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Worms – what's new: some recent developments in worm control in ruminants and camelids

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Abstract: Worms are economically important, being the number one disease in sheep in Australia, and number two (after bloat) in cattle in southern temperate regions of Australia (possibly number one if liver fluke is included). Faecal worm egg counting (WEC) is still the standard diagnostic tool, with useful adjuncts such as the lectin binding assay and the *Haemonchus* Dipstick Test being tested and deployed in recent years. There has been a shift away from prescriptive, calendar-based strategic worm control programs for sheep to modified strategic programs with an increased emphasis on integrated parasite management and treatments based on worm egg count monitoring rather than visual assessment or the calendar. Use of effective combinations and drench rotation are recommended. Monitoring of drench efficacy by way of post-drench WEC in small and large ruminants and alpaca is recommended.

Resistance has worsened during the last 10 years with macrocyclic lactones (MLs) in particular being affected. The prevalence of sheep/goat worm resistance to the MLs is quite high in certain areas. Resistance has been identified as a significant issue in cattle worms in New Zealand, and it appears to be an emerging issue in Australia as well. Two new drench groups have appeared on the horizon in the last two years. One, the AADs (aminoacetonitrile derivatives), represented by monepantel (Zolvix® (Pfizer)) was recently launched in Australia, about 1.5 years after the world launch in New Zealand. Another, the spiroindoles (represented by derquantel, marketed in combination with abamectin as Startect® (Pfizer)), was released in NZ in July 2010. In the past in Australia, first reports of resistance to new drench groups have typically appeared just five years or so after the launch of the new chemicals. Taking measures to manage resistance will hopefully ensure that resistance does not develop so rapidly in the case of the AADs and spiroindoles.

Keywords: worms, faecal egg counting, integrated parasite management, resistance.

Worms – why bother? (new costings)

According to a 2006 review of the cost of endemic diseases in Australia, internal parasites are the number one disease of sheep, closely followed by external parasites (lice and flies) (Sackett et al. 2006).

For cattle in the southern temperate zones of Australia ('cattle-southern'), gastrointestinal parasites¹ (excluding liver fluke) are ranked second after bloat in economic importance. Liver fluke is also categorised as a 'high economic cost disease' (Sackett et al. 2006). If liver fluke is included with gastrointestinal parasites, then worms may be the number one disease in cattle (southern) (see Figure 1).

As an aside, the cost of pestivirus, undoubtedly an important disease agent in cattle, was not modelled for various reasons.

Worms are also among the most important endemic disease agents of goats and camelids such as alpaca.

New tests?

The long-term standard method of diagnosing endoparasitism in livestock has been the faecal worm egg count (WEC), employing the McMaster technique or variations of it. This technique is based on the fact that roundworm (nematode) eggs float in

saturated salt (specific gravity 1.2) or sugar solutions, enabling counting using chambers (e.g. Whitlock) and microscopes (40x or 100x). Liver fluke eggs are somewhat denser and will not float in salt solutions: laboratories use a sedimentation technique for this egg type instead.

Because the eggs of important roundworms of ruminants look similar, the faeces is mixed with vermiculite and incubated in order to produce larvae. The larvae from the cultured faeces are counted and differentiated by genus 7-10 days later.

WECs are sometimes complemented by necropsy and total worm counts. Plasma pepsinogen is also sometimes used in cattle as indication of damage to the abomasum from *Ostertagia ostertagi* (brown stomach worm).

Considerable thought and effort has gone into devising tests that are faster, cheaper and more accurate than the WEC (e-noses, immunological assays, etc), and also bypass the need for larval differentiation. But so far WEC remains the standard diagnostic tool.

However, there have been some useful additions to WEC.

One is the lectin assay developed by Palmer and Morcombe (1996). In this test peanut lectin, which binds to *Haemonchus* (barbers pole worm) eggs, is used to differentiate *Haemonchus* from other roundworm genera, without having to resort to larval culture and differentiation.

¹ Liver fluke is commonly included as a gastrointestinal parasite because of the intimate association of the liver with the GI tract.

Another is the *Haemonchus* Dipstick Test (HDT). This test uses existing dipstick technology to detect occult ('hidden') blood in faeces, and works on the well-founded assumption that the most likely cause of haemoglobin being present in sheep faeces is the blood-sucker, *Haemonchus contortus*.

Haemonchus infections in small ruminants and alpaca typically yield very high WECs, due to the fecundity of adult female *Haemonchus* worms (approximately 10,000 eggs per female per day, compared to just a few hundred for the important scour worms (small brown stomach (*Teladorsagia* (*Ostertagia*) *circumcincta*) and black scour worm (*Trichostrongylus* spp)).

However, the prepatent period² for the important roundworms of ruminants is about three weeks. Under extreme conditions (warm weather and ongoing rain), *Haemonchus* burdens can build up very rapidly, with significant blood loss occurring before WECs rise, about 18-21 days after initial infection. The HDT has the advantage that it can detect significant *Haemonchus* burdens (by way of haemoglobin in faeces) a week or more before the WEC rises. The test is also relatively quick, cheap, and easy.

However, the HDT is an adjunct to rather than a replacement for WEC.

The humble WEC remains one of the pillars of good worm management, at least in small ruminants and alpaca.

Spitting on worms (saliva test for host resistance)

Currently in Australia, WECs are used to determine the host resistance status of rams.

In NZ, a saliva test has been developed, originally by Gavin Harrison, formerly of Agresearch, Wallaceville. This test, now commercialised, measures immunoglobulin A to a carbohydrate larval antigen (CarLA) found in 3rd stage worm larvae of many nematode species. (It was originally isolated from *Trichostrongylus colubriformis*, one of the black scour worms commonly found in sheep). It remains to be seen if this will be available for and applicable to Merinos and other sheep in Australia.

New approaches

Move away from the prescriptive, and toward more worms in refugia

The original WormKill program (launched in 1984, for the summer rainfall zone of northern NSW) was the first of the modern worm control programs in Australia and

arguably one of the most successful (in terms of adoption rate) of any extension program let alone worm control programs (Newman 1984).

WormKill 1984 was simple and prescriptive, with strategic drenching being performed on specific dates. For the first 5-10 years, the program was highly successful, especially against *Haemonchus*, until closantel-resistant *Haemonchus* began to appear from the late 1980s. By the mid-1990s resistance to closantel, a cornerstone of WormKill 1984, was common in northern NSW, and worm control become complicated and uncertain once again, even with the newly released (1995) moxidectin (Cydectin[®]) taking over from closantel as a persistent drug efficacious against *Haemonchus*.

The first Australian reports of resistance to ivermectin (released in Australia in 1988) were in the early 1990s: one from northern NSW (*Haemonchus* – Le Jambre 1993) and the other from Western Australia (*Ostertagia* – Swan et al. 1994).

In the main WA sheep zone, which has a Mediterranean climate, summer drenching was practised, with one or two drenches a year. The advantage of drenching when there are few worms on pasture (hot, dry summer, on cereal stubbles) is that there is very slow reinfection of sheep and little carry over of worms into the next season. A disadvantage is that there are few worms 'in refugia' (generally, eggs and larvae on pasture, therefore not selected for drug resistance) at the time of drenching. It is thought that 'the refugia principle' explains the relatively rapid development of ivermectin resistance in WA after just a few years and relatively few uses of ivermectin.

The last decade or so has seen a move away from prescriptive strategic programs, aiming to balance acceptable productivity with reduced selection for drench resistance.

Accordingly, in the late 1990s, DrenchPlan (the program for sheep worm control in the non-seasonal and winter rainfall areas of central and southern NSW) dropped the second summer drench as an automatic treatment, recommending drenching on the basis of WECs instead. Likewise, it was recommended that the first summer drench should be based on WEC in drought or very dry years, because of the paucity of worms 'in refugia'.

In WA, on the basis of work supported by the Sheep CRC³, there has been a move away from drenching all sheep at the beginning of summer. Young sheep are still treated at this

²The prepatent period is the time it takes for a new infection to become patent, i.e. to produce worm eggs in the faeces of the host.

³ Australian Sheep Industry Cooperative Research Centre (www.sheepcrc.org)

time, but adults are now treated in the following April, when there are more worms in refugia (Woodgate and Besier 2009)

So, there has been an increased emphasis on having sufficient worms in refugia, especially in non-seasonal and winter rainfall areas, and in areas where livestock are commonly grazed on crop stubbles.

Combinations are cool

Although combination broad-spectrum drenches have been available for some time, notably benzimidazole ('BZ', 'white') combined with levamisole ('LEV'; 'clear') drenches, there has been increased emphasis on using combinations of drenches if possible. This has been reflected in the number of combination broad-spectrum sheep drenches coming onto the market. There are even combination drenches available now for cattle (e.g. macrocyclic lactone (ML, 'mectin') + LEV), albeit only in New Zealand at this stage.

The theory behind combination drenches is that resistance will develop somewhat more slowly if unrelated actives are used in combination rather than sequentially. This is based largely on computer-modelling studies, but is accepted by most parasitologists, at least in the southern hemisphere.

Combinations will be most effective (in terms of efficacy and reduced selection for drug resistance) when the actives are highly effective in their own right, and their spectra of activity, and persistency, are similar. And, the more actives, the better.

Targeted treatments and more WECing

Various Sheep CRC projects have looked at targeted treatments, whether of mobs or individuals.

This is because worms are not evenly distributed among individuals in a mob or even between mobs. It is well known that a minority of animals in a mob produce most of the worms.

Also certain classes of stock, notably periparturient and lactating ewes, and lambs and weaners, produce a disproportionate number of worms on a property.

Of course, there are some old and simple technologies for targeted treatment. A well known one is the FAMACHA[®] system from *Haemonchus*-endemic regions of South Africa. This entails categorising individual sheep (or goats) on the basis of eye colour charts, with eye colour being an indication of degrees of anaemia arising from *Haemonchus* infections. Pale-eyed sheep are treated, and also tagged. Animals that are repeatedly treated (and tagged) are culled.

With the advent of automated drafting systems and electronic capture of data and identification of individual animals, there has been interest in using measures such as changes in bodyweight as a possible indicator of animals requiring drenching. It's early days yet.

WECing and better sampling of the mob

Traditionally when worm egg counts in a mob are monitored, 10 samples (either directly from animals or fresh dung piles on pasture) have been taken. This has always been a compromise due to cost considerations, but may not be adequate in larger mobs.

Additionally diagnosticians have often preferred 10 individual egg counts (as in 'WormTest Gold' in I&I NSW laboratory at EMAI, Menangle) rather than a bulk count of 10, or two bulk counts of five individual samples (as in 'WormTest Basic'). Against this it is argued that the distribution of WECs in a mob (a skewed rather than a normal distribution) is generally known in livestock such as sheep, and the range and distribution of WECs in a mob can be deduced from the average WEC.

So, there are moves afoot to:

- use bulk counts instead of individual counts when monitoring worm burdens (but not in drench efficacy testing), and
- increase the number of animals sampled, for example: sample 10% of the mob with a minimum of 10 and a maximum of 40-50 individual animals or dung piles sampled, with two pellets being taken from each dung pile.

(Lewis Kahn (UNE) and Maxine Lyndal-Murphy (DEEDI⁴), personal communications).

WECing in cattle

Alas, WECing is less useful in cattle than in small ruminants and alpaca. From around weaning (6-8 months of age), WECs in cattle tend to fall and become less correlated with worm burdens. Yearling and adult cattle with significant worm burdens may or may not have high WECs.

WEC is still a useful tool in cattle, and should increasingly be used, for example, 14 days post-drenching, to provide a quick check of drench efficacy (14 days in cattle, as opposed to 10 days (7-14 days) in sheep because of the effect of persistent ML-based cattle pour-ons, although theoretically at least the pre-patent period of some cattle *Cooperia* spp can be as short as 12 days).

⁴Department of Employment, Economic Development and Innovation, Queensland

WEC – liver fluke

Any positive fluke count in ruminants or alpaca is significant and requires action. The single most important fluke drench on an affected property in south-eastern Australia is in April/May, with the onset of frosty weather. A highly efficient flukicide (i.e. triclabendazole based) should be used at this time.

IPM, or 'prevent, detect and respond'

Worm control still depends very much on drenching, but there is an increasing emphasis on an integrated approach to worm management.

A recently coined phrase, 'Prevent-detect-respond', (Gareth Kelly (UNE), personal communication) is one way of describing the elements of IPM:

- Prevent: control exposure to infective larvae through grazing management; and ameliorate deleterious effects of worm infections through genetics and nutrition.
- Detect: regularly and systematically monitor worm burdens (and drench efficacy), largely through WECing, and monitor condition of animals.
- Respond: assess WEC results using a drench decision aid; use the appropriate, effective drench if indicated; modify management if necessary in light of monitoring of WEC and production targets.

Vaccines and biological control?

Work continues on vaccines, for example the "Moredun" *Haemonchus* vaccine by David Smith and others (Smith and Zarlenga 2006, Anon 2010). With few exceptions (for example, the lungworm vaccine for cattle used in Europe) there is no prospect of 'worm' vaccines for livestock on the market in the near future.

Malcolm Knox (CSIRO, Armidale) and others developed a biological control method using a nematophagous fungus, *Duddingtonia flagrans* (a fungus that eats worm larvae), that occurs naturally on Australian farms. It was developed to commercial stage but, in Australia at least, it has yet to reach the market.

Drench resistance –what's new

With the exception of monepantel (Zolvix[®]), recently released in Australia (September 2010), there are few if any drenches registered for use in small (or large) ruminants in Australia that are not affected to some degree by drench resistance.

The resistance situation, particularly with respect to the macrocyclic lactones, has changed markedly in the last 5-10 years. And

it is no longer just an issue for small ruminants.

Small ruminants and alpaca

Sheep, goats, and alpaca⁵ are grouped together here as they tend to carry the same worm species.

The prevalence of resistant worms in goats may be even higher than for sheep because goats generally metabolise drenches faster than sheep, with consequent effects on drench efficacy (and selection for resistance), unless dose rates are adjusted appropriately. (Because of differences in pharmacokinetics, veterinarians generally prescribe somewhat higher dose rates for goats and alpaca than those applicable for sheep.)

Resistance is generally and simply defined as failure of a drench to reduce worm egg counts post-treatment by 95% or more.

Resistance becomes apparent because of an increased frequency of resistance alleles in a worm population. Some drenches were never highly effective against some worms. This is not resistance. An example is naphthalophos (Rametin[®], Combat[®]), which from the outset was only moderately effective against *Teladorsagia* and *Trichostrongylus* spp worms in sheep.

Table 1 gives an overview of how common resistance to sheep drenches is in Australia. Some of the estimates may well be underestimates.

Resistance in Australian cattle worms not an issue??

Recent studies in NZ have shown that resistance is now an issue in cattle worms.

Waghorn et al. (2006) reported on a study of calves on 62 beef farms in the North Island of New Zealand. Faecal egg count reduction tests were performed, evaluating oral formulations of ivermectin, levamisole and albendazole, at recommended dose rates.

The results are summarised in Table 2.

Of the 27 species of nematodes recorded in cattle in NZ, three are considered important: *Ostertagia ostertagi* and *Trichostrongylus axei*⁶ because of their pathogenicity, and *Cooperia oncophora* because it is prevalent and widespread. (Although the production costs of not controlling drench-resistant *Cooperia* is currently being examined by Leathwick and others in New Zealand.)

The first report of confirmed cattle worm resistance in NZ was in 1987, and involved

⁵Alpaca can carry sheep and goat worms as well as camelid worms.

⁶*T. axei* is not considered to be a particularly pathogenic species in cattle or other livestock in Australia.

benzimidazole-resistant *Cooperia oncophora*. Things have progressed from there.

Resistant cattle worms in Australia?

In Australia there have been very few reports of resistance to cattle drenches, just two reports of benzimidazole ('BZ' or 'white') drench resistance in *T. axei* (stomach hair worm) (Eagleson and Bowie 1986; Eagleson et al. 1992), which fortunately is not a major worm species in this country. Anderson and Lord (1979) also reported thiabendazole and levamisole resistance in *Ostertagia ostertagi* in cattle.

However, during recent years there have been more field reports of suspected resistance to cattle drenches. And this year confirmed resistance from south-east Queensland was reported by Lyndal-Murphy et al. (2010).

This was the first Australian report of macrocyclic lactone ('ML', 'mectin') drenches failing against subtropical small intestinal worms (*Cooperia* species) in cattle - and possibly barber's pole worm (*Haemonchus placei*).

Rendell (2010) reported the results of field studies in Victoria, analysing post-treatment worm egg count reduction in calves on 13 Victorian properties. The results are summarised in Table 3.

Rendell's paper is the first Australian report of macrocyclic lactone-resistant *O. ostertagi* in the refereed literature. He noted that the frequency of resistance in *O. ostertagi* to BZ, LEV and IVM and in *Trichostrongylus* spp. to BZ in his study was higher than that in the 2004-05 New Zealand survey (Waghorn et al. 2006) whereas the resistance frequency in *Cooperia* spp. to IVM and BZ was less. So, resistance appears to be an emerging issue for Australian cattle producers.

Arguably the routine drenching of adult cattle for roundworms in Australian extensive grazing systems is of dubious value and may increase selection for drug resistance.

Also, the frequent use of endectocides such as moxidectin against cattle tick in areas such as north-eastern NSW and parts of Queensland may be a factor in the development of resistance in endoparasites. Route of administration of cattle anthelmintics may also be a factor. Cattle producers are encouraged to check drench efficacy by conducting a WEC 14 days after treatment.

Managing resistance

- Use drenches known to be effective based on a worm egg count reduction test (WECRT). Consider using combinations of unrelated drenches.

- Rotate between effective drenches or drench combinations from different drench families (within season/year, from one drench to the next).
- Use the correct dose using equipment known to be accurate. Calculate dose based on the heaviest animals in the mob.
- Follow the label.
- Avoid unnecessary drenching
 - especially of adults.
 - during droughts or prolonged dry spells.
 - immediately before or after moving sheep onto clean paddocks, for example un-grazed cereal stubbles or paddocks that have been sheep-free for extended periods.
- Avoid using long-acting drenches pre-lambing.
- Manage lambing paddocks after lambing. Resistant worms are likely to accumulate on the lambing paddock between lambing and weaning. If ewes were drenched pre-lambing, worms that were resistant to that drench will have accumulated on the lambing paddock after lambing, especially if a long-acting drench was used. After ewes and lambs have left the lambing paddock, dilute the resistant worms on this paddock by grazing with wormy sheep that have not been drenched recently.

New drenches

The beginning of September 2010 marked the arrival of spring as well as Australia's first new drench in 22 years. Ivermectin, the first of the macrocyclic lactone (ML) drenches, was released in 1988; now we have monepantel ('Zolvix', Novartis), representing the new drench group known as the AADs (amino-acetonitrile derivatives).

Pfizer has a new drench in the pipeline too. This one, called 'Startect', was released in July in New Zealand, which means we might see it in Australia in 12-18 months. Startect® is a combination of new and old. The new part is derquantel, representing the new action group, the spiroindoles. Derquantel is combined with the ML, abamectin.

So, where does a new drench fit? Do we use it or keep it on the shelf? And if we do use it, bearing in mind that we tend to break new drenches (drench resistance) after about five years, how do we use it?

We should use not shelve Zolvix, but use it well. Being new and highly effective, Zolvix will do a good job on worms in its own right,

but it can also keep the older drench actives alive for longer.

Zolvix should be used in rotation with older actives, and the evidence suggests rotation within a season has the edge on annual or longer rotations.

But this is where many producers will fail. A sensible system of drench rotations is not possible if the efficacy of the various drenches or drench combinations on a property is unknown.

Combinations should be considered. The consensus is that using a combination of unrelated broad-spectrum actives will raise efficacy and delay resistance more than using drench actives one at a time, unless severe resistance affects all the actives.

Use of combinations and rotation of drenches can go together. For example, ML-based triple (or quadruple) combination and an organophosphate (OP)-based combination may be options that still work well for many if not most producers. In this case the ML-based combination could be used, followed by the OP-based combination at the next drench, with Zolvix being the next drench in line.

Farmers may not wish to use combinations; that is their choice. Zolvix should at least be considered as part of a rotation of drenches known to be effective.

Extension – what's new

The stand-out development in the last several years has been the development of WormBoss (wormboss.com.au), a web-based extension tool and national repository of information regarding sustainable sheep worm management. WormBoss is a project supported by the Australian Sheep Industry Cooperative Research Centre and Australian Wool innovation. The WormBoss website still attracts a number of hits and pages opened above industry averages.

Conclusions

- Worms are high impact economic disease agents in our livestock industries.
- Like worms themselves, the economic cost of worms is largely invisible.
- Drug resistance is a very important issue for our grazing industries.
- Regular monitoring of worm burdens – and drench efficacy – is an important part of an integrated approach to worm management.
- New drench actives are welcome, but they are not the answer to sustainable worm control, and they should be managed well.

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Appendix

Table 1. Estimated prevalence of drench-resistant* sheep worms in Australia (modified from Love 2007)

Benzimidazole (BZ, 'white')	Approximately 90% of properties.
Levamisole (LEV, 'clear')	Approximately 80% of properties ('scour' worms**); resistance no longer rare in <i>Haemonchus</i> .
BZ + LEV combination	Approximately 60% of properties ('scour' worms).
Macrocyclic lactone (ML, 'ectin') (Avermectins (ivermectin, abamectin) and milbemycins (moxidectin))	Now common. 70+% of sheep farms in WA have ML (ivermectin)-resistant <i>Teladorsagia (Ostertagia)</i> . ML (ivermectin) -resistant <i>Haemonchus</i> in northern NSW and southern Queensland are common (70+% of farms; possibly 30+% of farms for moxidectin). ML-resistant <i>Teladorsagia (Ostertagia)</i> are thought to occur on 30+% of farms in southern NSW and other non-seasonal to winter rainfall areas of south-eastern Australia
Naphthalophos	Confirmed resistance rare. Two recorded cases in Australia (goats, Queensland) – Green et al. (1981) (<i>Haemonchus</i>), Le Jambre et al. (2005) (<i>Trichostrongylus</i>).
Closantel	Resistance in <i>Haemonchus</i> is common (80+% of farms in some areas) in northern NSW and SE Queensland. Many isolates are also ML-resistant. Small number of closantel-resistant strains of liver fluke in Australia.
Triclabendazole	Small number of resistant strains of liver fluke in Australia.

Notes: * Drench efficacy < 95 per cent. Prevalence of ML-resistance: these estimates refer to avermectin (ivermectin, abamectin) resistance. The prevalence of resistance to moxidectin, which is more potent, is currently lower (but approximately 30% of farms in the New England region of NSW (Nielsen and Bailey, unpublished data). In addition, Nielsen's and Bailey's assessment of the data suggests that, on the great majority of properties in the NSW Northern Tablelands, worm eggs are re-appearing somewhat sooner after treatments with moxidectin products than used to be the case. **Scour worms: mainly *Trichostrongylus* (black scour worm) and *Teladorsagia (Ostertagia)* spp (small brown stomach worm).

Sources: J Lloyd, D Palmer D, J Boray, GW Hutchinson, RB Besier, R Woodgate, R Nielsen (Veterinary Health Research, Armidale), JN Bailey, PI Veale, M Lyndal-Murphy and D Hucker (personal communications); Besier and Love (2003), Love 2007.

Table 2. Cattle worm resistance in New Zealand

	% farms with <95% FECR		
	Albendazole	Levamisole	Ivermectin
All species	76	6	92
<i>Ostertagia</i> sp	33	9	9
<i>Trichostrongylus</i> sp	14	0	5

For each anthelmintic and worm species, the number of farms ranges from 21 to 62. FECR: faecal egg count reduction.

All species: undifferentiated faecal worm egg count.

Table 3. Proportion of properties with resistance (Rendell, 2010)

	<i>Ostertagia ostertagi</i>	<i>Trichostrongylus</i> spp.	<i>Cooperia</i> spp.	At least one species
BZ	5/11 (45%)	4/7 (57%)	1/11 (9%)	7/13 (54%)
LEV	3/3 (100%)	0/3 (0%)	0/3 (0%)	3/3 (100%)
½IVM	8/10 (80%)	0/6 (0%)	7/10 (70%)	10/10 (100%)
Full IVM	5/11 (45%)	1/7 (14%)	6/11 (55%)	8/13 (62%)

Resistance defined as <95% WEC reduction and the lower 95% confidence limit was <90%. BZ – benzimidazole. LEV – levamisole. 1/2IVM – ivermectin at half dose (0.1mg/kg). IVM – ivermectin at full dose (0.2mg/kg).

Figure 1. Cost of endemic diseases in Australia

