



Progressive Control Pathway (PCP)

2nd West Eurasia FMD Lab Network meeting

Advances in diagnostic tests

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Objectives and worplan

Develop “next generation” ready-to-use kits

For FMDV antigen detection / serotyping

- ✓ Simplified – sandwich ELISA
- ✓ Lateral flow devices (LFDs)



For antibody detection

- ✓ Ab to FMDV serotypes O, A, Asia 1 (SP antibody)
→ simplified – competitive ELISAs
- ✓ Ab to FMDV NSP (3ABC) = already available

Complexity of conventional ELISA format

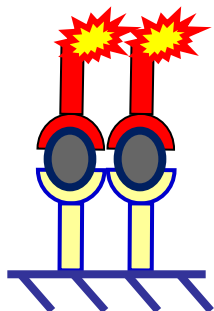
FMDV Antigen detection

- ✓ Serotype specific reagents for double-sandwich ELISA
 - Rabbit polyclonal capture antibody
 - Guinea pig polyclonal indicator antibody (+anti-GP IgG conjugate)
 - Control antigens (inactivated) supplied separately
 - **Four incubation steps**

FMDV Serotype-specific serology

- ✓ Serotype specific reagents for LPBE or SPCE
 - Rabbit polyclonal capture antibody
 - Guinea pig polyclonal indicator antibody (+anti-GP IgG conjugate)
 - FMDV antigens (inactivated)
 - **Four-five incubation steps**

New FMDV antigen detection/serotyping kit sandwich ELISA – Monoclonal Antibodies



DETECTOR MAb PO-conjugated

FMDV Antigen (positive sample)

CATCHING MAb (coated)

Catching Ab	Unique detector Ab pan-FMDV	FMDV specificity
type O	1F10	O
type A	1F10	A
type Asia 1	1F10	Asia 1
Type C	1F10	C
pan-FMDV	1F10	FMDV



- ✓ **FMDV control antigens already present in relevant wells**
- ✓ **All stabilized reagents included (conjugates, buffers, chromogen)**
- ✓ **Stable at 5°C (> one year)**

FMDV antigen detection/serotyping KIT

Evaluation on field samples

External evaluation (field samples)

- Kimron Veterinary Institute, Israel
- Indian Immunologicals Limited, India
- SAP Institute, Turkey

Demonstration/training

- Kenya
- Egypt

Supply (FAO, EUFMD programs - 2011)

- Afghanistan, Pakistan, Tajikistan
- Iran, Armenia, Azerbaijan, Georgia

LFD – Availability and Application

- ✓ FMDV Pan-specific test (based on pan-FMDV Mab 1F10)
- ✓ SAT2 specific test (based on a pan-SAT2 specific Mab)
- ✓ Asia 1 specific test (based on a pan-Asia1 specific Mab)
 - Validated on 160 epithelium samples at IAH
 - Available through EUFMD
- ✓ Other type specific tests
 - Under development

Serotype-specific serology

"Ready to use" ELISA kits

FMDV serotypes O, A, ASIA 1

State-of-the art

- ✓ **Prototypes kits developed and evaluated**
 - Kits for Ab to serotype O tested in some countries
 - Kits for types A & Asia 1 now ready for production
 - Shelf-life up to 2 years
- ✓ **Test run in only 2-2.5 hours (fast & easy)**
 - Plates provided pre-coated with FMDV inactivated antigens
 - Only two incubation steps (sera & conjugated Mab)
 - Incubations at Room Temperature
- ✓ **Current production capability: small-medium scale**

November 2010

Trainings

December 2008



Conclusions

- ✓ New antigen detection & serotyping ELISA kits for Eurasian FMD available
- ✓ Eurasian serotype-specific Ab ELISA kits validated, ready for production (**production scale?**)
- ✓ LFD for pan-FMD detection (O, A, C, Asia1) in use
- ✓ Asia1-specific LFDs launched

Gaps

- ✓ SAT-specific Ab/Ag ELISAs under development
- ✓ Other serotype-specific LFDs to follow (**do we need?**)

Discussion points

✓ Antigen detection & serotyping ELISA

- New ready-to-use kit:
 - Sensitivity similar to polyclonal ELISA
 - Better type-specificity
 - User friendly
- Pros: easier than RT-PCR, enables serotyping
- Limit: sensitivity (lower than RT-PCR)

✓ Serotype specific-Ab detection

- Limit:
 - Imperfect type-specificity
 - Different assays per each desired serotype

✓ Kits expiring date

- Check controls, usually kits can be used longer after expiration!!