



Anti-Bovine Virus Diarrhoea Virus Monoclonal Antibody labelled with Fluorescein Isothiocyanate

BIO 316

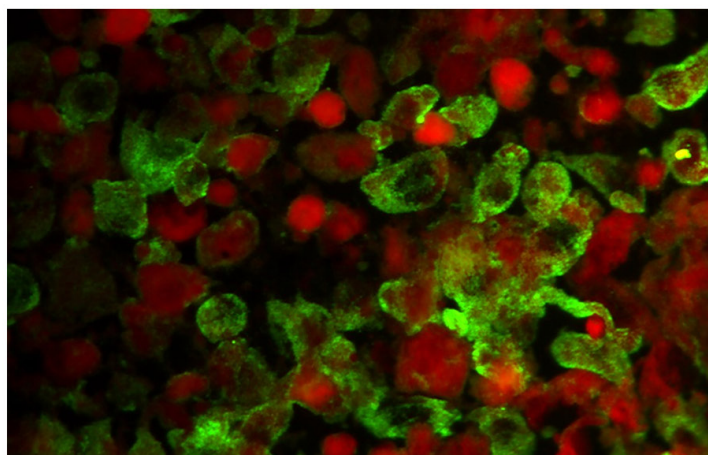
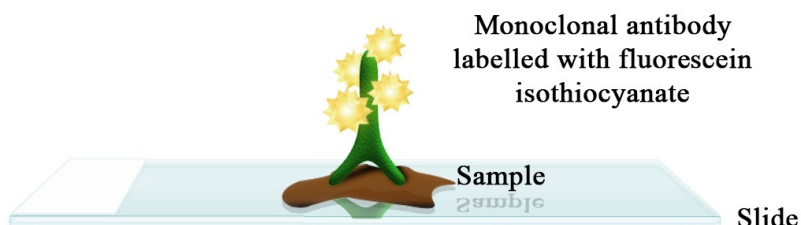
Reagent for direct immunofluorescence

REAGENT FOR DETECTION OF BVDV ON TISSUE SECTION OR CELL CULTURE

INTRODUCTION

BVD--bovine virus diarrhoea--and mucosal disease (MD) complex are two different clinical disorders caused by the same virus. BVD is the result of an acute infection in susceptible animals. Onset may occur at any time after birth. BVD has a brief course and low mortality. Mucosal disease, in contrast, is a deadly disease of low morbidity. It develops in viraemic animals that have been contaminated in utero. The characteristic of this in utero infection is the existence of specific immunotolerance that prevents the animals from producing antibody against the infective strain but not against another, antigenically different, BVD strain. These persistent carriers, which can live for years without developing clinical signs of the disease, can be detected by laboratory screening tests only. The direct immunofluorescence assay allows one to detect the presence of BVDV on frozen tissue sections made from fragments of respiratory organs (lungs), digestive organs (buccal or oesophageal mucosa, intestines), or lymphoid tissue (lymph nodes, spleen, Peyer's patches). The reagent can also be used to identify the virus's presence on an infected cell culture.

EXAMPLE OF RESULTS





Fix the cell preparation (cell cultures or tissue sections) for 15 minutes at room temperature with one of the following fixators :

- Paraformaldehyde 2 % in PBS
- Acetone solution (9 volumes of acetone and 1 volume of water).

Rince with PBS.

Dilute the conjugate twentyfold with a PBS-Evans blue solution made up according to the following formula:

PBS - Blue Evans

NaCl:	8 gr
KH ₂ PO ₄ :	0.2 gr
KCl:	0.2 gr
Na ₂ HPO ₄ . 2H ₂ O:	1.15 gr
Blue Evans:	0.01 gr
NaN ₃ :	0.1 gr
H ₂ O	1 L

Incubate the sample with the fluorescein-labelled conjugate for 1 hour at room temperature. At the end of this incubation period rinse the cell preparation with a PBS solution. Dry the cell preparation, then add the mounting medium prepared as follows:

Mounting medium

Glycerol	9 volumes
PBS	1 volume

Examine the cell preparation under a microscope equipped for detecting fluorescence.

COMPOSITION: One vial of 500 µl

STORING THE CONJUGATE: The conjugate must be stored at 4°C. It must never be frozen.

STABILITY: One year at 4°C

