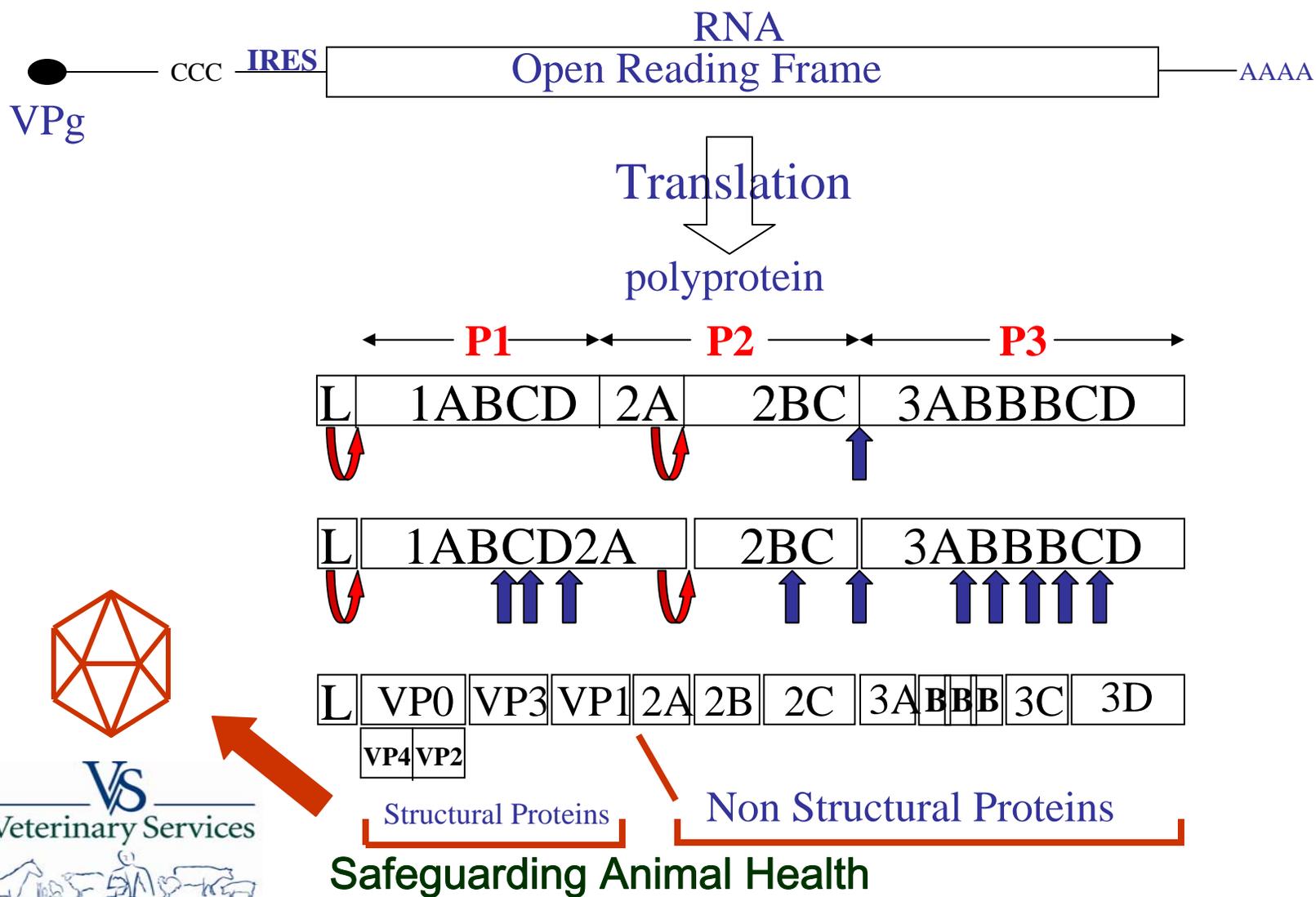
- The virus affects artiodactylae : cattle, swine, goats, sheep, buffalo.
- Lesions in mouth, feet, teats, etc.
- Viremia
- The virus can become persistent: Carrier state







Genome Organization



Samples for FMD:

Epithelium from lesions

Vesicular fluid

Oral swabs

Nasal swabs

Blood

Milk

OP fluid

Diagnostic Tests for Agent Identification

- RT real-time PCR
- Antigen ELISA
- Virus isolation followed by serological method for serotyping.
- Complement Fixation

RT real-time PCR

- Samples
 - Epithelium
 - Vesicular Fluid
 - OP fluid
 - Blood, Serum
 - Swabs (nasal, oral)
 - Milk

- Primers
 - 3' D pol
 - 5' UTR

Antigen ELISA

Substrate

Rb Anti-GP conjugate

GP detector antisera Serotype specific

Test Sample (Antigen)

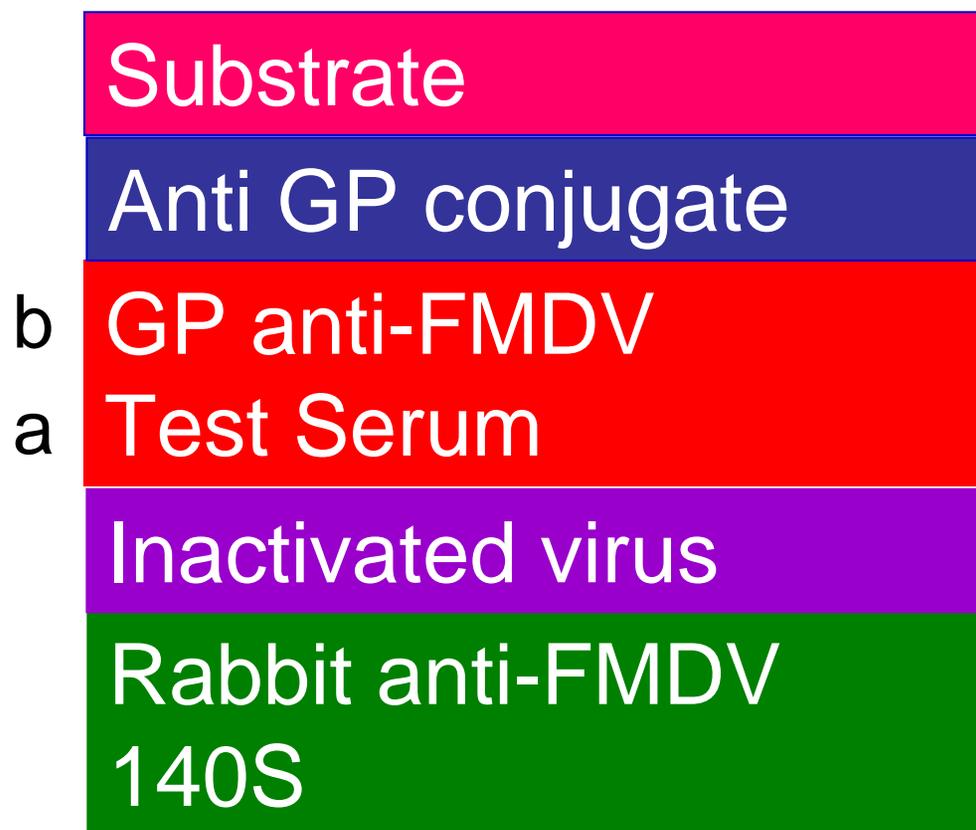
Rabbit Capture Antibody

Wells coated with Ab against all 7 Serotypes, VS and SVD

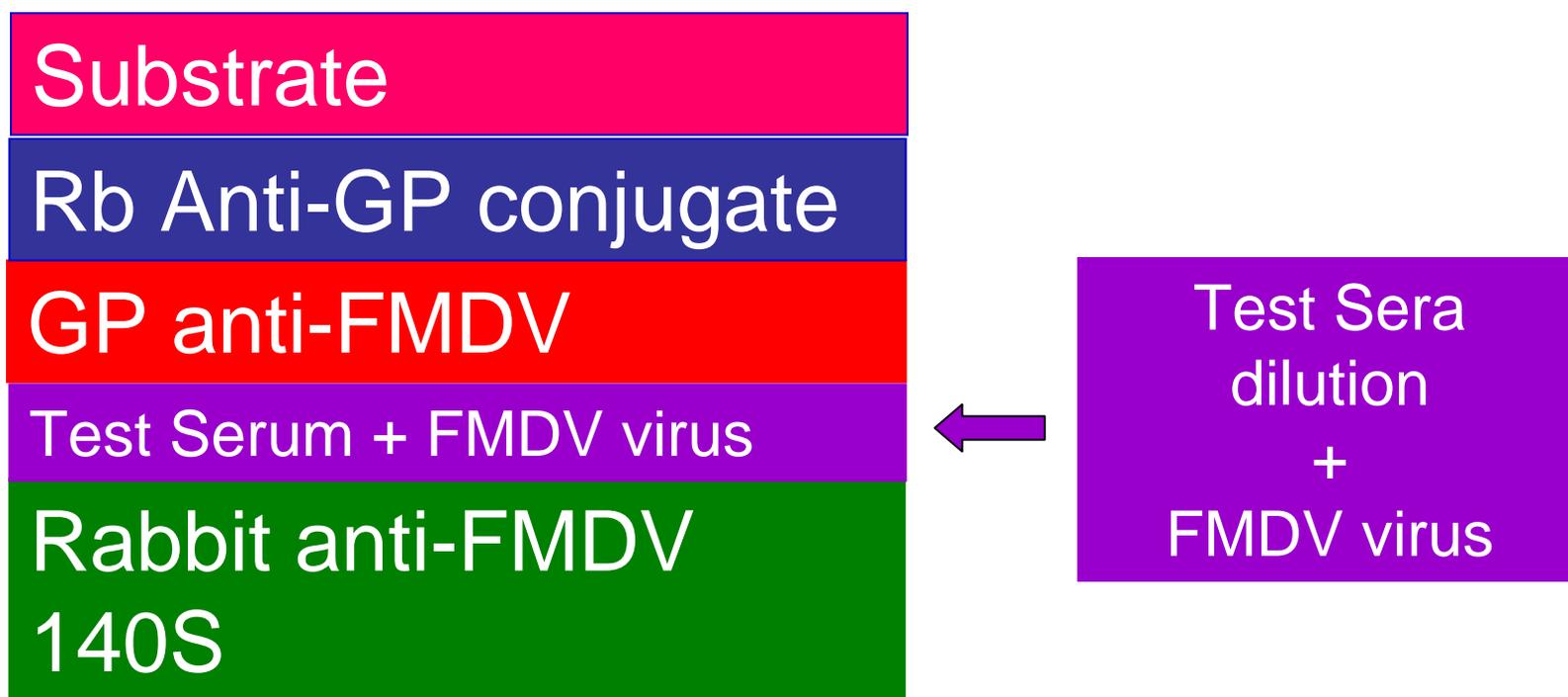
Tests for Antibodies to SP

- Solid phase competition ELISA
 - Surveillance for non vaccinated animals
- Liquid Phase Blocking ELISA
 - Surveillance for non vaccinated animals
 - Vaccine matching

Solid phase competition ELISA



Liquid Phase Blocking ELISA



Oesophago-pharyngeal OP fluid

- Detection of carriers
- Can also be used also for:
- Advanced or convalescent cases
- Suspected infection but absence of clinical signs

Testing of OP fluid

- Virus Isolation in cell culture followed by serological identification of the virus.
- RT real-time PCR.

Probang Cup



OP fluid – Site of Collection



Collection of OP fluid



Probang cup with OP fluid



Tests for Antibodies to NSP

- Indirect Enzyme linked Immunosorbent assay + EITB
 - Surveillance for vaccinated animals
- Competition NSP ELISA CEDI
 - Surveillance for vaccinated animals

Indirect Enzyme linked Immunosorbent assay

Substrate

Rb Anti-species IgG-HRP

Test Sera

Antigen 3ABC

Inconclusive results:

Vaccinated- confirmation by EITB

Non-Vaccinated – other NSP ELISA

Enzyme Linked Immunoelctro Transfer Blot EITB

Purified Recombinant NSPs
3A, 3B, 2C, 3D, and 3ABC

Separated in a PAGE gel
Transferred to Nitrocellulose

Enzyme Linked Immunoelctro Transfer Blot EITB

Substrate

Rb Anti-bovine - APh

Test Sera

Stripe with NSPs

Control sera: Positive
Weakly Positive
Cut off
Negative

Enzyme Linked Immunoelctro Transfer Blot EITB

Interpretation

Positive: if 3ABC, 3A, 3B, AND 3D, (+/-2C) stain equal or higher than positive control.

Negative: if two or more antigens stain below the control sera

Competition NSP ELISA CEDI

Substrate

MAb against 3ABC labelled

Test serum

3ABC

MAb against 3ABC

Ceditest

- Commercially available (Cedi Diagnostics B.V.)
- Detects antibodies to NSPs: 3ABC
- Blocking ELISA
- Good for all serotypes
- Good to detect antibodies to all susceptible animal species

Summary Table

- Surveillance
 - Detection of antibodies to structural proteins
 - Solid phase competition ELISA
 - Liquid phase blocking ELISA
 - Virus Neutralization
 - Detection of antibodies to non-structural proteins
 - Ceditest (3ABC)
 - Indirect Enzyme linked immunosorbent assay
 - EITB
 - AGID - VIAA
 - Oesophagopharyngeal fluid testing
 - Virus Isolation
 - RT real-time PCR

Thank you !