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**ANTI-BOVINE BHV-4 MONOCLONAL ANTIBODY LABELLED WITH
FLUORESC EIN ISOTHIOCYANATE (BIO 286)**

(Reagent for direct immunofluorescence)

**REAGENT FOR DETECTION OF BOVINE HERPES VIRUS 4 (BHV-4) ON TISSUE SECTION
OR CELL CULTURE**

I - PROCEDURE

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

- Acetone solution (9 volumes of acetone and 1 volume of water).
- Acetone (1 V) + Ethanol (1 V)
- Ethanol
- Isopropanol

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.

II - STORING THE CONJUGATE

The conjugate must be stored at 4°C. It must never be frozen. It will keep at least 1 year at 4°C.