Antigen detection ELISA kits

FMDV ANTIGEN DETECTION ELISA
and SEROTYPING OF FMDV O, A, C, ASIA 1, SAT 1 and SAT 2
5 plates/kit (10 samples/plate)

Antibody detection ELISA kits

SP Ab

SOLID-PHASE COMPETITIVE ELISA (SPCE) FOR ANTIBODIES SPECIFIC TO FMDV SEROTYPE O

SOLID-PHASE COMPETITIVE ELISA (SPCE) FOR ANTIBODIES SPECIFIC TO FMDV SEROTYPE A

SOLID-PHASE COMPETITIVE ELISA (SPCE) FOR ANTIBODIES SPECIFIC TO FMDV SEROTYPE Asia 1

SOLID-PHASE COMPETITIVE ELISA (SPCE) FOR ANTIBODIES SPECIFIC TO FMDV SEROTYPE SAT 2

SOLID-PHASE COMPETITIVE ELISA (SPCE) FOR ANTIBODIES SPECIFIC TO FMDV SEROTYPE SAT 1

Each SP-Ab ELISA kit is composed by 5 plates,
90 test sera/plate: single serum dilution
45 test sera/plate: two serum dilutions
22 test sera/plate: four serum dilutions

NSP Ab

FMDV 3ABC-TRAPPING INDIRECT ELISA
5 plates/kit (45 test sera/plate)

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Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna "Bruno Ubertini"

BRESCIA - Headquarter
Via Bianchi 7/9, 25124 Brescia (ITALY)
Tel. +39 030 2290.1 - Fax +39 030 2425251
http://www.izsler.it

Contacts :
santina.grazioli@izsler.it ⟶ for technical issues
estero.clienti@izsler.it ⟶ for commercial issues

I.Z.S.L.E.R.

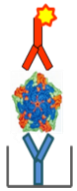
Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna Brescia, Italy

FMD DIAGNOSTIC KITS

developed in collaboration with The Pirbright Institute, UK

FMDV ANTIGEN DETECTION ELISA
Serotyping of
FMDV O, A, Asia 1, C, SAT 1 and SAT2

TEST PRINCIPLE



DETECTOR MAb PO-conjugated

FMDV Antigen (positive sample)

CATCHING MAb (coated)

The assay is a sandwich ELISA performed with selected combinations of anti-FMDV monoclonal antibodies (MAbs), used as coated and conjugated antibodies.

The test can be applied for detection and typing of FMD viruses in homogenates of epithelium vesicles and in vesicles fluid.

Only in these clinical samples the FMD virus usually achieves the concentration required to provide a positive signal in ELISA assays.

The kit is designed for detection and typing of FMD viruses of type O, A, Asia 1, C, SAT 1 and SAT2. A pan-FMDV test, detecting any isolate of type O, A, C and Asia1 and in addition some isolates of the SATs serotypes, is included in the kit to complement the specific typing and to detect FMD viruses which might escape binding to the selected type-specific MAb.

Microplates are supplied pre-coated with catching MAbs and with positive controls already incorporated onto plates.
Test samples (epithelium suspensions or vesicle fluids) are incubated with the coated MAbs. The FMD virus present in samples is captured by the homologous type-specific MAb and by an universal pan-FMDV MAb (these are the catching antibodies). After washing to remove unbound material **two detector conjugates** (**A** and **B**) are distributed in definite rows (as illustrated in the scheme below).
In case of positive samples, the conjugated MAb binds to the FMD virus trapped by the catching MAb, forming an Ag-Ab complex. After incubation, the unbound conjugate is removed by washing, and the TMB-chromogen solution is delivered into wells. A colorimetric reaction develops if the conjugate has bound to the sample antigen: the colour development is proportional to the amount of viral antigen present in the test sample. After addition of a stop solution, the optical density of the developed colour is read by a microplate photometer.
The test is fast - 2.5 hours approximately at room temperature - and simple - the precoated plates supplying the different type-specific reagents whilst the operator handles only two immunological reagents, corresponding to the detector conjugates.

Example of results and interpretation

Interpretation		POS O	POS Asia1	POS A	untyped	POS SAT2	NEG	POS SAT1	POS C	NEG	NEG	POS contr	NEG contr	
		Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6	Sample 7	Sample 8	Sample 9	Sample 10			
Catching Mabs		1	2	3	4	5	6	7	8	9	10	11	12	
Type O	A													Conjugate A
Type A (1st Mab)	B													
Type A (2nd Mab)	C													
Type Asia 1	D													
Type C	E													
Pan-O,A,C,Asia1	F													
Type SAT 2	G													Conjugate B
Type SAT 1	H													

positive sample reaction

negative or positive reaction

positive controls

ELISA kits are supplied with five ELISA plates.
Up to ten samples can be tested in each plate.

References

S. Grazioli, N.P. Ferris G. Dho G. Pezzoni, A. Morrison, V. Miulet, E. Brocchi. Development and validation of a simple serotyping ELISA based on monoclonal antibodies for the diagnosis of foot-and mouth disease virus serotypes O, A, C and Asia 1. TED 2020;00:1-11.DOI:10.1111/tbed.13677

S. Grazioli, N. Ferris, G. Dho, E. Spagnoli, E. Brocchi. Ready-to-use ELISA kit for FMDV diagnosis and serotyping tailored for Africa. Sess. Res. Gr. St. Tech. Committee of the Europ. Comm. Control FMD - Jerez, Spain, 29 Oct-31 Oct 2012.

SOLID-PHASE COMPETITIVE ELISA (SPCE)
FOR ANTIBODIES SPECIFIC TO
FMDV serotypes O, A, Asia 1, SAT 1 and SAT 2
N. 5 kits, one per each serotype

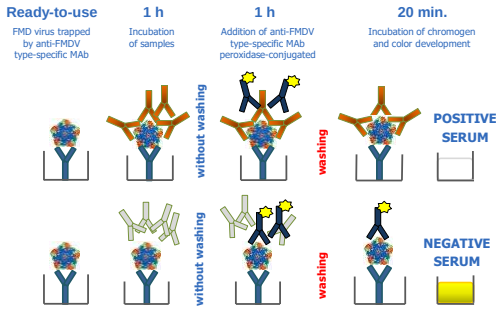
INTRODUCTION
Animals infected or vaccinated with Foot-and-Mouth Disease Viruses (FMDV) elicit antibodies against proteins of the viral capsids, also called structural proteins (SP). Anti-SP antibodies cannot distinguish between infected and vaccinated animals and are mostly serotype-specific. Therefore, different reagents must be used for each of the seven FMDV serotypes and specific assays may be required even for antigenically distant variants within a serotype.

Applications of SP-serology in FMD diagnosis include:
- Serosurveys during outbreaks/epidemic (in association with other diagnostic assays)
- Exit strategy to an FMD epidemic (to regain FMD free-status)
- Evaluation of vaccinal coverage (vaccination campaigns) and vaccine induced immunity
- Serological monitoring in countries at risk of FMD introduction (ex. buffer zones)
- Import/export, animal movement/trade (including exotic species, circus, zoo etc)

Serological assays commonly used for the detection of anti-SP antibodies are ELISA and Virus Neutralisation test. ELISA assays are easier, faster, suited for large scale and automation; in addition, they can make use of inactivated FMDV antigens and can consequently be adopted in a laboratory without high containment biosecurity status.

TEST PRINCIPLE
Each assay is a solid phase competitive ELISA (SPCE) based on a peroxidase-conjugated, neutralizing monoclonal antibody (MAb), specific for one FMDV serotype. Its binding to the homologous FMDV serotype immobilized onto the solid-phase is inhibited by antibodies against that serotype present in test sera. The test can be applied to measure antibodies in serum or plasma samples of FMDV infected or vaccinated animals of any susceptible species.
ELISA microplates are supplied pre-coated with inactivated FMDV antigens captured by homologous serotype-specific MAbs.
Appropriately diluted sera are incubated with the trapped antigen, enabling the specific antibodies present in the sample to bind to the antigen. Then, the anti-FMDV serotype-specific MAb, conjugated with peroxidase, is dispensed: its reaction with the homologous antigen will be inhibited by antibodies of positive sera previously bound to the virus, while in case of negative sera the conjugated MAb can bind to the FMDV antigen. After incubation, the unbound conjugate is removed by washing and the TMB substrate/chromogen solution is delivered into wells. A colorimetric reaction develops if the conjugated MAb has bound to the virus, i.e. if test serum is negative, while colour development is inhibited by positive sera. Antigen and conjugate concentrations are pre-calibrated in order to give a suitable reaction value (OD). After addition of a stop solution, the optical density of the developed colour is read by a microplate photometer.

The test is fast - 2.5 hours approximately at room temperature - and simple.



Each kit is supplied with five ELISA plates.

Serum samples can be screened at a single dilution 1/10, or in two dilutions for a semi-quantitative test (1/10, 1/30) or in four dilutions for titration (1/10, 1/30, 1/90, 1/270). Ninety or 45 or 22 test sera (with 1, 2 or 4 dilutions per each serum respectively) can be examined in one plate.

References:

Grazioli S, Brocchi E, Tranquillo V, Parida S, Paton D. Development of solid phase competitive ELISAs based on monoclonal antibodies for the serology of FMDV serotypes SAT1 and SAT2. The global control of IMD tools, ideas and ideals: Report. European Commission for the Control of Foot-and Mouth Disease Session of the Research Group of the Standing Technical Committee: 14-17 October 2008 Erice, Italy

*Anna B. Ludi, Alison Morris, Simon Gubbins, Amin Asfor, Madeeha Afzal, Clare F. Browning, Santina Grazioli, Efrem Alessandro Foglia, Ginette Wilsden, Alison Burman, Emiliana Brocchi, David J. Paton *, Donald P. King*
Cross-serotype reactivity of ELISAs used to detect antibodies to the structural proteins of foot-and-mouth disease virus
Viruses. 2022 Jul 8;14(7):1495. doi: 10.3390/v14071495.PMID: 35891476

FMDV 3ABC-TRAPPING ELISA
anti-NSP antibodies

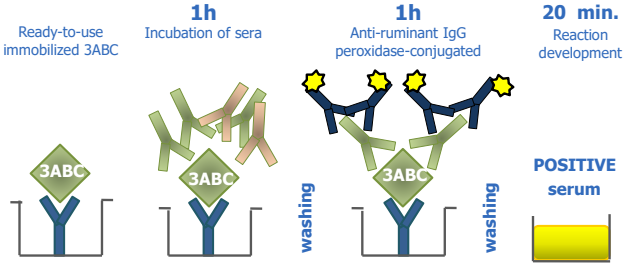
INTRODUCTION
The test is a trapping-indirect ELISA for the detection of antibodies to the non-structural polypeptide 3ABC of FMD virus in serum or plasma samples of **large and small ruminants.**
In Foot-and-Mouth Disease (FMD) investigations, the differentiation of infected from vaccinated animals is based on the identification of antibodies against the viral non structural proteins (NSP). FMDV vaccines consist of partially purified, inactivated virus particles: the vaccine purification eliminates NSP produced during virus replication. Therefore, animals vaccinated with purified vaccines will develop antibodies against viral structural proteins (SP) only, whilst animals infected elicit antibodies against both SP and NSP.

Several NSP are produced during viral replication. NSP are mostly common to the seven FMDV serotypes and, among NSP, the polypeptide 3ABC is indicated as one of the most immunogenic. The detection of anti-3ABC antibodies is indicative of a current or past infection with any of the seven FMDV serotypes, so the test reveals infected animals regardless their vaccination status.

TEST PRINCIPLE
The **FMDV 3ABC-TRAPPING ELISA KIT** uses an anti-3A specific monoclonal antibody (MAb) coated to the solid phase to trap the recombinant 3ABC polypeptide expressed in *E. coli*.

Microtitre plates are supplied pre-coated with the 3ABC antigen captured by the MAb.

Appropriately diluted test sera are incubated with the trapped antigen, enabling the specific antibodies present in the sample to bind to the 3ABC. After washing to remove unbound material, an anti-ruminant IgG, peroxidase-conjugated MAb is dispensed: the anti-ruminant IgG binds to the FMDV antibodies of the positive samples immune-complexed with 3ABC. After incubation, the unbound conjugate is removed by washing, and the TMB-chromogen substrate is delivered into wells. A colorimetric reaction develops if the conjugate has bound to the sample antibody: the colour development is proportional to the amount of antibodies present in the test sample. After addition of a stop solution, the optical density of the developed colour is read by a microplate



photometer.

Kits are supplied with five ELISA plates.

Each sample is dispensed in duplicate wells, one well of which contains antigen (3ABC) and one which does not (buffer). Forty-five samples plus three control sera can be examined in one ELISA plate.

Reference

Brocchi E, Bergmann IE, Dekker A, Paton DJ, Sammin DJ, Greiner M, Grazioli S, De Simone F, Yadin H, Haas B, Bulut N, Malirat V, Neitzert E, Goris N, Parida S, Sorensen K, De Clercq K. Comparative evaluation of six ELISAs for the detection of antibodies to the non-structural proteins of foot-and-mouth disease virus. Vaccine. 2006 Nov 17;24(47-48):6966-79