BIOVET®

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BIOVET.CA

MFIA KITS

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Summary

Photos : Manufacturing ELISA kits - © Patrick Roger, Photographer

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BIOVET®

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Better diagnostic, better medicine.™

About Biovet

We operate a CFIA-accredited laboratory, offering a full range of innovative diagnostic services for veterinarians. Our team is always attentive to market requirements and developments and is committed to providing quality products and services every day.

Biovet is also the only Canadian company to develop, manufacture and market a range of high-performance multiplex serology products, ELISA diagnostic kits and qPCR products for animals.

Biovet holds a CFIA establishment licence for veterinary biologics (Licence No: 49).

Key facts and figures

PARTNER
IN OVER
250
CLINICS

SOLD
IN NEARLY
50
COUNTRIES

AFRICA,
OCEANIA,
NORTH & SOUTH
AMERICA ,
ASIA,
EUROPE

NEARLY
100
DEDICATED EMPLOYEES

In October 2019, Antech Diagnostics, a subsidiary of Mars Petcare, acquired Biovet. It was a natural merger of two like-minded organizations with a shared commitment to veterinarians. Now, we have more resources to work with, so we can make our service offering even better for our 800 clients.

How does Xmap Technology Work?

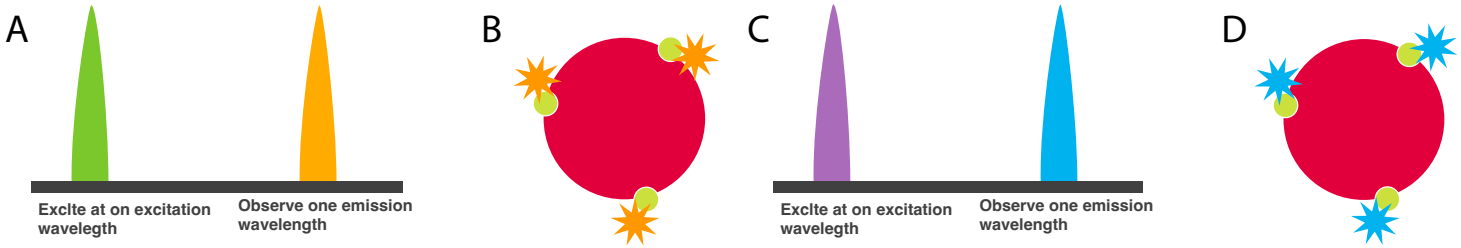
By **Combining advanced fluidics**, **optics** and **digital signal** processing with the proprietary microspheres (beads) technology, xMap Technology enable high multiplexing in **single sample volume**.

The **microsphere** used for MFIA assay are of 6.5µm diameter. They are coated with excitable dyes. When a beam of light strikes the bead, it emits a **signal specific to its color.** There are 500 different colored beads.

The **surface of the beads** is impregnated with iron containing magnetite particles. This feature allows the **use of magnets** to rapidly remove the beads from reaction suspension to speed up processing protocols.

Basic principle of the MFIA test

- **Multiple light sources** from the **Luminex** device excite the different dyes in the beads, enabling the device to identify each bead in the mixture and the **fluorescent reporter signal** captured during the assay.
- These unique features make it possible to **mix several antigens** and thus **detect several diseases in an animal** in the same assay. This **detect antibodies against several pathogens** and **reduces the volume of sample** required for testing.
- With this concept, **several different techniques** can be carried out:
    - ELISA-type immunoassays
    - nucleic acid assays (PCR)
    - enzyme activity assays
- 2 Kits produced and used by Biovet : MFIA PRRS NA, EU and Mycoplasma hyopneumoniae and Salmonella serotyping.



Description of one of the Biovet MFIA kits

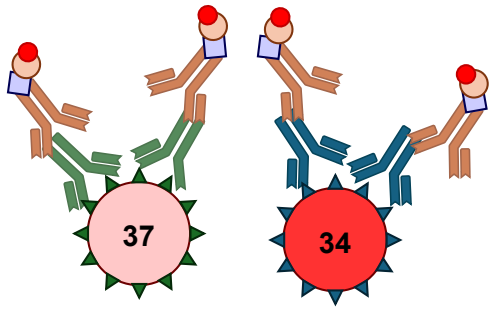
Kit Swinecheck MFIA PRRS NA,PRRS EU, SIV, PCV2, *M. hyo*
The kit contains the following components :

Component	Quantity
Sample Diluent	2 x 75 mL
PRRSV type 2 (NA) Positive Control (ready-to-use)	2 mL
PRRSV type 1 (EU) Positive Control (ready-to-use)	2 mL
Swine Influenza A Positive Control (ready-to-use)	2 mL
PCV2 Positive Control (ready-to-use)	2 mL
M. hyopneumoniae Positive Control (ready-to-use)	2 mL
Negative Control (ready-to-use)	3 mL
Microsphere Mix	28 mL
Wash Buffer (10X)	2 x 100 mL
Detection Antibody	55 mL
SA-PE Conjugate	55 mL
Microtiter Plate	5

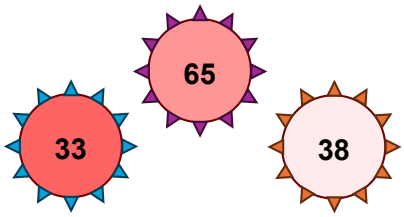
MFIA kits

MFIA assay developed BY BIOVET

The analyzer will read the SA-PE signal separately for each set (region) of microspheres.



- PRRSV type 1
- SIV



- PRRSV type 2
- *M. hyopneumoniae*
- PCV2

LEGEND

Microsphere Region 34 coated with PRRSV type 1 (EU) Ag

Microsphere Region 33 coated with PRRSV type 2 (NA) Ag

Microsphere Region 37 coated with SIV Influenza virus Ag

Microsphere Region 65 coated with Mycoplasma hyopneumoniae

Microsphere Region 38 coated with PCV2 Ag

Positive PRRSV1 antibody

Positive SIV antibody

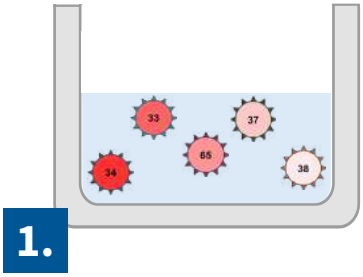
Negative antibody

Detection antibody

SA-PE Reporter

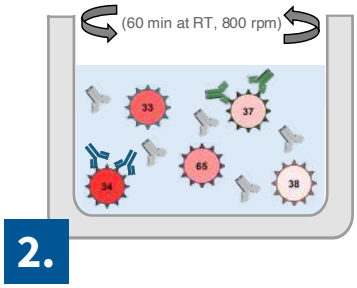
Magnet

MFIA assay developed BY BIOVET



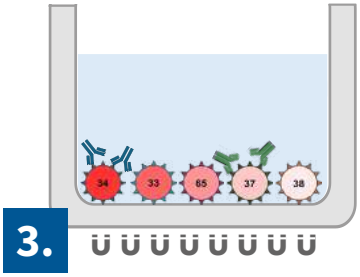
1.

Five distinct microsphere sets (Region) coated with different antigens are added to the wells.



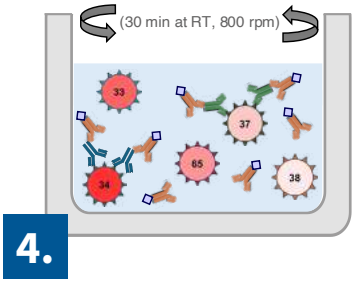
2.

Diluted serum samples are added to the wells and the plate is incubated under agitation.



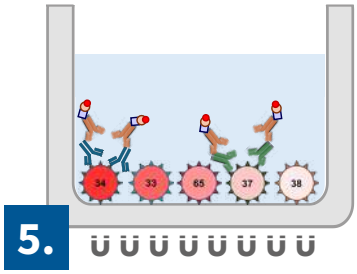
3.

After the incubation, the plate is moved on a magnetic separator and wells are washed.



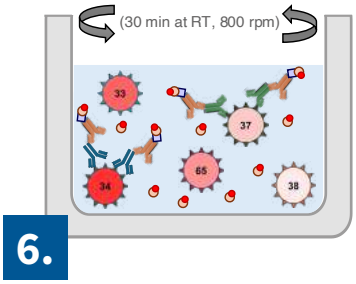
4.

The plate is then removed from the magnetic separator and the detection antibody is added, followed by an incubation.



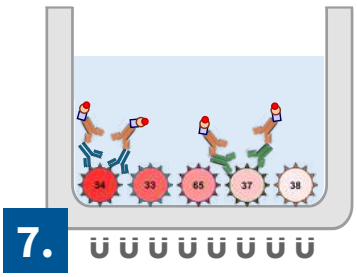
5.

After the incubation, the plate is moved on a magnetic separator and wells are washed.



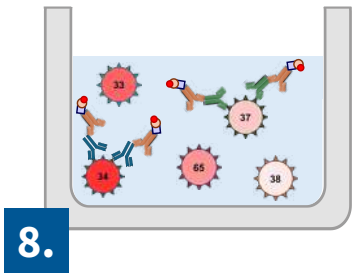
6.

The plate is then removed from the magnetic separator and the SAPE Reporter is added, followed by an incubation.



7.

After the incubation, the plate is moved on a magnetic separator and wells are washed.



8.

The plate is then removed from the magnetic separator and microspheres are resuspended in the wash buffer. The plate is then read by the analyzer.

MFIA assay

DIFFERENT READERS AVAILABLE

Reading time and the number of different beads each instruments can read vary.

1. Magpix, no longer sold, can read more than 50 different beads, and takes an hour to read a 96 well plate.
2. LX200 can read 100 different beads and takes 45 minutes to read a 96-well plate.
3. FlexMAP 3D and xMap Intelliflex can read 500 different beads and take 15 minutes to read a 96-well plate.

XMAP INTELLIFLEX SYSTEM

FLEXMAP 3D SYSTEM

LUMINEX 200 SYSTEM

MFIA assay

RESULTS INTERPRETATION

The analyzer will read the wells and give the Median Fluorescence Intensity (MFI) signal for each set of microsphere/antigen. Subtract the MFI value obtained for the negative control from those of the samples or the positive control to obtain the corrected MFI. If the negative control is run in duplicate, use the mean MFI for subtraction.

Calculate the S/P ratio for each set of microsphere/antigen as follow:

$$\text{S/P ratio} = \frac{\text{(Sample MFI - Negative Control MFI)}}{\text{(Positive Control MFI - Negative Control MFI)}} = \frac{\text{Sample corrected MFI}}{\text{Positive Control corrected MFI}}$$

The following cut off values were chosen:

Result	Negative	Positive
PRRSV type 2 (NA)*	S/P < 0.30	S/P ≥ 0.30
PRRSV type 1 (EU)*	S/P < 0.30	S/P ≥ 0.30
M. hyopneumoniae	S/P < 0.30	S/P ≥ 0.30
SIV	S/P < 0.15	S/P ≥ 0.15
PCV2	S/P < 0.30	S/P ≥ 0.30

* PRRSV type 1 and 2 antigens share common epitopes. PRRSV positive antibody samples usually cross-react with both antigens. The PRRSV type involved corresponds to the antigen with the highest S/P.

The following criteria must be met in order to validate the test :

- Each sample requires the analysis of at least 50 microspheres per antigen for valid data. Rerun any sample with a lower microsphere count for any antigen.
- The mean MFI signal for the Negative Control must be < 1 500 for PRRSV NA (type 2), PRRSV EU (type 1), SIV and M. hyopneumoniae, and < 2 500 for PCV2. Higher Negative Control signals can indicate a systematic error for the assay plate and require repeating the assay plate.
- The « corrected MFI » signal for the Positive Controls must be ≥ 5 000 for all controls. Lower Positive Control signals can indicate a systematic error for the assay plate and require repeating the assay plate.

MFIA assay

DIFFERENT READER AVAILABLE

1

REDUCES
the volume of sample required for testing various pathogens.

2

REDUCES
the quantity of consumables used for assays (tips, dilution plates).

3

REDUCES
the amount of antigen required for testing various pathogens.

4

OBTAIN
more results with **less technical time**.

5

GREATER
sensitivity than ELISA.

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