

Bovine Diagnostic Testing

Resource Booklet



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Bovine Viral Diarrhoea (BVD) Diagnosis and Control Clinically, there are three forms of disease due to Bovine Pestivirus infection:

- 1. A persistently infected (PI) form which may/may not have clinical signs
- 2. An acute transient form characterised by fever and diarrhoea and short term immunosuppression. These animals will mount an immune response and clear the virus in 10-14 days. Subsequently, they are Ab positive and virus negative.
- Mucosal disease (MD) only occurring in PI animals. PI animals are infected by a noncytopathogenic strain of the virus.

A subsequent spontaneous mutation of the virus to a cytopathogenic strain within the PI animal results in MD, characterised by sero-mucoid nasal secretions, severe erosive lesions in the oral and intestinal mucosa, diarrhoea and death.

A key element for persistence of infection in cattle is the ability of the virus to cross the placenta in naïve (non-immune) cows and infect the foetus. Infection of naïve pregnant cows has different outcomes depending upon the gestation period:

positive and virus

negative).

GESTATION PERIOD WHEN BVD INFECTS NAÏVE COW 120-150 days > 150 days < 40 days 40-120 days Foetal death. Foetal abortion or birth Foetal abortion or Birth of normal calf which may be small of persistently infected birth of calves with calves immunologically congenital defects, and weak, but has a tolerant to the virus especially of the competent immune (i.e. virus positive but CNS (e.g. cerebellar response (antibody

antibody negative).

EPIDEMIOLOGY

Infection is considered widespread in Australia. Persistently infected (PI) animals are considered the most important source of infection in a herd, as these animals excrete large amounts of virus continuously throughout their lives from nasal discharges, saliva, semen, urine, tears, and milk.

Disease often occurs when a susceptible animal is introduced into an infected herd or a PI animal is introduced into a susceptible herd. Conditions when pestivirus is likely to infect a herd

hypoplasia). Usually

antibody positive with

or without detectable

virus.

- Close contact between cattle, eg across fences
- Recent introduction of a PI animal or pregnant cow carrying a PI calf (Trojan calf) into a herd of susceptible pregnant females.
- Introduction of new cattle into a closed breeding herd.
- When the breeding herd has access to other cattle on the property.



IDENTIFYING AND DIAGNOSING PESTIVIRUS

Clinical signs of pestivirus can vary depending on the time of infection.

Clinical signs that would lead a producer to suspect pestivirus include:

- Early-term abortion or embryonic loss.
- Temporary infertility.
- Increased susceptibility to other diseases.
- Weak, stunted or deformed calves.
- Ill-thrift and wastage.

LABORATORY DIAGNOSIS FOR BVD

BVD virus can be detected in **blood**, milk and skin using either an antigen capture ELISA or RT-PCR (reverse transcription polymerase chain reaction).

An antibody ELISA is used to detect BVDV antibodies in serum and milk.

PCR is very sensitive and individual or pooled serum and milk samples can be tested.

1. Ab ELISA:

Gives low/medium/high titres.

A high titre result indicates recent infection in the last 6 months is likely whereas lower titres aren't related to a recent infection (eg recent abortion).

Can be pooled on serum or milk. Pooled serum or milk from the vat checks herd exposure for recent infection.

2. Ag ELISA:

Serum or whole blood on individual animals to identify a PI (PCR is more sensitive than Ag ELISA).

3. PCR: individual sample

Serum or whole blood on individual animals.

To detect a PI (more sensitive than the Ag ELISA test).

4. PCR: ear notch

(Live animal) or aborted foetus tissue. To detect a PI.

5. PCR: pooled samples

Pooled serum or whole blood. Up to 20 samples pooled in the lab (send individual samples into lab and we will pool them here). If a pool is PCR +ve then the individual serum/blood samples are tested to identify the PI individual.

6. PCR on milk

From vat: herds can be screened.

If PCR is negative, there is no PI animal present in herd.

If positive, then continue on to pooled serum or whole blood PCR sampling to identify the PI individual.

BVD DISEASE CONTROL

Defining the BVDV status of animals in a herd and identifying any PI animals are the first stages toward controlling or eradicating the disease. This is done using a combination of PCR, antigen and antibody ELISA tests.

To investigate a herd's BVDV status:

In lactating animals

a bulk milk sample can be collected from the vat and tested for antibody by ELISA. If the antibody result is high, a PI could be present and a milk sample should then be tested by PCR. If a milk sample is positive on PCR, stratify the herd by production and serum sample the poorest performing 10% of the herd first. Test for virus by PCR, eliminate any PI animals, then recheck another milk sample by PCR.

In non-lactating animals

- Ideally take serum samples from 15 animals (nine animals minimum) in each age group of cattle you wish to investigate, and test for antibody by ELISA to establish if BVDV is present in the herd (15 serum samples are pooled together in the laboratory and testing gives a 95% chance of finding a seropositive animal).
- Take serum samples from *all* cattle. Gribbles Veterinary can pool the submitted serum samples into batches of 20 to be tested by PCR. When a positive pool is found, the individual samples making up that pool will be tested in an antigen capture ELISA. This will identify both PI and transiently infected animals. Skin can also be tested.
- If all serum pools or milk sample from a herd test negative by PCR, the animals sampled can be considered clear of infected animals and a biosecurity/ vaccination program put in place.
- For surveillance, annual testing is recommended. The herd may be bled and tested by antibody ELISA (individual serum samples from 15 yearling animals) and a bulk milk antibody test on the lactating animals. Virus screening of all calves to be kept is also recommended.

 Biosecurity can be maintained by pretesting new introductions to the herd for virus, with possible vaccination for on-going protection. Quarantine a newly bought pregnant cow until she calves because her calf could be PI (Trojan PI). Then test the calf for virus, once born and before colostrum is given (colostrum will interfere with antibody and virus testing).

PREVENTION STRATEGIES FOR PESTIVIRUS

- Defining the pestivirus status of a herd by serological testing of herds.
- Identifying and culling persistently infected animals
- Only buying cattle from other uninfected properties.
- Purchasing cattle from properties with no history of trading, agistment or cattle turnover, compared to cattle trading properties or where agistment is run.
- Keeping newly purchased cattle away from the breeding herd, especially if in early pregnancy.
- Ensuring replacement females have developed a strong immunity before joining.
- Vaccinating to control the disease in cattle.



Mastitis is an inflammatory change of the mammary gland and can be caused by physical or chemical agents, but the majority of cases are usually a result of bacterial infection. Over 100 different microorganisms have been isolated from mastitis samples, but *Staphylococcus aureus*, *Streptococci*, *Mycoplasma bovis* and members of the *Enterobacteriaceae* are among the more common organisms responsible. Mastitis is also characterised by an increase in somatic cells, and by pathological changes in the mammary tissue.

In order to accurately diagnose mastitis, it is vital that the milk sample is collected to ensure that the potential pathogen in the sample comes from the inside of the mammary gland and not from contamination on the outside of the udder. It is also essential the sample be taken prior to administration of antimicrobials (systemic or intramammary).

Most bacteria causing mastitis are considered either contagious or environmental. Contagious pathogens are found within infected udders and are spread most commonly at milking time (splashes, milking machine and other fomites).

GRIBBLES PCR PATHOPROOF™ MAJOR 4 IDENTIFIES:

- Staphylococcus aureus
- Streptococcus agalactiae
- Mycoplasma bovis
- Streptococcus uberis

GRIBBLES PCR PATHOPROOF™ COMPLETE16 IDENTIFIES:

- Mycoplasma bovis
- Mycoplasma spp.
- Staphylococcus aureus
- Streptococcus agalactiae
- Streptococcus uberis
- Streptococcus dysgalactiae
- Escherichia coli
- Enterococcus spp. (including E. faecalis and E. faecium)
- Staphylococcus spp. (including all major coagulase-negative Staphylococci)
- Klebsiella oxytoca (and/or K. pneumonia)
- Serratia marcescens
- Corynebacterium bovis
- Trueperella pyogenes and/or Peptoniphilus indolicus
- Staphylococcal ß lactamase gene (penicillin- resistance gene)
- Yeast
- Prototheca spp.

Some of the isolates from bovine mastitis and their natural habitats are listed below:

MICROORGANISM	NATURAL HABITAT
Staphylococcus aureus	Udder lesions, mucous membranes (contagious)
Streptococcus agalactiae	Intra-mammary (contagious)
S. dysgalactiae	Environmental and teat lesions
S. uberis Mycoplasma bovis	Environmental predominantly Contagious
Enterococcus faecalis	Faeces and skin
Escherichia coli	Faeces, bedding (generally in housed cattle)
Klebsiella pneumoniae	Faeces, bedding (generally in housed cattle)
Enterobacter aerogenes	Faeces, bedding (generally in housed cattle)
Serratia marcescens	Faeces and soil
Trueperella (Arcanobacterium) pyogenes	Skin and mucous membranes
Nocardia asteroides	Soil
Pseudomonas aeruginosa	Water, soil or faeces
Bacillus cereus	Grain feed, intramammary preparations
Prototheca zopfii	Mud, soil, faeces, water



Abortion in any species can be due to infectious or noninfectious causes.

Samples to collect:

FOETAL

- fresh cotyledon (for fungal culture)
- fresh brain, lung or spleen for PCR
- fresh stomach contents or lung for culture
- fixed brain, lung, heart, and cotyledon (if available) for histopathology.

MATERNAL

Serum – affected cow and five previously aborted cows or herd mates if insufficient aborted animals. Used for serology if indicated eg BVD antibody testing

Unless specific testing instructions are received, fresh material will be placed on hold pending the results of histology.

COMPLETE BOVINE ABORTION PANEL SPECIMENS

Cow serum (BVD AGID), fresh fetal stomach contents/lung (bacteriology); fresh foetal heart and lung (BVD, Neospora and Lepto PCR); fixed fetal brain, heart, lung, cotyledon (histopathology).

NEOSPORA

- Fixed foetal brain (even if mushy), heart, lung, +/- skeletal muscle
- Fresh foetal brain tissue for PCR (brain tissue is the most sensitive for Neospora PCR diagnosis)
- Abortion is most common in the second trimester.

BVD

- Fresh foetal lung/spleen tissue for PCR
- Fresh serum from cow (for Ab AGID)
- Fixed foetal tissues for histopathology to rule out other causes
- The pathology of BVD in the foetus is complex (see BVD chapter).

LEPTOSPIROSIS

- Fresh foetal lung/spleen tissue for PCR
- Fixed foetal brain, heart, lung and placenta
- Check if the herd is vaccinated. Aborting cows are often (though not always) sick.

MYCOTIC ABORTION

- Stomach contents or spleen and fresh placenta (only if not contaminated)
- Fixed placenta and foetal tissues
- Usually sporadic abortions
- Aspergillus, Mucor, Absidia, Rhizopus.

OTHER BACTERIAL CAUSES OF ABORTION (USUALLY A SPORADIC CAUSE)

- Fresh stomach contents or lung for culture
- Fixed fetal brain, heart, lung and placenta
- Common bacteria include Listeria monocytogenes, Trueperella (Arcanobacterium) pyogenes, Ureaplasma diversum, Salmonella
- NB. Tritrichomonas foetus and Campylobacter foetus venerealis infections can cause venereal disease and infertility but occasionally cause abortion. We have a PCR test for both.

NONINFECTIOUS CAUSES OF ABORTION

- Maternal pyrexia causing fetal heat stress can lead to abortion.
- *Macrocarpa* and pine needle poisoning can cause late term abortions
- *Theileria* leading to cow anemia and fetal anoxia (NB. transplacental transmission of *Theileria* is low, ie in transplacental infection of Theileria is not the cause of abortion).

O24 Trace Elemen

Trace Element Testing in Cattle

Trace element deficiences can cause or exacerbate disease and ill thrift.

Trace element profile for copper, Vit B12 (Cobalt) and GSHPx: requires Gel and EDTA tubes.

COPPER (Cu)

Copper <u>Deficiency</u> Specimens: serum (N = 10), or liver (N = 5)

Copper <u>Toxicity</u> Specimens: serum (live animal) and kidney (dead animal).

Copper (Cu) is stored in the liver so serum copper levels don't give an estimation of the liver reserves until the reserves are very low. But, if more than two serum coppers are low in a group then the liver copper levels of all animals in the group will also be low.

General Information on Cu

As an animal becomes copper depleted, liver copper is the first to fall. When this is severely depleted, then serum copper falls and eventually copper at essential sites becomes low. Clinical syndromes associated with Cu deficiency occur when the enzymes needing Cu are affected. Low serum copper means that the liver Cu stores are low and Cu deficiency could be the cause of the clinical signs.

Serum is a poor indicator of liver stores and is not the recommended sample when information on storage levels is required.

Avoid sampling sick animals (eg mastitis, facial eczema), since serum copper increases in inflammatory conditions.

Copper deficiency signs include ataxia, osteoporosis, growth rate depression, infertility, and hypopigmentation of hair. Zinc, at concentrations used to prevent sporidesmin toxicity, can impact on the copper status of cattle (one of the prophylactic effects of zinc may be to reduce copper availability).

Copper availability in pasture is affected by the sulphur, iron, molybdenum and zinc concentrations in pasture and soil.

SELENIUM (Se)

Specimens: serum, whole blood (EDTA) or liver (n = 5)

Number of animals to test for mob = 5 (liver, serum or whole blood)

Serum selenium is very stable as is whole blood selenium.

Liver or whole blood can be used to diagnose selenium toxicity.

Serum Se – measures current intake and approximates liver concentration.

Whole blood selenium and glutathione peroxidase (GPx) – these two tests correlate after steady state reached 3 months after dosing. GSHPx can be used to assess selenium status of stock if they have been grazing the same soil type and no selenium supplementation for three months.

Liver selenium - same as serum selenium; it measures current intake (50 mg liver needed).

General Information on Se

Selenium absorbed from the diet or supplements gets translocated to serum and liver within hours. There is no storage organ for selenium. Serum and liver are good indicators of the current selenium status of the animal (within the last month).

Glutathione peroxidase (GPx), changes more slowly with changing intake, because it is incorporated into red cells during haematopoiesis. Therefore GPx predominantly reflects the selenium intake 3 months previous.

GPx gives a reliable indication of current selenium status for stock that have been grazing the same pasture if no selenium has been supplemented in this period, either directly or through the fertiliser. Whole blood selenium measures both the selenium level in serum and in the RBCs, therefore lies between serum selenium and GPx levels in its responsiveness to changes in selenium intake.

COBALT/VITAMIN B12

Specimens: serum (n=10) or liver (n=5)

Optimum number of animals to test for deficiency = 8 (serum) and 4 (liver).

Cobalt is measured indirectly in animals by measuring vitamin B12 a cobalt containing vitamin.

General Information on Co/B12

Order of susceptibility to cobalt deficiency (high to low) is lambs>adult sheep>calves> kids >fawns>adult goats> cattle>deer.

Sample during the most susceptible time of year.

In marginal areas where the incidence of cobalt deficiency varies from year to year it is recommended to check vitamin B_{12} status at weaning time. Liver is the storage organ for vitamin B_{12} but it also correlates well with treatment responses. Concentrations are falsely increased in animals with liver disease (eg. facial eczema). Serum concentrations in cobalt deficient animals rise within 24-48 hours of an increase in cobalt intake.

IODINE

Specimen: serum (N = 3-5)

Inorganic iodine is a very stable compound.

Inorganic iodine measures the iodine intake of the animal over the previous 2-3 days and the effect of oral or parenteral treatment with iodine supplements.

Serum thyroxine (T4) is <u>not</u> a useful test to detect iodine deficiency in farm animals.



ZINC (Zn)

Specimens: Serum (n =10) or liver (n = 4) Serum preferred.

There are no significant mobilisable stores of zinc in the body.

Serum Zn levels reflect dietary levels.

High levels of calcium, soil and sulphur in the diet can reduce zinc availability to the animal.

Do not test sick animals or cows within two weeks of calving as these processes depress serum zinc concentrations.

REFERENCES FOR TRACE ELEMENTS

1. Further in depth discussion of trace element deficiencies including soil types and areas of Victoria affected are available at:

vro.depi.vic.gov.au/dpi/vro/vrosite.nsf/ pages/trace_elements_pastures

2. Optigrow Trace Element Monitoring Manual. Anonymous, 1997 MAF Quality Management

3. Trace element disorders in SA

www.pir.sa.gov.au/__data/assets/ pdf_file/0011/49619/Trace_Element_ disorders_in_SA.pdf

4. Hosking WJ, Caple IW, Halpin CG, Brown AJ, Paynter DI, Conley DN, North-Coombes PL. Trace Elements for Pastures in Victoria; for the Trace Element Review Committee of the State Dept of Agriculture and Rural Affairs, Victoria, 1986.



INDIVIDUAL FECs:

Faecal Egg Counts (FECs) are a means of estimating the worm burden of an individual or population. Individual FECs allow subjective assessment of the worm burden of an animal, to confirm a diagnosis of parasitism with nematodes or coccidia, or to assess efficacy of recent treatment. Approximately 4-5 grams for cattle is a minimum sample – approx 2 tablespoons.

Collection should be made directly from the rectum by gloved finger, or from freshly passed faeces in a small clean holding yard, since eggs can hatch or desiccate quickly after deposition on pasture.

For herd investigations, ten individual animal samples are recommended. These should reach the lab within 2 days of collection. Otherwise, they should be stored at 4°C to prevent eggs hatching. Individual samples are most suitable for animals that are unwell or in poor body condition. They are more accurate in younger animals than in older mature animals that may have immunity to nematodes and thus suppress their egg release.

FEC interpretation is subjective and best considered in light of the clinical presentation and management system. Parasite factors e.g. species, life cycle stage; host factors e.g. age, immunity, gastrointestinal throughput rate and food intake; environmental factors e.g. season must all be taken into consideration to estimate the gastrointestinal nematode burden and its likely significance in each host. FECs can be artificially reduced in animals with diarrhoea (volume/ dilution effect), or with delay in sample processing, and artefactually increased by dehydration and gut stasis, or collection of faeces from the gut post-mortem; these can allow accumulation of eggs in a small area.

BULK FECs:

Bulk faecal samples are useful to get an overview of the gastrointestinal nematode parasite status of the flock. Bulk sampling averages ten individual FECs meaning high and low individual FECs don't overly influence results. It is best to do one bulk test per group of animals, e.g. young stock, females and males, or per paddock if animals are grazed separately.

Please specify "Bulk FEC" on the submission form: if you submit 10 individual samples without specifying further, you will be charged for and provided with 10 individual FECs. Up to 10 animals may be submitted per group; more than this will be divided for greater accuracy into two or more bulk FECs. Pellets should ideally be taken from the animal's rectum, or immediately after defaecation from the ground. (If faeces are collected randomly from the pasture, it is likely that some will be older and therefore eggs may have hatched, artificially reducing the egg count. The number sampled in this way may not be as accurate.). Where a drench history is known, it is possible to evaluate the efficacy of the treatment programme. The drench resistance test is useful when a particular product is being investigated.

If the anthelmintic drug used is one without ongoing effect i.e. after treatment ingested larvae are able to mature immediately, the minimum period before a significant FEC would be seen is 3 weeks, and this only in conditions of heavy pasture contamination.

Where longer-acting modern drugs are used there should not be faecal egg output for many more weeks, depending on product, and any significant number of eggs seen in the faeces of the group may indicate a problem with administration of, or resistance to, the drug. Resistance is indicated if the mean egg count at resampling, relative to the initial sampling is not at least 90% reduced.

Many problems of drench resistance are due to insufficient drug volume being administered, which is ineffective and leads to development of resistance, so check that the heaviest animals in each group are the ones used to calculate the dose of drug given to all animals. Pasture management also plays a large role in good worm management.

DRENCH RESISTANCE TESTING -STRONGYLE PCR TESTING

Strongyle PCR testing has replaced the older method of larval culture and is cheaper and faster.

An FEC is done and a PCR to determine which Strongyle species have survived.

The test includes a faecal PCR test combined with a FEC.

1. The FEC reports the total parasite burden quantitatively (Strongyle and *Nematodirus* eggs per gram) The PCR reports the total Strongyle burden semi-quantitatively (0-5+) to species level, plus the relative abundance (%) of each.

Strongyle species: Haemonchus spp, Ostertagia ostertagi, Trichostrongylus spp, Cooperia oncophora, Oesophagostomum radiatum, Bunostomum spp and pannematode are included in the PCR.

(Note: the pan-nematode assessment reports nematode antigens other than the species listed. A larval culture would be required to identify what these are.)

TOTAL WORM COUNTS

These are the most accurate assessment of the total worm burden of an individual and are less affected by dehydration, diarrhoea, variation in worm fecundity and host immunity and suppression of egg production.

The animal must be sacrificed for this test.

Collect the entire contents of the abomasum in a bucket making sure the abomasal surface is thoroughly washed. Wash the contents of the first half of the small intestine into another container. If large intestinal counts are required, wash the entire contents of the caecum and colon into a third container. If volumes allow, submit all the gut contents in three separate leak-proof containers (e.g. Agee jars). If volumes are too large, thoroughly mix the contents and take a measured aliquot (e.g. 10%) and specify this on the submission form.

The results will be calculated as a percentage to allow interpretation of the

individual worm burden;

Worms are also identified to genus or species level as appropriate to allow planned treatment.

SERUM PEPSINOGEN

A high pepsinogen level in groups of calves generally indicates a heavy *Ostertagia sp.* worm burden (e.g. above 30,000). Interpretation of a high level in a single animal needs to be done with caution, as there are other causes of increased levels such as abomasal ulceration and reduced renal perfusion. Pepsinogen concentrations remain high for a long period after abomasal damage (over 60 days following a single challenge).

Pepsinogen concentrations also tend to increase with age from 1 U/L in calves less than six months to 1-3 U/L in older stock. This occurs irrespective of infection. In cattle greater than 2 years of age, pepsinogen levels will usually be approaching or above the top end of the normal range of 2.6 U/L. Because of this non-specific increase, the test has limited value in cattle older than 2 years of age.

Pepsinogen is excreted in urine, which means that levels in serum increase as renal perfusion decreases. A high pepsinogen can therefore be misleading in an individual sick animal. Serum pepsinogen is of limited value in sheep, goats and deer because severe parasitism usually involves a much larger part of the gastrointestinal tract.

LUNGWORM

The diagnostic tests for lungworm include examination of faeces for larvae or examination of tracheal wash fluid. In a group of animals where lungworm is suspected, it is recommended to check at least 5 individual faecal samples for lungworm larvae. Approximately 30-50 g of faeces is required from each cow. If these are all negative, then tracheal washes can be done before this infection can be ruled out. Tracheal washes are likely to be more sensitive than faecal examinations.

LIVER FLUKE

The diagnostic tests for liver fluke (*Fasciola hepatica*) include examination of the faeces for eggs, detecting antibody in serum and bulk tank milk and post-mortem or slaughterhouse examination of bile ducts.

Fasciola hepatica is a poor egg layer, which means that the sensitivity of a faecal test in an individual infected animal is about 65%. This test will also miss prepatent infections. It is recommended that the faeces from at least 5 animals be examined for fluke eggs. 10 g is required for export samples; 4-5 g is usually adequate for other samples if more cannot be gathered.

Milk: submit individual milk samples. We will pool them into groups of 5 in the lab prior to ELISA testing.



NEONATAL DIARRHOEA

Neonatal diarrhoea is often a multifactorial problem involving colostrum, nutrition, husbandry, climate and infectious agents. The age of the animal is important. In many instances, the infectious agents suspected can be ruled out based on this knowledge.

SAMPLE COLLECTION

1. Faeces or colonic content

2. Fixed small and large intestine from fresh intestine (freshly dead animal).

Many of the causative infectious agents are transiently present, or produce villous atrophy that is obscured by autolysis; therefore, remember that intestinal mucosa commences autolysis within 15 minutes of death, hindering diagnosis. If fresh samples cannot be obtained, think carefully before requesting histopathology interpretation. For histology, take multiple 1 cm long segments and immerse in abundant formalin.

Bovine diarrhoea: there are 5 panels for different ages of cattle. Panels 1-4 are for calves and panel 5 is for adult cattle.

1	Faecal Panel 1. (1-2 day old calf)	Faecal culture, Salmonella, E.coli K99
2	Faecal Panel 2. (2 day - 3 week old calf)	Faecal culture, Coronavirus, Rotavirus, Cryptosporidium, Salmonella, E.Coli K99
3	Faecal Panel 3. (3 week old to pasture calf)	Faecal culture, Coronavirus, Rotavirus, Cryptosporidium, Salmonella, Coccidia, FEC
4	Faecal Panel 4. (Calf on pasture)	Faecal culture, Coronavirus, Rotavirus, Cryptosporidium, Salmonella, Yersinia, Coccidia, FEC
5	Faecal Panel 5. (Adult)	Salmonella, Yersinia

SPECIFIC INFORMATION FOR SOME AGENTS

1. *Escherichia coli* – Apart from histology and culture, Enterotoxigenic E. coli in calves can be diagnosed by antigen k99 detection with ELISA.

2. Yersinia sp. is not included in the calf scour panels; therefore, consider requesting it in addition to the calf scour panels above; in calves that are getting close to weaning.

3. *Cryptosporidium:* Pathogenic infections with Cryptosporidium sp. are uncommon in calves > 3 weeks old; therefore, we routinely do not recommend testing for Cryptosporidium sp. in calves > 3 weeks.

OTHER TESTS

GGT – Levels of GGT in neonatal calves < 5 days old are directly proportional to the quantity of colostrum absorbed.

We recommend taking **5-10 sera samples** from affected and in-contact calves. The half-life of GGT in serum is relatively short and so this test can only be interpreted in calves less than 5 days of age.



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06/04/2016	DOB: 01/05/195	1
TEST, LYMPHOCYTOS	IIS	
HAEMATOLOGY GENERA	<u>د ۵</u> ک	
06/04/2016	DOB: 01/02/197	3
TEST, RASHID		
PROSTATE SPECIFIC AG.	<u>0</u> >	
05/04/2016	DOB: 01/09/197	3
TEST, MONOCYTES		
HAEMATOLOGY GENERA	د <u>۵</u> ک	
05/04/2016	DOB: 01/02/195	3
TEST, RASHID		
PROSTATE SPECIFIC AG.	<u>0</u> >	
04/04/2016	DOB: 01/01/197	3

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