A guide to diagnostic sampling - blood smears

This guide on veterinary blood smears for diagnostic investigations is a key part of haematological testing. It will help you to perform blood smears for laboratory submission as well as helping you examine cell morphology and blood borne parasites.

Remember to include a detailed history when completing the DPIRD Diagnostic Laboratory Services submission form. If you suspect an exotic, reportable or zoonotic disease, contact our Diagnostic Laboratory Services sample receival or the duty pathologist prior to submitting samples on 0(8) 9368 3351 or DDLS@dpird.wa.gov.au.

Perform blood smears when you suspect:

- blood-borne protozoal or mycoplasmal conditions
- · haemolytic conditions
- anaemia
- · haematologic neoplasia.

Equipment required for performing blood smears:

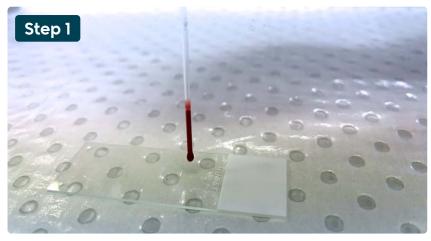
- · microscope slides with frosted ends for writing on
- spreader slide use a specific spreader slide with bevelled edges, or a second clean microscope slide
- capillary (microhaematocrit) tubes
- · pencil to label the slides
- blood collected in an EDTA tube
- pot of sterile water to clean the spreader slide between uses.

Top tips for making good smears:

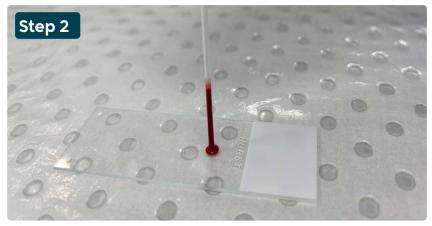
Making great blood films takes practice; don't be discouraged by less than perfect smears. Submit all blood smears, perfect or otherwise, as areas of the film may be suitable for examination.

- Collect blood in an EDTA tube and make the smears when back at the clinic
- Use clean, high-quality microscope slides
- · Aim for a blood droplet size of 4 mm diameter
- Optimise spreading speed for length and a good feathered edge
- Hold the spreader slide at 30-40 degrees to achieve optimal smear length
- Maintain even contact throughout the spreading motion.

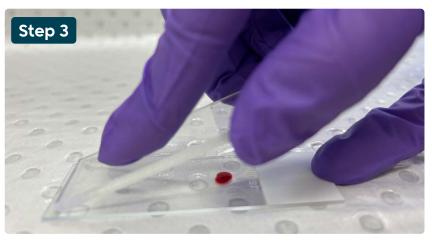




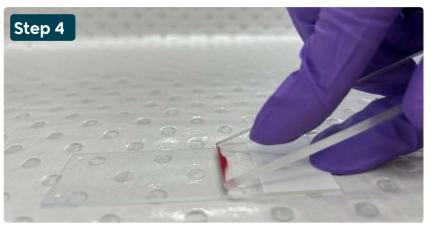
- Fill a capillary tube 3/4 full with blood
- · Hold the capillary tube vertically over the slide
- Allow 1 drop of blood to form on the end of the tube.



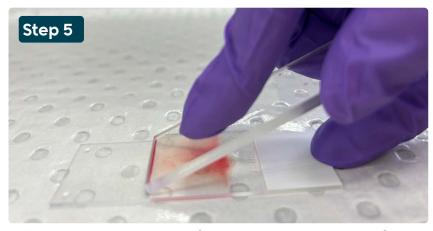
- The tube onto the slide about 0.5 cm from the frosted area
- Leave a drop of blood about 4 mm in diameter on the slide.



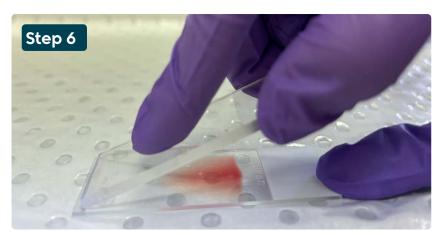
- Hold the spreader slide or second microscope slide at a 30-40 degree angle at the end of the slide (i.e. in front of the blood droplet)
- Ensure the short edge of the spreader slide is in even contact with the lower slide
- Pin the lower slide to prevent it from moving.



- Using a smooth motion, draw the spreader slide back through the entire drop of blood
- Allow the blood to spread evenly along the edge of the spreader slide.

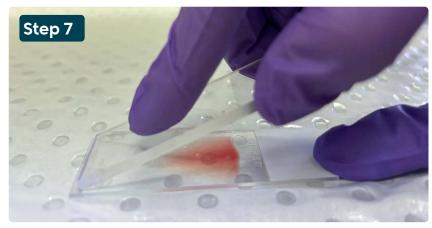


 Push the spreader slide forward along the length of the lower slide.

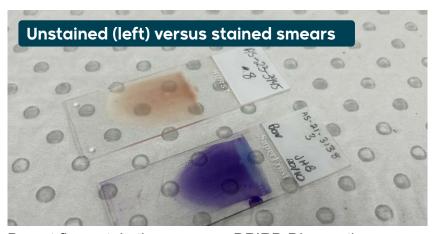


Maintain a constant smooth motion, angle and even contact.

Note: blood is being dragged behind the spreader slide, not in front of the slide.



- An optimal smear is ¾ the length of the slide and has a feathered edge
- Leave the slide to air dry and make more smears if required
- · Pack smears individually into slide holders.



Do not fix or stain the smears – DPIRD Diagnostic Laboratory Services (DDLS) will do this under controlled conditions to optimise the staining process.

Troubleshooting blood smear errors

Problem	Solutions		
Short smear	Use a larger droplet of bloodDecrease the angle of the spreader slideDecrease the speed of the spreader slide.		
Long smear / no feathered edge	 Use a smaller drop of blood Increase the angle of the spreader slide Increase the speed of the spreader slide. 		
Thick smear	 Use smaller drop of blood Decrease the angle of the spreader slide Increase the speed of the spreader slide. 		
Thin smear	 Use a larger drop of blood Increase the angle of the spreader slide Decrease the speed of the spreader slide. 		
Smear has waves and ridges	es and • Increase the speed of the spreader slide		

Reference: https://www.vet.cornell.edu/animal-health-diagnostic-center/laboratories/clinical-pathology/samples-and-submissions/hematology



The blood droplet on the left is too big and will result in a thick smear. The blood droplet on the right is the preferred size.



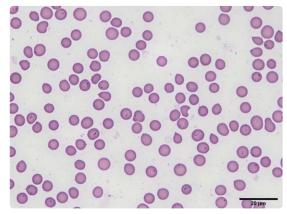
Slide 1 – perfect smear Slide 2 – smear technique interrupted in middle Slide 3 – smear was skewed Slide 4 – blood droplet too thick Slide 5 – smear too short.

Infectious diseases in Australian ruminants diagnosed by blood smear

Disease	Species	Transmission	Disease status
Anaplasmosis	cattle	tick-borne	endemic – northern Australia
Babesiosis	cattle	tick-borne	endemic – northern Australia
Mycoplasma ovis (eperythrozoonosis)	sheep	iatrogenic, blood-sucking insects (midges, mosquitoes, flies)	endemic
Bovine anaemia due to Theileria orientalis	cattle	tick-borne	endemic to parts of eastern Australia and South west western Australia

Bovine erythrocytes

infected with protozoal parasites from the *Theileria* orientalis group. The parasite is known as a piroplasm when it is within an erythrocyte. Piroplasms appear in erythrocytes from day 10 post-infection. Naïve, young, pregnant or immune-compromised animals may develop severe anaemia and mortalities can be as high as 30 per cent in a herd.



Example only.

Not a diagnostic resource

Haemaphysalis longicornis (common bush tick)

Ticks are vectors for many protozoal and mycoplasmal parasites. For *Theileria* spp. transmission in the tick is known to be trans-stadial or



life stage-to-life stage. A larva or nymph stage tick transmits the parasite to the next animal it feeds on. Trans-ovarial transmission (transmission from infected females to their larvae) does not occur. Control of ticks and good sanitation when using needles and surgical equipment minimises the introduction of these infectious parasites into naïve populations.