



Diagnostic and Analytical Services

Veterinary Laboratory Service

## COLLECTION OF SAMPLES FROM BULLS FOR CULTURE OF CAMPYLOBACTER FETUS AND TRITRICHOMONAS FOETUS

*Campylobacter fetus* subsp *venerealis* and *Trichomonas foetus* inhabit the mucosa of the glans penis, prepuce and the distal portion of the urethra. Therefore, preputial scrapings (smegma), preputial washings and semen samples are all suitable for culture.

Please let us know if you are testing for *Campylobacter*, *Trichomonas* or for both so we can send you the required media. Store media at 4<sup>0</sup>C before use. Discard unused media after 4 weeks.

### Sampling method A - wet method: Collection of preputial washings.\*

#### Equipment available from the laboratory:

1. Sterile screw-capped universal container each containing 20 ml of phosphate buffered saline pH7.2 (PBS).
2. *Campylobacter* enrichment transport medium (CETM).
3. *Trichomonas foetus* 'InPouch' test kit.

#### Equipment you need to prepare:

1. Sterile insemination pipette connected by plastic or rubber tube to a 20 ml sterile disposable syringe. If more than one bull is tested use new equipment or disinfect and rinse pipette/syringe between bulls.
2. A smaller syringe or pipette (1-2 ml) for inoculating the CETM and 'InPouch' test kit.

Introduce the pipette to the full length of the preputial cavity and hold the preputial orifice firmly closed with one hand around the pipette to prevent PBS from escaping. Inject 20 ml of PBS into the prepuce and wash/massage thoroughly with one hand, forcing the saline up and down along the penis several times. Withdraw the pipette close to the orifice where the washing fluid has accumulated and withdraw the fluid into the syringe. Transfer the washings to the universal container and stand to allow impurities to settle, then **inoculate the CETM with 1 ml of the supernatant** and the **'InPouch' trichomonas test kit with 0.5 ml of sedimented cells** from the bottom of the container.

\* This technique is better tolerated by bulls than the smegma collection method (Method B - dry method), consequently less physical restraint is required.

### Sampling method B - dry method: Collection of preputial scrapings.

#### Equipment available from the laboratory:

1. Sterile screw-capped universal container containing 20 ml of phosphate buffered saline pH7.2 (PBS). Use only 4 ml, see below.

2. Campylobacter enrichment transport medium (CETM).
3. *Trichomonas foetus* 'InPouch' test kit.

**Equipment you need to prepare:\*\***

1. Plastic rigid bevelled pipette. If more than one bull is tested use new equipment or disinfect and rinse pipette/syringe between bulls.
2. Rubber bulb.

Introduce the pipette to the full length of the preputial cavity. Squeezing and releasing the bulb, scrape along the dorsal surface of the penis and the surrounding preputial mucosa for half to one minute. Control the placement of the penis with a hand externally on the wall of the prepuce. Keep the opening in the bevelled end of the pipette directed downwards onto the penis. The angle of the 'handle' of the pipette should be downwards, away from the animal's body. Withdraw the pipette with the bulb compressed to avoid picking up too much debris from around the preputial opening. Flush out the scrapings with 4 mL of PBS into the universal container, stand to allow impurities to settle and **inoculate the CETM with 1 ml of the supernatant and the 'InPouch' trichomonas test kit with 0.5mL of sedimented cells** from the bottom of the container.

\*\* The laboratory recommends Method A and no longer supplies bulbs and pipettes for Method B. Pipettes should be approx 600 mm long and the scraping end should be cut obliquely approx 60°. The material (10 mm OD, nylon, high pressure plasticised tubing) is obtainable from Bearing Service Centre, 9 Barney St, North Parramata, ph: 02 9683 1355. The 60 mm diameter rubber bulbs are obtainable from Lab Supply, 260 Horsley Road, Milperra 2214, ph:02 9738 3838.

**Method C: Collection of semen.\*\*\***

**Equipment available from the laboratory:**

1. Campylobacter enrichment transport medium (CETM).
2. *Trichomonas foetus* 'InPouch' test kit.

Semen is collected by one of the standard techniques, using either artificial vagina, electro-ejaculator or massage of the accessory sex glands. The CETM is inoculated with 1 ml and 'InPouch' test kit with 0.5 ml of semen.

\*\*\*Culture of semen samples are not as reliable as culture of samples collected from the prepuce.

**Forwarding samples to the laboratory.**

The inoculated containers and test kits should be clearly marked. Information regarding the nature of the sample, animal identification and the time and date of collection should be included. Forward samples to the laboratory within two days.

The optimal temperature range for the test samples during transport is 18-37°C.

**It is important to avoid refrigeration and freezing or exposure of collected samples to temperature above 37°C.**