

primefact

DIY worm egg counting

December 2017, Primefact 1613, first edition Stephen Love, Veterinarian/Research Officer (Parasitology), Armidale

Introduction

There are many types of internal parasites in the world that affect many different species of animals. A common diagnostic method to assess the size and significance of worm burdens is to count the eggs shed by these worms in faeces or excreta. This is called a faecal egg count (FEC) or worm egg count (WEC).

This Primefact outlines procedures for doing your own ("DIY") worm egg counts for grazing livestock including free range poultry. NSW DPI also provides courses on how to do worm egg counts (See Tocal Skills Training in 'References').

Types of internal parasites

They can be divided into groups:

Nematodes (roundworms)

Roundworm eggs generally float in saturated salt (NaCl) solution. Common roundworms in ruminants include barber's pole worm (*Haemonchus*), black scour worm (*Trichostrongylus*) and brown stomach worm (*Teladorsagia; Ostertagia* in cattle).

The large roundworm (*Ascaridia galli*) is a common worm in poultry. It has a direct life cycle, although, as with *Heterakis*, earthworms can be transport hosts. *A. galli* lives in the intestine and has moderate pathogenicity. Others include the caecal worm (*Heterakis*, also common in poultry) and various *Capillaria* species, which mostly occur in the intestines. *Heterakis* can act as a vector for the causative agent of the protozoan disease, 'blackhead' (enterohepatitis; histomonosis) in turkeys.

Trematodes (flukes)

These can be important parasites of grazing livestock such as ruminants. The most important

fluke affecting ruminants in Australia is liver fluke (*Fasciola hepatica*). Liver fluke eggs are relatively dense and do not float in saturated salt (sodium chloride) solution, which has a specific gravity of 1.2. Labs mostly use a sedimentation technique rather than flotation to count fluke eggs.

Cestodes (tapeworms)

A variety of tapeworms, for example, *Raillietina* spp, are found in the intestines of poultry. Tapeworms have indirect lifecycles.

The common intestinal tapeworm of sheep and goats is *Moniezia*. It rarely has a significant impact on the animal.

Protozoans

Coccidia are protozoans (single-celled microorganisms) that can affect all domestic animals. There are many species of coccidia affecting livestock, including poultry, and most are hostspecific.

Basic parasite life cycles

The two main types of worm life cycles are direct and indirect.

Direct life cycles

Direct life cycles only involve one host. Many (but not all) roundworms have a direct life cycle. The sexually mature adult parasite lives in the host and produces eggs that are expelled in the faeces. Larvae hatch from these expelled eggs, develop through stages to the infective form, and are ingested by the host. In some cases, larval development through all stages occurs within the egg which is ingested by the host. The infective larvae migrate to the preferred site of infection (predilection site) within the host and develop into mature adults, which then reproduce. And so the cycle goes on.

www.dpi.nsw.gov.au

Indirect Life Cycle

Indirect life cycles involve two different types of hosts. Tapeworms often have an indirect life cycle.

The host that is infected with the sexually mature parasite is called the definitive or final host. The adult produces eggs that are passed out in the faeces. The infective larvae then enter another type of host, the intermediate host. Examples of intermediate hosts are ants or beetles in the case of *Raillietina*; fleas in the case of the common dog and cat tapeworm; and a pasture mite in the case of the sheep intestinal tapeworm.

The parasite develops further in the intermediate host and then, by various means, depending on the particular parasite, comes into contact with the definitive host. The parasite then develops to the adult stage within the definitive host.

Individual or bulk worm egg counts?

The following is largely drawn from an article by Kahn (2017).

As mentioned previously, WormTests identify the number of worm eggs in faeces (WEC). This is a good indication of the worm burden of sheep and goats. Some laboratories also do 'larval cultures' (also called 'larval differentiation' or 'worm type') to ascertain the types of roundworms present and what proportion they are off the overall roundworm burden. This can be useful for interpreting the WormTest result and deciding on treatment.

Various laboratories and suppliers, including veterinarians, provide WormTest kits for their clients. The procedure described by the testing laboratory should be followed.

Faeces must be collected from freshly deposited dung piles or from individual sheep, regardless of method (individual or bulk). If the samples are being used to establish Australian Sheep Breeding Values (ASBV) for WEC, samples must be collected from individual sheep.

Individual worm tests

For individual WormTests, faeces from individually-deposited faecal piles are collected into separate containers. These are then analysed individually in the laboratory, or by you if 'DIY'.

Usually 10 containers are provided in WormTest kits. The lab report will show the individual WEC of the 10 sheep, although animal IDs may be unknown if samples are collected from piles

deposited on pasture. Usually the report also shows the average of the 10 WECs.

When the aim is to provide WECs for calculation of ASBVs, faeces are collected from individual sheep, not from faeces on the ground.

Table 1. Individual versus bulk worm egg counts (Source: Kahn, 2017)

	Individual method	Bulk method
Strengths	Shows the variation in WEC among animals in a mob	Samples a larger proportion of the mob
	Essential for determining ASBVs	Results more likely to reflect the true mob average
	Essential for DrenchTests	
	Essential for post- treatment DrenchCheck	
Weaknesses	Samples a smaller proportion of the mob	Does not show variation in WEC among animals in a
	Results may not reflect the true mob	mob
	average	Cannot be used for determining ASBV
		Cannot be used for DrenchTests
		Should not be used for post-treatment DrenchCheck

For more information on 'DrenchTest' (faecal egg count reduction test) and 'DrenchChecks' (WEC before and after drenching a mob of sheep), see wormboss.com.au

Bulk worm tests

This requires faeces from individually-deposited faecal piles to be collected into a single bulk container, the contents of which are then thoroughly mixed. Then 4–5 subsamples from this bulk sample is analysed in the laboratory (or by you).

For bulk WormTests, samples are taken from 20–40 faecal piles (equivalent to 20–40 sheep). Twenty is enough except where barber's pole worm is an issue or if the mob or herd has over 200 animals.

Collect the same amount from each pile: for adults, about three pellets from each pile or the equivalent amount if the faeces is soft or runny. For weaners, take 5 pellets per pile. Laboratories vary in how they report results. They may provide the WEC of the subsamples; they will definitely provide the average of WEC values for the different subsamples.

Kahn (2017) discusses in some detail the strengths and weaknesses of the two methods. They are summarised in a table, reproduced above (Table 1).

Keep in mind that either method is better than not testing at all!

Faecal egg counting procedure

(Modified McMaster technique. Reference: Hutchinson GW, 2009)

Following is a standard method, including materials required, for doing worm egg counts using faeces (mammals) or excreta (poultry).

Although a faecal worm egg count (WEC) is not always directly related to the number of worms in a host, particularly in yearling and adult cattle, it often it gives a reasonable idea of the level of infection, in small ruminants, for example.

Different species of worms produce different numbers of eggs. And some worms species are more pathogenic (causing more damage to their host) than others.

Due to their lower density (compared to, say, liver fluke eggs), the eggs of roundworms, and some other parasite eggs, float in saturated solutions of salt (sodium chloride). Sometimes other saturated solutions are used, for example, sugar or zinc sulphate. (See RVC/FAO in References).

Floating the eggs separates them from other faecal material, and they can be counted at the under surface of a counting slide. By using known quantities of samples and reagents, the number of parasite eggs per gram (epg) of faeces can be determined.

The WEC is the most routinely performed test to estimate the worm population in an animal. The standard method for faecal worm egg counting (McMaster technique and variants) is derived from original methods developed by the CSIRO McMaster Laboratory in NSW. The details, for example, the amount of faeces weighed out and hence multiplication factors, vary from lab to lab, but the principles are the same. Other variations include using volume not weight to measure out the faecal sample, and also, as used in a University of New England lab, to mix faeces in water, not saturated salt solution, with the saturated salt being added at a later stage, i.e. into the chamber of the counting slide (UNE Micro Method; see References).

Safety

All faecal samples should be regarded as potentially infectious and appropriate hygiene procedures including hand washing should be followed when handling faeces.

Materials required

• Electronic balance / scales: capable of weighing 3 grams and accurate to 0.1 gram.

• Compound microscope (Figure 7): 40x and 100x final magnification (4x and 10x objectives with 10x eyepieces), mechanical stage, binocular recommended. A monocular microscope is acceptable but less comfortable if you are examining many samples.

• A mechanical stage makes it much easier to move the slide as you count.

• Faecal jars: small jars/vials capable of holding 60 ml of solution with some space at the top (to reduce spillage when mixing).

• Battery hydrometer: Available from most hardware or auto parts stores (for assessing car battery acid). Use this to monitor the specific gravity of the salt solution.

 Counting slide: Whitlock Universal counting chamber. http://www.whitlock.com.au/slides/Spcuni6.htm

• Spatula or blunt knife blade: for weighing out faeces, and stirring, unless a rod is preferred for the latter.

• Paper towel.

• Loading rod (pipette), syringe or syringe barrel with mesh covering one end, for example ~1 mm flyscreen mesh, or wire mesh with approximately 12 meshes per cm. This is for loading prepared samples into the chamber. Disposable plastic bulb pipettes may also work but can easily get blocked with plant material in the excreta. Some filter the faeces-salt solution mix through a tea strainer or similar before loading the pipette.

Figure 1. Disposable plastic bulb pipette and, below that, a 'plastic' tubular pipette (or 'loading rod') with stainless steel mesh (sieve) on the bevelled end. These are just examples of what can be used.



Image credit: S Love

• Water bottle.

• Container of clean water for rinsing the loading rod between samples.

• Damp cloth/sponge.

• Non-abrasive tissues suitable for cleaning glassware on microscopes.

• Worksheet to record results.

• Air blowing apparatus for agitation (e.g. aquarium pump). This is used when filling the loading rod (optional).

• Permanent marker pen.

Solutions

Saturated salt solution (specific gravity: 1.2)

One method of preparing solution The following method is used in some labs.

Place 5-6 large cups of swimming pool salt (cheaper than table salt) in a 9-10 litre plastic bucket. Three quarters fill with hot tap water and stir vigorously for 1-2 minutes. Allow to cool.

There should be a small amount of un-dissolved salt in the bottom of the bucket after the solution cools. Always measure specific gravity (density relative to water) with a battery hydrometer. To do this insert the hydrometer in the solution, and fully squeeze the rubber bulb, sucking up sufficient solution to allow the hydrometer to float freely in the solution. The specific gravity of the solution should read '1.2' (or '1200' depending on the units used). If the solution is below 1.2, dissolve a cup of salt in a small amount of hot tap water and add to the bucket. Check the solution again when cool.

One litre of saturated salt solution with a specific gravity of 1.2 weighs 1.2 kg. One litre of plain water weighs 1.0 kg.

It is important to measure the specific gravity as this will affect the number of worm eggs floating to the top of the slide during the egg counting procedure.

Another method (which may be better)

Add 400g of salt for each litre of hot water. This amount of salt is just above what is required (~357grams; see references) to make a litre of saturated salt solution at 25 degrees Celsius. When the solution has cooled, there should be some undissolved salt in the bottom. Nonetheless, always check the specific gravity. Using weight rather than volume of salt has another advantage: the so-called 'bulk density' of salt (not the same as 'density') varies depending on how fine it is.

Quality control

Occasionally you should submit samples to a certified laboratory to ensure that your own counts are comparable to those obtained by an experienced, qualified technician. WormTest sampling kits are available from NSW DPI (State Veterinary Diagnostic Laboratory – see www.dpi.nsw.gov.au), and other providers, and can be used for this purpose. Thoroughly mix (homogenise) a sample or samples of faeces. Do an egg count on one half of each sample and submit the other half (halves) to the lab.

Procedure

• Label sample jars identifying the animal name/number and note the details on the worksheet.

• Using the spatula weigh 3 g (+/- 0.5g) of faeces from each animal into each faecal jar. Clean the spatula on a clean damp cloth between samples.

(Different weights can be used. For example some labs may use 2.5g, but the multiplication factor at the end will have to be adjusted accordingly).

• If there is less than 3 g of faeces available, record on the worksheet the exact weight of faeces used.

• Cover faeces with approximately 10 ml (2 teaspoons) of water (to soften faeces, if necessary).

NOTE: Samples can be left at 4 °C if necessary after this step. Hard samples (more of an issue with sheep, especially with heavy barber's pole worm burdens) may require soaking before they are soft enough to mash.

• Mash faeces and mix with the water using the spatula, cleaning the spatula between each animal faecal sample.

NOTE: Samples that have been prepared with saturated salt solution should not sit for more than 30 minutes before counting as the salt may distort the eggs. For this reason, only fill the number of chambers that can be counted in approximately 30 minutes.

Preparation of the samples

1. Ensure that the faeces/excreta have been thoroughly mashed to break up pellets, if present, before adding saturated salt solution.

2. Add salt solution to 60 ml line on the jar.

(Different labs might use different volumes. The multiplication factor at the end will have to be adjusted accordingly).

3. Mix solution well with spatula or rod (glass or plastic) to ensure faeces and salt solution are thoroughly combined. (Laboratories sometimes use a mechanical or air powered mixers, or a hand-held kitchen mixer, until all faeces are broken up and well mixed. A downside might be more air bubbles). Stir the sample (in a north-south and east-west stirring (paddling) motion – as opposed to circular stirring, which doesn't mix as well. If necessary, add one or two drops of methylated spirits to disperse any air bubbles.

6. Immediately after stirring, pipette a sample (aliquot) from the suspension in the jar, holding your index finger or thumb over the end of the sieved loading rod (Figure 1) (if you are not using a pipette with bulb) when withdrawing it from the solution. This is to hold the collected sample inside the rod. Note the following:

a. Drain pipette or rod before collecting sample.

b. Lightly bounce the pipette while filling to prevent blockage of sieve by fibre.

c. Tilt the mixture if necessary to ensure adequate filling of the pipette.

d. Aim to take the sample from the centre of the jar, but this is not critical if the contents of the jar are well-mixed right up to the time a sample (aliquot) is drawn up. Depending on the pipette or rod you are using, and the shape of the jar, you may need to place the end of the rod near the bottom of the jar to ensure you draw up enough solution to fill the chamber in the counting slide. This is not an issue if using a syringe or pipettes with a bulb.

(Another variation: some strain the sample through a tea strainer rather than using a loading rod with mesh on the end).

7. It may help to dab the end of the pipette with a tissue to remove bubbles from the surface of the mixture.

8. Hold the slide with the 'veranda' facing away from you.

9. It may help to softly breathe into the counting chambers of the slide. This 'wets' the surface and allows easier loading of the sample.

10. Hold the pipette horizontally and fill the chamber in one smooth action without producing bubbles. Completely fill either one chamber of the counting slide (sheep, goat, deer) or two chambers for species that tend to have fewer eggs in their faeces/excreta (cattle, horse,

alpaca, native animals). Load the slide from left to right.

11. Rinse loading rod in clean water between faecal samples.

12. Place counting slide on a piece of paper towel next to the microscope (with the 'veranda' facing towards the back. This helps to keep the bottom of the chamber clean and dry. Water and salt are not good for microscopes.

With some microscopes, you may need to face the 'veranda' to the front (it does not matter too much), so that you can see all parts of the chamber when you are counting. It depends on the range of motion your microscope's mechanical stage, if your's has a movable stage.

13. Before counting allow approximately one minute after preparation for the eggs to float to the top of the counting chamber.

Figure 2. Eggs commonly seen in faeces of grazing livestock (mammals), with the 'strongyle' (aka 'trichostrongyle', 'strongylid') egg type being most common. Regarding poultry: neither ascarid (fig.3) nor *Capillaria* eggs are shown here, but the poultry roundworm *Trichostrongylus tenuis* has a 'strongyle' type egg and *Capillaria* eggs are similar to *Trichuris* (whipworm) eggs.



Image credit: Anne Oakenful, EMAI, NSW DPI

Figure 3. Ascarid eggs. This example is *Ascaridia galli* which infects birds.



Image credit: Nisha Sharma, UNE

Notes on Figure 3: *Heterakis* (poultry caecal worm) eggs are a little smaller than, but otherwise similar to,

those of *A.galli*. Note the thicker egg wall of ascarids compared to the 'strongyle' types. Ascarid-type eggs are commonly seen in other livestock, e.g. young horses, and pigs (and also in humans). The egg here (magnified somewhat!) is approximately $80 \times 50 \mu m$ (microns), roughly the size of the 'strongyle' eggs in Figure 2 above. For comparison, a human hair on average is about 100 microns wide (range: 50-180 microns) and a red blood cell is approx. 8 microns in diameter.

Counting worm eggs

1. Place the counting slide on the microscope stage with the 'veranda' facing away from you. Consistent orientation of slides must be maintained to ensure accurate identification of samples. The important thing is to be systematic.

2. Using 40x total magnification (4x objective and 10x eyepiece(s)), focus on the air bubbles just beneath the top glass of the slide. Air bubbles appear as perfectly round doughnutshaped structures with a bright clear centre, and thick black outline. For closer examination, 100x final magnification can be used, but for routine counting, 40x is best as you can see the full width of each lane at that magnification.

3. Begin counting at the top left hand side of the slide.

4. Count all eggs within the double line boundaries, i.e. 5 lanes for each chamber. See the diagram of the counting slide (Fig.5). When counting eggs that fall on the line, either count those on the top and the right lines, or on the bottom and left lines. You choose: the important thing is to be systematic in order to avoid double counting.

5. Count eggs in each consecutive chamber and record the results on the worksheet.

Table 2. Multiplication for calculating eggs per gram (epg) of faeces/excreta, assuming 3 grams of faeces, 60ml of salt solution and a Whitlock Universal slide (0.5ml chambers).

Animal	No. of chambers	Multiplication for epg result (x)
Sheep	1	40
Goats	1	40
Cattle	2	20
Horse	2	20
Poultry (4g sample) ¹	1	50
Alpaca	2	20
Native animals	2	20

¹Poultry (or other animals): when a 4g sample of excreta is used, made up to 60ml with flotation

solution (usually salt solution), and a slide with 0.3 ml chambers is used. See appendix for more information.

Figure 4. Layout of Whitlock Universal Counting slide. This slide has 4 chambers, each nominally holding 0.5ml.



Image credit: NSW DPI-'Tocal Skills Training' (Profarm) Worm Egg Counting Course Manual / Anne Oakenful.

Microscope slides

The Whitlock Universal slide is specifically designed for parasite worm egg counts in large and small animals. Currently (at 1 March 2017) they cost \$145 including GST. They are glass sides and do last a long time if looked after. (Dropping on a concrete floor is not advised) These slides are commonly used in veterinary diagnostic laboratories, including the NSW DPI veterinary laboratory.

The McMaster (3 \times 0.3ml) and Paracytometer (2 \times 0.6ml) slides have been discontinued, having been superseded by the Universal.

Whitlock Universal slides are available from:

JA Whitlock & Co., PO Box 51, Eastwood NSW 2122. Phone: +61 2 9638 1142.

Web site: www.whitlock.com.au

Acrylic slides may be cheaper. A disadvantage of these is that they can gather quite a few scratches over time.

Slides are also available from sources apart from the one listed above.

Figure 5. Whitlock Universal counting slide



Image credit / source: JA Whitlock http://www.whitlock.com.au

Microscopes

There are a number of different sources of microscopes. Prices vary depending on the quality.

Figure 5. Compound biological microscope



The basic microscope requirements for faecal egg counts are:

- Compound microscope with 40x 100x magnification (10x eyepiece with 4x and 10x objectives).
- Binocular eye pieces (monocular can be used however it can cause eye strain if doing multiple samples).
- Mechanical stage.
- Power supply for lighting.

The web is a good source of information on microscope products, prices and suppliers. Also enquire about the availability of periodic servicing.

Most laboratory suppliers have a microscope range, including **digital microscopes**, that covers the basic needs required for egg counts. Some examples, in alphabetical order:

- A.I.S. Australian Instrument Services Pty http://www.ausinst.com.au/catalogue/
- EggsAct Pty Ltd

http://www.faecaleggcountkit.com.au

- Logical Interface. http://www.logint.com.au or http://www.microscopeshop.com.au/binocular-microscope.aspx
- Lomb Scientific

www.lomb.com.au

 ProSciTech https://www.proscitech.com.au/

- Southern Biological http://www.southernbiological.com/
- Other service providers:

http://www.wormboss.com.au/teststools/professional-service-providers.php

Ebay occasionally have microscopes on sale and can be worth looking at from time to time to see what is available.

Universities, colleges and schools sometimes dispose of second-hand microscopes.

Hydrometer

Battery hydrometers, which can used for checking specific gravity of salt solutions (as well as battery acid etc) are readily available at most hardware stores, and auto parts stores. They are relatively cheap, about \$10, and are a good investment to ensure that all flotation solutions are at the required specific gravity to float parasite eggs.

Figure 6. 'Trichinelloid' or *Capillaria* – type egg. Similar to *Trichuris* (whipworm) egg in Figure 2. '*Capillaria*' are generally hair-like worms found in a range of animals, including poultry, and in a variety of different locations in different hosts (e.g. crop, intestine, caecum in poultry). Some *Capillaria* worms have changed names. This one below (4 eggs in lung of a rufous bettong (small Australian marsupial)) was formerly *Capillaria* sp nov 7 (1992), now *Eucoleus potoroi.* (The arrows are pointing to various features of the eggs).Note the clearly visible polar plugs of the egg in the centre.



Image: WH Johns. Source: Love and Reddacliff, 1992.

Appendix – extra notes

Calculations

How do you calculate the egg count (eggs per gram of faeces) from the raw egg count (the number of eggs you counted in the counting slide (whether one or two chambers))?

Eggs per gram (epg) of faeces =

eggs counted in scanned area of slide x volume of faeces-flotation solution mix

divided by

weight of faecal sample x volume of the counting chamber(s).

Example 1

Assume this is the method you use:

3g of faeces is put into a jar (plus a small amount of water to aid breaking up / mixing the faeces). Then flotation fluid (commonly saturated salt (NaCl) solution) is added to make a total volume of 60 mls. After thoroughly mixing, a pipette (or syringe or sieved loading rod) is used to draw some of this fluid up for transfer to a chamber in a Whitlock Universal slide. (These have 5 x '0.5'ml chambers). (In this method, usually one chamber is counted per sheep sample, and two chambers for each cattle sample).

Say there are 55 eggs counted. The EPG = $55 \times (60) / (3 \times 0.5) = 2200$ The multiplication factor in this method is 40 (60/(3 x 0.5)). In this example, the 'raw egg count' of 55 times 40 = 2200 eggs per gram (epg) of faeces,

Example 2

Say your method uses a 4 g faecal sample, made up to 60 ml, and a slide with 0.3ml chambers is used. The multiplication factor is $(60) / (4 \times 0.3) = 50$.

Volume of chambers

Different versions of counting slides may have chambers with different volumes, e.g., 0.3, 0.5 or 0.6 mls. These are 'nominal' volumes and refer to the volume of fluid under the lines scored on the slide. The actual volume of the chamber is a little bigger than its 'nominal' volume.

Size of mesh for sieve/filter

What size mesh is used for the sieve designed to filter out extraneous material (plant material etc.) from the faeces-water-flotation solution mixture?

Hutchinson, 2009 (~page 17, line 770), suggests a mesh with 12 meshes per centimetre.

Acknowledgments

Some of the information in this PrimeFact is derived from the NSW DPI Profarm Worm Egg Counting Course Manual, by Anne Oakenful, revised by Stephen Love.

Many thanks to Dr Peter Hunt, Senior Research Scientist, CSIRO Armidale, for helpful comments and suggestions while this Primefact was being prepared.

References and more information

Hutchinson GW, Hutchinson GW (2009), Nematode Parasites of Ruminants: Australian and New Zealand Standard Diagnostic Procedures. Sub-Committee on Animal Health Laboratory Standards. Accessed March 2017 at http://www.agriculture.gov.au/animal/health/labor atories/procedures/anzsdp/nematode-parasitesruminants Also at:

https://wormmailinthecloud.wordpress.com/2017/ 04/03/nematode-parasites-of-ruminantsaustralian-and-new-zealand-standard-diagnosticprocedures-and-variations/

Kahn LP, 2017. Individual or bulk worm egg counts: is there a difference? ParaBoss / WormBoss feature article, October 2017. Accessed November 2017 at http://www.wormboss.com.au/news/articles/tests/ individual-or-bulk-worm-egg-counts-is-there-adifference.php.

Love S and Reddacliff G, 1992.Verminous bronchitis and bronchiolitis in potoroid marsupials associated with a new *Capillaria* sp. Journal of Wildlife Diseases.

NSW DPI "Profarm" egg counting course: http://www.dpi.nsw.gov.au/content/agriculture/pro farm/courses/faecal-egg-counts

Merck Veterinary Manual

http://www.merckvetmanual.com/poultry/helminth iasis/overview-of-helminthiasis-in-poultry (Not from a uniquely Australian perspective, but still useful).

ParaBoss: www.paraboss.com.au Includes WormBoss, FlyBoss and LiceBoss. Cattle-related material to be added ~ 2019.

Profarm: see Tocal Skills Training.

RVC/FAO: Royal Veterinary College / FAO Guide to Veterinary Diagnostic Pathology: Flotation fluids – general purpose. Accessed March 2017 at

http://www.rvc.ac.uk/review/parasitology/Flotation /Flotation_fluids/General.htm Salt: solubility/solution stability: Maximum solubility of NaCl in water at 25°C is 357 mg/ml. NaCl is unusual in that its solubility does not increase appreciably with temperature, since at 100°C, the solubility is 384 mg/ml. The solubility of NaCl in water is decreased by adding HCl; it is almost insoluble in concentrated HCl. The density of a saturated solution at 25°C is 1.202 g/ml. A saturated solution (23% w/v) freezes at -20.5°C (5°F). Accessed December 2017 from https://www.sigmaaldrich.com/content/dam/sigma -aldrich/docs/Sigma/Datasheet/6/71386dat.pdf Also see 'RVC/FAO'.

Standard parasitology procedures. For variations on these (including the 'UNE micro method method), and some resources: WormMail 2017-04-03

https://wormmailinthecloud.wordpress.com/2017/ 04/03/nematode-parasites-of-ruminantsaustralian-and-new-zealand-standard-diagnosticprocedures-and-variations/ Accessed April 2017.

Tocal Skills Training, formerly PROfarm. Information on course, "Faecal egg count for worms":

https://www.dpi.nsw.gov.au/content/agriculture/to cal-skills-training/courses/faecal-egg-counts

UNE 'Micro Method'.

https://wormmailinthecloud.wordpress.com/2017/ 11/30/une-micro-method-for-doing-worm-eggcounts-on-bulk-faecal-samples-sheep-goats-etc/ Accessed 2017-11-30.

Whitlock slides and prices: http://www.whitlock.com.au/slides/PricewGST2017b.html (Accessed March 2017).

WormBoss: www.wormboss.com.au

© State of New South Wales through the Department of Industry, Skills and Regional Development, 2017. You may copy, distribute and otherwise freely deal with this publication for any purpose, provided that you attribute the NSW Department of Primary Industries as the owner.

Disclaimer: The information contained in this publication is based on knowledge and understanding at the time of writing (December 2017). However, because of advances in knowledge, users are reminded of the need to ensure that information upon which they rely is up to date and to check currency of the information with the appropriate officer of the Department of Primary Industries or the user's independent advisor.

ISSN 1832 6668

HPE Content Manager reference: INT17/261810