Meat & Livestock Australia SUPER PIRD S2005/NO3

## Dung Beetles and Internal Parasites of Sheep

# **Final Report<sup>1</sup>**

Granite Borders Landcare Committee PO Box 400 Tenterfield NSW 2372 Ph / Fax 02 67363500



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#### I. Executive Summary

The project identified the dung beetles that attacked sheep faeces as being: native species *O. atrox*, *O. australis*, *O. capella*, *O. chepara*, *O. dandalu* and *O. granulatu*s; introduced *O. gazella*, *E. africanus*, *E. intermedius* and *O. pecuarius*. Dung ball rollers *Sisyphus sp.* also removed sheep dung.

The project originally intended to monitor dung beetle activity and then attempt to correlate this with worm egg counts in sheep grazing the monitored paddocks. This proved to difficult as many factors such as host immunity, age, etc. influence faecal egg counts. Instead, the project moved to estimate numbers of infective larvae developing in sheep faeces attacked by dung beetles compared with dung beetle free faeces. Early experiments with infective larvae were confounded by low numbers of parasites developing in either control or dung beetle attacked faeces.

A series of small experiments were undertaken to investigate larval culture techniques that both produced a large number of parasites but still could estimate the effect of dung beetles on the system. These studies developed a culture container that could reliably recover larvae from buried faeces and also estimate the amount of faeces being attacked by dung beetles. The project found that burying sheep faeces manually actually enhanced the amount of larvae recovered compared with faeces on the surface of soil when moisture was limited.

Larvae developing underground were found to stay there until there was sufficient moisture to allow them to move to the surface. If burying faeces by dung beetles was purely a mechanical movement of faeces, there would be a potential for larvae to develop underground and be sequestered there until rainfall enabled them to move to the surface. These sequestered larvae could represent a "time bomb" for sheep farmers by commencing infection of sheep immediately following rain, even when the sheep grazing the pastures were parasite free prior to rainfall. However, when dung beetles were included in the culture containers the "time bomb" effect was ruled out.

The action of dung beetles in burying faeces actually reduces the numbers of larvae obtained consistent with the total amount of faeces removed from the surface by the beetles. Dung beetles were found to remove approximately 1g of faeces/beetle from freshly deposited sheep dung. Thus dung beetle activity results in a reduction of parasitic larvae on pasture.

A field day will be held to communicate these results to farmers this Spring 2009.

### A. People directly involved with MLA SUPER PIRD S2005/NO3 Dung Beetles and Internal Parasites of Sheep

Andrew Biddle, Northern New England RLPB Jane Boyd, MLA Super PIRD S2005/N03 Jennie Coldham, Project Manager, Granite Borders Landcare Committee Jane Growns, MLA Super PIRD S2005/N03 Malcolm Knox, CSIRO Livestock Industries Leo Le Jambre, Consultant - MLA Super PIRD S2005/N03 Pam Wilson, Project Officer, Northern Tablelands Dung Beetle Express

#### Site monitors:

Cam Banks - Uralla Rob & John Chappell - Dundee L & W. Chapman - Bundarra/Guyra David Worsley - Nullamanna Jude Cox - Deepwater Sandra Smith - Mingoola NSW DPI - Wellingrove.

#### II. Introduction and background

The Northern Tablelands Dung Beetle Express was an initiative of Granite Borders Landcare Committee in partnership with the Southern New England Landcare Co-ordinating Committee, Northern New England and Armidale Rural Lands Protection Boards. It is supported by GLENRAC and GWYMAC Landcare organisations.

The original project commenced in late 1998 and was funded by the Natural Heritage Trust, Australian Geographic Society and the North West Catchment Management Committee. The project aimed to increase dung beetle activity across the Northern Tablelands from The Summit in Queensland to Walcha in New South Wales.

In order to achieve this 20 monitoring sites were established and the information collected was then used to select sites for beetle harvests and releases across the project area. The project was very successful and while a key focus is still increasing dung beetle activity and promoting the benefits of dung beetles, the Dung Beetle Express has undertaken several new projects.

The Bundaberg Rum Bush Fund financed a joint project between the Dung Beetle Express and the Malpas Catchment Committee in 2004. The project aimed to improve water quality by reducing nutrient runoff in the catchment. This project involved releasing colonies of dung beetle species which had not previously been located in this area.

The Dung Beetle Express then expanded on this work with a National Landcare Program funded project in conjunction with the Northern Rivers Catchment Management Authority. This project was primarily concerned with improving nutrient cycling by increasing beetle species abundance and richness in the catchment area. Dung burial reduces the amount of organic matter and nutrients entering river systems thus enhancing water quality.

Meat and Livestock Australia have supported the Dung Beetle Express by providing funding through a Producer Initiated Research and Development project (PIRD) and then a Super PIRD.

The Super PIRD funded an important project looking at the relationship between dung burial and internal parasites of sheep. Over the first twelve months of the project, the species of dung beetles utilizing sheep dung were identified and subsequent attention was given to studying worm survival. The questions addressed were whether worm eggs can survive burial and, if so, can larvae migrate to the soil surface where they could infect sheep? The answer to these questions are important to producers who may be using a rotational grazing system as part of an integrated pest management program as burial could prolong the viability of worm larvae.

This information regarding the project's directions were distributed over the internet.

#### III. Objectives

The project began with 5 goals (measurable outcomes):

- a) To investigate the extent of sheep dung burial by dung beetles
- b) To determine what species are utilising sheep dung
- c) To investigate any correlation between faecal egg count numbers and dung beetle activity
- d) To investigate the effect of drenching on dung beetle communities
- e) To introduce or redistribute suitable dung beetle species if results support taking this action.

#### IV. Methodology

#### Initial Methodology

Over the first twelve months of the project it was found that 12 species are utilising sheep dung, 6 introduced and 6 natives. It was difficult to quantify just how much dung was being buried, however, that at sites with good beetle activity an average of <sup>3</sup>/<sub>4</sub> of the pellets placed on the traps are buried or removed (ball rolling species being suspected in the case of removal).

Egg counts increased as beetle numbers rise – this was not considered significant in determining the effectiveness of dung burial in reducing worm burdens in sheep as conditions which favour dung beetles also favour the development and dispersal of worm larvae. However, it made the interpretation of the effectiveness of trying to correlate dung beetle activity with worm egg counts in sheep.

Consequently, while there was value in continuing the current methodology the project staff felt that more information could be gained by making some changes. Dr.Leo Le Jambre joined the project at this point as a consultant and helped design a larval recovery project designed to produce results that would be quantifiable and of significance to sheep producers.

The new methodology project will look at whether pellets buried by dung beetles retain viable larval numbers and whether larvae can migrate to the soil surface thus re-infecting pastures – a factor which may impact on producers using rotational grazing as part of an integrated pest management program.

#### New Methodology:

The project changed emphasis to concentrate on larvae on pasture rather than worm egg counts in sheep. The aim was to have several different pots in a laboratory situation and a few on landholder's properties. The location of the landholder trial would be determined using data on species recovered over the past twelve months.

#### V. Dung Beetles Utilizing Sheep Faeces:

Native beetles were *O. atrox*, *O. australis*, *O. capella*, *O. chepara*, *O. dandalu* and *O. granulatus*. Introduced: *O. gazella*, *E. africanus*, *E. intermedius* and *O. pecuarius* (*O. pecuarius* preferred the larger, clumpy dung rather than pellets). We know that introduced species either *Sisyphus rubrus* or *S. spinipes* (perhaps both) also attacked sheep dung but as they are dung ball rollers they removed themselves and the dung before proper identification at the species level could occur.



Onthaphagus granualatus – native dung beetle



Sisyphus spinipes – introduced ball rolling dung beetle





Euoniticellus africanus - introduced dung beetle

Onitis pecuarius – introduced dung beetle

#### **VI. Experiments**

### A. Experiment 1. Experiment to determine effect of dung beetles on larval development on landowners properties and under laboratory conditions

#### **Culture Containers**

Dung beetles and worm larvae were made to interact within the confines of pots filled with soil. These pots were filled with soil to a distance of 4-5cm from the rim and then a layer of turf was placed on top of the soil. It was considered that this would most closely mimic the conditions encountered by both by larvae developing in sheep faeces and by dung beetles feeding on the faeces. Then 15 g of fresh sheep faeces containing *Haemonchus contortus* eggs was placed on top of the soil. If the faeces were to be buried by hand, a hole was bored through the turf and the faeces placed down the hole.

The pot was then placed in a bucket and the bucket covered with cling wrap to help keep the faeces moist. A pot in a bucket and covered with cling wrap will henceforth be referred to as a "Culture container". (See Figures 1-5.) In the first experiment, the faeces were misted with 5ml of water at the commencement of the trial. The culture containers in the "on farm" study at Deepwater, Dundee, Nullamanna, Uralla and Bundarra/Guyra were left uncovered for 24 hours to allow dung beetles to fly onto the faeces (Figure 6). The number and treatment of culture containers in the laboratory trial were: 3 seeded with *O. gazella* (introduced) – 3 pairs of beetles added, 3 seeded with *O. australis* (native) – 3 pairs added, 3 seeded with *O. granulatus* (native) – 3 pairs added, 3 unburied and beetles excluded (control), 3 buried by hand at 10 – 15 cm and 3 buried by hand at 3-5 cm.



Figure 1. Culture Container with 15g of sheep faeces

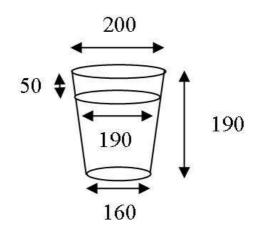


Figure 2. Dimensions of pot within bucket.



Figure 3. Jane Boyd burying sheep faeces in Culture Container



Figure 4. Jane Boyd releasing dung beetles into Culture Containers



Figure 5. Pam Wilson setting up cultures



Figure 6. Pam Wilson with co-operating landholder Rob Chappell, Dundee.

#### Collection of larvae

*Haemonchus* and other sheep parasitic nematode eggs in sheep faeces require favourable environmental conditions (moisture and warmth) to hatch. When these conditions are present, the eggs hatch to release first stage larvae. These feed on bacteria in the faeces, grow and molt to second stage larvae which also feed on bacteria.

The second stage larvae molt to form the third stage larvae which are the infective stage. However, the second molt is incomplete and the infective larvae remains encased in the second stage skin which prevents from feeding but provides protection against drying out. Once the third stage or infective larvae stops feeding they begin to wander and move through moisture films. Since they are also negatively geotropic they move out of the faeces and up films of moisture that occur on pasture plants due to either rain or dew. Consequently, the third stage larvae that developed from the faeces would move to the highest points on the surface of the pot containing the soil and then be collected for counting.

#### Haemonchus contortus infected sheep faeces

The *H. contortus* strain used to infect the sheep faeces in all of the experiments described in this report was the Kirby strain. This strain was originally isolated from the University of New England Kirby farm in 1987 and is susceptible to all anthelmintics. Kirby is passaged at CSIRO's FD McMaster Research Laboratory and the sheep faeces containing nematode eggs were provided by this laboratory.

### Results of trial including collaborators and laboratory MLA TRIAL - COMMENCED 28/2/07

TRIAL PLOT	NO.OF WORMS	Collected	Sheep	% Burial			Number of Larvae		ae		
			Cheep	70 <b>D</b> uniu	6/03/07	13/03/07		12/04/07			
	•		•	•							
Uralla S1	10,400 epg	28/02/07	377		8	143					
Uralla S2	10,400 epg	28/02/07			0	0					
Bundarra/Guyra S1	10,400 epg	28/02/07			(4 soil) 0	2	0	0		0	
Bundarra/Guyra S2	10.400 epg	28/02/07			0	0	0	0		0	
Dundee S1	10.400 epg	28/02/07		0	50	0	0		0	0	
Dundee S2	10,400 epg	28/02/07		60%	0	0	15		(5 soil) 12	0	
Nullammanna S1	10,400 epg	28/02/07			0	2	0	0		0	
Nullammanna S2	10,400 epg	28/02/07			0	0	0	0		0	
Deepwater S1	10,400 epg	28/02/07			0	10	4	0		0	
Deepwater S2	10,400 epg	28/02/07			0	0	0	0		0	
Operatural C4	00.000 and	07/00/07	070			0	0	[	0	0	0
Control S1	26,600 epg	27/02/07	376	0	0	0	0		0	0	0
Control S2	26,600 epg	27/02/07			90	0	82		10		0
Control S3	26,600 epg	27/02/07		0	0	0	0		0	0	0
0. gazella S1	26,600 epg	27/02/07		30%	0	0	0		0	0	64
0. gazella S2	26,600 epg	27/02/07		30%	0	0	7		5	0	36
0. gazella S3	26,600 epg	27/02/07		20%	180	0	9		74	0	0
0.australis S1	26,600 epg	27/02/07		35%	0	0	15		31	0	43
0.australis S2	26,600 epg	28/02/07		20%	0	0	0		0	0	5
0.australis S3	26,600 epg	28/02/07		20%	0	0	21		74	0	3
0.granulatus S1	26,600 epg	28/02/07		5-10%	0	0	0		0	0	0
0.granulatus S2	26,600 epg	28/02/07		30%	24	0	0		0	0	10
0.granulatus S3	26,600 epg	28/02/07		5-10%	0	0	0		0	0	0

TRIAL PLOT	NO. OF WORMS	Collected	Sheep	% Burial	Number of Larvae						
					6/03/07	13/03/07	20/03/07	23/03/07	27/03/07	3/04/07	12/04/07
3cm S1	7,800 epg	28/02/07	374	15%	0	0	4		7	0	24
3cm S2	7,800 epg	28/02/07		15%	0	0	0		0	0	0
3cm S3	7,800 epg	28/02/07		15%	0	0	10		4	0	0
12cm S1	7,800 epg	28/02/07		20%	96	2	0		2	0	13
12cm S2	7,800 epg	28/02/07		20%	0	0	0		10	0	70
12cm S2	7,800 epg	28/02/07		20%	0	0	2		5	0	15
Mixed S1	7,800 epg	28/02/07		85%	0	0	0		0	0	72
Mixed S2	7,800 epg	28/02/07		50%	0	0	0		5	0	23
Mixed S3	7,800 epg	28/02/07		50%	0	0	0		0	0	10

#### Results of farmer and laboratory based trials

The results of both the farmer and laboratory based trials indicated that either not enough larvae were surviving or were failing to be recovered determine the influence of dung beetles on larval development. The small numbers of larvae obtained in this trial did not allow for any meaningful comparisons between dung beetles or between buried or surface cultured faeces. Thus the next steps were to improve the conditions of culturing larvae and subsequently recovering them under culture conditions that would allow an interaction with dung beetles.

A series of small trials were conducted to determine the best way to collect larvae from pots. It was found that larvae did not move down the outside of the pot and that scraping the surface layer of soil from the pot and placing it in a funnel and flooding it with water, the larvae could be collected from the bottom of the funnel. The successful development of larvae depended on 5ml of water being added to each pot with surface faeces each day for a period of 3-5 days at the start of culturing. The counting of parasitic nematode larvae could be made easier if the soil was heated at  $70^{\circ}$ C for 3 days prior to loading the pots in the culture containers. This treatment killed the free-living soil nematodes which could be confused with the parasitic larvae.

#### B. Experiment 2: Larval development and survival in surface and buried faeces

#### Aims:

- 1. To compare the relative survival rates of larvae developing in sheep faeces placed on the soil surface with those developing in faeces buried under the soil.
- 2. To determine suitable conditions for incubating cultures out of doors.
- 3. To provide some experience for the Glen Innes team in the culture and collection of parasitic nematode larvae

#### Materials and Methods

Soil to be used in pots was heated to 70°C for 3 days prior to use, in order to kill soil nematodes and micro-predators. After heating, the soil was mixed with water to produce a damp, but not wet, consistency and then placed into the pots. The level of the soil was brought up to 0.5 cm from the top of the pot. Then 15 g of fresh sheep faeces was placed on top of the soil. If the faeces were to be buried, 4 cm of soil was added to the pots, then 15 g of faeces and then covered with the remaining soil to 0.5 cm from the top of the pot. The pot was then placed in a bucket and the bucket covered with cling wrap to help keep the faeces moist. A pot in a bucket and covered with cling wrap will henceforth be referred to as a "Culture container". The faeces in the containers were misted with 5 ml of water each day for five days. If the container had buried faeces, the surface of the soil was misted.

Armidale – One culture container with buried faeces was placed in a bucket and covered with cling wrap outside against a South facing wall with a roof shielding against the sun except in the early morning and late afternoon. A second two culture containers were incubated in the laboratory at 25°C. One had the faeces on the surface the other had buried faeces.

Glen Innes- Two culture containers were set up with 15g of faeces buried in the soil. One was placed outside in a similar position as the Armidale container and the other placed indoors.

After 7 days culturing the top 2cm of the soil in each culture container was scraped off and placed in cheesecloth. This was then suspended in a funnel or rain gauge and covered with water. Since larvae cannot swim, the V shaped container concentrated the larvae that crawled out of the soil. After 4 hours the supernatant was taken off and the larvae at the bottom of the funnel or rain gauge were ready to be counted.

If the culture container had buried faeces, the pot was then placed in one inch of water, which made the soil wet enough for the larvae at the bottom of the pot to move toward the surface. The top layer of soil was scraped off each day for a period of 3 days to ensure that all third stage

larvae that had managed to develop in the buried faeces had time to reach the surface. Each surface scraping was placed in water as above to concentrate the larvae prior to counting.

#### Results

Table 1. Numbers of larvae recovered from trials at Glen Innes and Armidale

Armidale

		Number of larvae collected					
	Indoor buried	Indoor surface	Outdoor buried				
Day 7	1725	7450	0				
Day 10	164400	300	0				
Day 11	450	0	0				

#### Glen Innes

	Numb	er of larvae collected
	Indoor buried	Outdoor buried
Day 7	1600	0
Day 9	1797	0
Day 10	2752	0
Day 11	200	0

#### Results and Discussion

It can be seen from the above Tables Culture Containers that get even a small amount of sun do not produce any larvae. This is most likely due to overheating in the closed container. Secondly, there was a very poor harvest of larvae from the indoor pot with 1mm of water being added to the surface faeces. However, the most impressive harvest of larvae occurred in the Culture Containers that had the buried faeces. Very few of these larvae moved to the surface until the soil became very moist and the larvae could move through the film of moisture surrounding the soil particles. The soil evidently provided sufficient moisture and protection from drying out to allow a high percentage of eggs to develop all the way to infective larvae. This is an important finding.

The action of dung beetles in burying the faeces could provide the nematode eggs with a safe and suitable environment to develop to infective larvae even when the conditions on the surface of the soil is unsuitable for larval development. Once the larvae developed under the soil they appear to stay put until conditions produce a film of moisture on the soil particles that would allow them to reach the surface. Consequently, dung beetles may be providing the parasites with a safe haven during dry periods and once the soil became very moist the larvae could then migrate to surface and infect sheep within days after a heavy rain. What is not known is whether the feeding and other activities of dung beetles kill the eggs during the process of dung burying. It is important to test the effect of burying by dung beetles as opposed to hand burying on larval development. It is also important to repeat this experiment with replicated samples so that the effect of burying can be tested statistically.

Important findings of work carried out 2007-2008

- 1. Sufficient moisture must be added to the Culture Containers to allow for larval development. One ml/day of water was not enough to ensure development of larvae even in cling wrap seal containers. Future studies will add 5ml over the entire soil surface to each container during the development phase of the larvae.
- 2. Culture Containers must be kept out of any direct sun. If the culture of larvae is carried outdoors the containers should be placed on the North side of a building or under trees.

- 3. Burying sheep faeces appears to protect developing eggs/larvae from desiccation and allows the parasites to reach infective stage even when there is not enough moisture to enable development on the soil surface.
- 4. Once larvae develop to the infective stage in buried faeces, they remain buried until there is sufficient moisture to provide a film of moisture over the soil particles for them to move to the soil surface.

The encouraging results of these parasite trials were communicated to MLA in a Progress Report.

#### C. Replicated trial to estimate the effects of burying sheep faeces on larval survival

#### Aims:

To compare the production of larvae in cultures with faeces placed on top of the soil with those with faeces buried in the soil with and without the addition of moisture.

#### Materials and Methods

The cultures were set up as described in the preceding experiment except that each culture was replicated 3 times. The cultures receiving rain were misted each day for 3 days to the equivalent of 5mm of water over the total surface area presented by the soil in the pot.

The larvae were harvested from the pots as described it the preceding experiment. Water equivalent to 20mm of rain was added to the pots containing buried faeces to enable the larvae to move to the surface.

#### Results

The cultures with buried faeces and surface faeces with rain produced similar numbers of larvae. Cultures with surface faeces and no rain produced significantly fewer larvae. These results are shown in Table 2 and Figure 7. An analysis of variance was carried out on the numbers of larvae recovered following the various treatments. The data was transformed to logs to the base 10 to stabilize the variance. The Bonferroni adjustment was used in the multiple comparison procedures to calculate an adjusted probability  $\alpha$  of comparison-wise type I error from the desired probability at the 0.05 level of family-wise type I error. The calculation guarantees that the use of the adjusted  $\alpha$  in pairwise comparisons keeps the actual probability of family-wise type I errors not higher than the desired level.

Table2. Total larvae recovered, no dung beetles

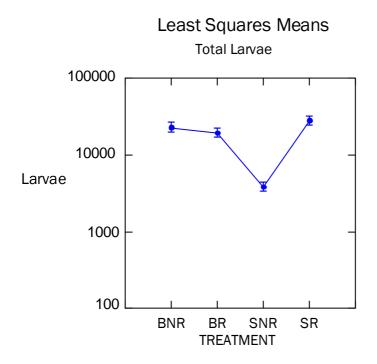
#### Analysis of Variance

Source	Sum-of-Squares	df	Mean-Square	F-ratio	Р
TREATMENT	1.410	3	0.470	40.335	0.000
Error	0.093	8	0.012		

Least squares means.

Treatment	Log Mean	Standard	Anti log	Number of reps
		error	mean	
Buried no rain (BNR)	4.359a	0.062	22856	3
Buried rain (BR)	4.291a	0.062	19543	3
Surface no rain (SNR)	3.585b	0.062	3846	3
Surface rain (SR)	4.448a	0.062	28054	3

Figure 7. The least square mean number of larvae harvested from the four treatments of sheep faeces. The abbreviations shown the Figure are: Buried no rain (BNR), Buried rain (BR), Surface no rain (SNR), Surface rain (SR).



#### **Discussion of Experiment C**

Burial of sheep faeces has no innate detrimental effect on the development and survival of *H. contortus* larvae compared to ideal conditions for larval development on the surface of soil. However, burial appears to enable more larvae to survive compared to surface faeces when rainfall is not optimum.

#### D. Replicated trial to estimate the effects of three species of dung beetles on larval survival

#### Aim:

To determine the effect of dung beetles on the development and survival of *H. contortus* larvae.

#### Material and Methods:

Initial weight of dung added to every pot was  $15 \text{ g} \pm 0.2$ . There were 4 replications in each treatment group. The culture technique was as described above except that dung beetles were added to some cultures. Netting was tied over the top of each pot to prevent the dung beetles from escaping. Pots were placed in buckets which were covered with cling film.

#### O. australis

2 males in each pot, replicates #1 and #2 had 6 females each; reps #3 and #4 had 7 females each

#### O. granulatus

Reps #1, #2, #3 had 1 male each; rep #4 had 2 males 3 females of this species were in each rep.

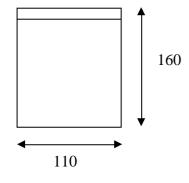
#### O. gazella

4 males in each rep Reps #1 and #2 had 6 females each, reps #3 and #4 had 7 females each

#### Timeline

27/2/09	experiment started
2/3/09	weighed dung remaining on surface, then gave 20 squirts of water
5/3/09	weighed dung remaining on surface, then gave 20 squirts of water
9/3/09	weighed dung remaining on surface, then scraped off top 1 cm of soil to collect nematodes, then added 550 ml (equivalent to 20 mm rain) using watering can
13/3/09	Scraped off next 1 cm of soil to collect nematodes. All species had some live beetles left, but most <i>O. gazella</i> were dead.

#### Collection of nematodes



Small mesh (old net curtain) bags with drawstrings were made up. Each was lined with a facial tissue before the soil was put in. The drawstring closed the top of the bag which was placed in the top of a rain gauge that was <sup>3</sup>/<sub>4</sub> full of water. Water was topped up to ensure all soil was under water. Samples were left for at least 4 hours before bag was taken out and most of water was removed by siphoning.

#### Results

There was a significant reduction in the number of larvae recovered from the culture containers that also contained dung beetles of all species. Burying produced the same number of larvae as did the surface faeces since surface faeces were watered as were the dung beetle pots. The data was transformed and analysed in the same way as the data in the previous experiment. The geometric and back transformed means are given in Table 3 along with the statistical analysis and are depicted in Figure 8.

Table 3. Statically analysis of total larvae recovered from dung beetle pots compared with dung beetle free pots.

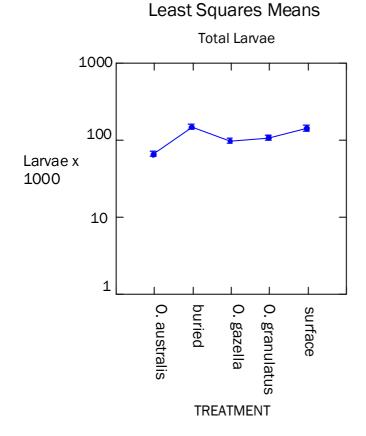
Source	Sum-of-Squares	df	Mean-Square	F-ratio	Р
TREATMENT	0.330	4	0.083	17.906	0.000
Error	0.069	15	0.005		

Least squares means.

Treatment	Log Mean	Standard	Anti log	Number of reps
		error	mean	
0. australis	4.711a	0.034	51404	4
Buried	5.066b	0.034	116413	4
0. gazella	4.882c	0.034	76208	4
0. granulatus	4.926bc	0.034	84333	4
Surface	5.047b	0.034	111429	4

Means with same letter following are similar.

Figure 8. The least square mean number of larvae harvested from the five treatments of sheep faeces.



There was a reduction in the amount of faeces left on the surface of the pot with dung beetles. The amount of dung left varied between the species of dung beetle as can be seen in Figure 9.

Figure 9. Weight of sheep faeces on the surface of control pots (no dung beetles) and culture containers that had dung beetles added to them. The weights were taken at day 6 from the start of culture.

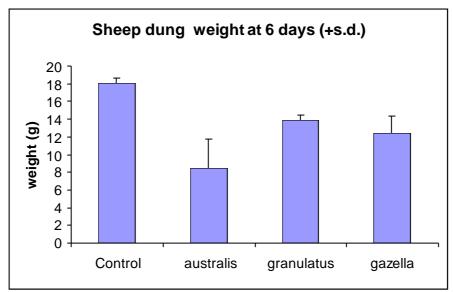


Table 4. The weights of faeces left on the surface of pots 6 days after culturing commenced. All of the beetle containing cultures had a decrease of faeces on the surface. The percentage reduction in weight is shown along with the expected number of larvae to be obtained from the culture if all the parasites in the missing faeces were destroyed.

6 days	Control	0. australis	0. granulatus	0. gazella
	18.7	12.3	13.9	11.2
	18.4	9.1	13.8	10.8
	17.7	8.1	13.4	15.1
	17.7	4.2	14.7	12.4
mean	18.1	8.4	14.0	12.4
s.d.	0.51	3.34	0.54	1.94
% reduction		54	23	32
Mean No. beetles		8	4	10
Dung removed/beetle (gram)		1.2	1.0	0.5
Mean larvae recovered	111429	51404	84333	76208
Expected if all dung		51795	92003	76079
buried killed eggs				

The reduction in larvae recovered in this experiment is what would be expected if the nematode eggs in proportion of faeces attacked by the three species of dung beetles were all killed (Table 4). Thus, since burial in itself has no detrimental effect, it appears that the action of dung beetles in shredding and chewing is lethal to parasite eggs and larvae.

#### VII. Discussion

#### Project Results

Bryan, 1973, found that at 100g of cattle of faeces per pair of *O. gazella* resulted in the faeces inside of the dung pat being reduced to a sawdust-like mixture. However, this destruction took place over a period of period of 160 days. Over this time, the addition of beetles to faecal pats at one pair per 100g faeces caused a reduction of 50% in the total larval recovery when compared with control pats with no beetles. Studying dung beetle activity in Durango Mexico Anduaga (2004) found that cattle dung pats were exploited mostly within the first 48 h after deposition. This author found that during the period of highest beetle activity, the mean amount of dung processed during 3 days of exposure was 39.01% (310.1 g fresh weight) of the 1 kg dung pats. At this time, traps caught approximately 2300 dung beetles in the cattle pats during the first day and less than 250 beetles visiting the dung on the second day. Consequently, the amount of dung being removed from the cattle dung pats was in line with that taken from the sheep dung in the present study.

A disadvantage to field studies reported in the literature is earthworms and other micropredators of dung were not excluded from the dung pats hence it is possible that amount of dung destroyed by dung beetles is somewhat inflated. A second disadvantage was that larval numbers were estimated by pasture samples and not by total larval counts that were obtained by collecting all of the larvae present in the Culture Containers used in the present studies. There is also a difference in the ecology of larvae developing in cattle dung pats compared with H. contortus larvae in sheep faecal pellets. Nematode larvae can remain in cattle dung pats for long periods. In dried out dung pats, rainfall of 50 to 100mm over a period of 2 or more days is required to facilitate migration. If moisture is not restrictive migration from cattle dung pats may take from 6 to 10 weeks before peak numbers of larvae appear on the surrounding herbage. Contributions to larval numbers on pasture can continue until the dung pats disintegrate and this may take up to one vear (Anderson et al., 1983). In contrast, nematode larvae in sheep faeces are not sequestered for such long periods. H. contortus larvae, in particular, leave the faecal pellets soon after developing to infective third stage. The reason for H. contortus larvae's quick exit from sheep faeces is that the larvae require a precipitation/evaporation ratio greater than 1 to develop (Anderson et al., 1978) Thus, the faecal pellets will be moist enough under these condition to provide a moisture film in which the larvae can move.

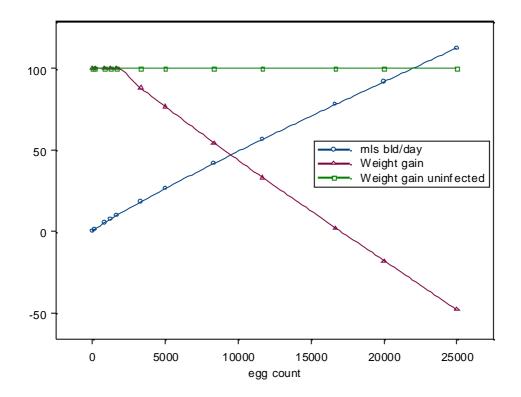
The results of the last two experiments demonstrate that burying sheep faeces can have a beneficial effect on numbers of larvae produced during a period lacking rain. Larvae developing under ground stay there until there is sufficient moisture to allow them to move to the surface. In the present study, the amount of moisture required was 20mm. If burying faeces by dung beetles was purely a mechanical movement of faeces, there would be a potential for larvae to develop underground and be sequestered there until rainfall enabled them to move to the surface. These sequestered larvae could have represented a "time bomb" for sheep farmers by commencing infection of sheep immediately following rain, even when the sheep grazing the pastures were parasite free prior to rainfall. However, the result of the trial including dung beetles in the culture containers has appeared to rule out the "time bomb" effect. The action of dung beetles in burying faeces removed from the surface by the beetles. Thus dung beetle activity results in a reduction of parasitic larvae on pasture.

Data from the present study indicates that all the larvae in the proportion of sheep faeces attacked by dung beetles are destroyed. The work carried out provided us with a means of quantifying the benefit of dung beetles in reducing parasitic larvae on pasture. The decrease in larvae developing on pasture is directly proportional to the weight of dung attacked by dung beetles. Thus if the beetles remove 50% of the dung being deposited on pasture there will be a 50% reduction in the numbers of parasite larvae developing from that dung. This reduction in pasture larval numbers cannot be directly related to parasite burdens in the sheep as there are many factors other than pasture larval numbers influencing infection levels. These include the age, condition, genetic constitution and previous exposure of the sheep.

#### VIII. Economic benefits of dung beetles

The effect of *H. contortus* infection on productivity of weaner lambs at pasture can best be predicted by haematocrits; for each further 0.01 proportional decrease in haematocrit there is a 0.03 reduction in live-weight gain (Albers et al, 1990). Haematocrit decreases can be determined from the daily blood loss of sheep (Albers and Le Jambre, 1983) and blood loss determined from faecal egg counts as demonstrated by Le Jambre, 1995. When this is calculated, the relationship is that shown in Figure 10 below. It is possible therefore to determine the effect that a given faecal egg count will have on liveweight gain. The graph emphasizes the value of diagnosis of parasitism based on faecal blood loss in order to identify and remove the infection before production loss occurs.

Figure 10. Displays the average daily weight gain of uninfected weaner lambs compared to the decrease in weight gain in weaner lambs with the egg counts shown on the x-axis. The blood loss in mls per day for a given egg count is also shown. Uninfected weaners are assumed to gain 100g/day



The worm world model (Barnes et al. 1995) estimates that susceptible sheep (lambs) have an average intake of pasture of 1kg/day dry matter and that the infection level in these sheep is (larval number/kg dry matter) X 0.5. *H. contortus* larvae commence sucking blood at around 11 days post infection but do not reach peak activity until 18-20 days post infection by which time egg laying has commenced. Even if farmers were to drench every 30 days with an effective anthelmintic it is quite possible for susceptible sheep to develop faecal egg counts of 5000eggs/g representing a loss of 25g/day in weight gain.

If lamb were worth \$5.00/kg that would equate to \$0.125/day/lamb lost to a *Haemonchus* infection that could easily be accumulated in 30 days. Lambs on pasture would produce approximately 1kg of faeces/day. Adult female *H. contortus* lay 10,000eggs/day; consequently the lambs' worm burdens would be 1000 worms including the male worms. Based on Barnes et al. a worm burden of 1000 worms after 30 days would represent lambs ingesting 66 larvae/day from pasture. At a stocking rate of 7 DSE/ha the lambs and their dams would be producing 12kg

of faeces per day. The present data suggests that 1 dung beetle would destroy all the worm eggs in 1g faeces. Therefore, 1000 dung beetles/ha would reduce the pasture infection level by 8%. Twelve thousand/ha would almost completely control nematodes on the pasture and would relegate haemonchosis to a rare phenomenon in the lambs.

Uncontrolled haemonchosis would kill approximately 20% of lambs and reduce weight gains by 25 – 50% in the survivors. All of the infective larvae would be developing from faeces being deposited on pasture. Considering that the value of lambs off 1ha of land is \$400.00 then uncontrolled haemonchosis would conservatively cost \$160.00. From this model then each 1000 dung beetles/ha has a value of \$13.00 to a sheep producing enterprise.

#### IX. Management changes to improve dung beetle habitat

Management is the key to increasing the number and variety of dung beetles and other beneficial insects that help break down dung. Since many properties in the New England Region run both sheep and cattle, both classes of livestock should be managed in a manner to improve the production of dung beetles. Dung beetle larvae are susceptible to some pesticides used for fly and internal parasite control for cattle and sheep.

Ivermectin (Ivomec and Doramectin) injectable, used at the recommended dose, reduced survival of the young of two species for 1 to 2 weeks in a study done in the USA (Fincher, 1996). Ivermectin pour-on reduced survival of the larvae for 1 to 3 weeks. Dung beetles feeding on dung from Australian cattle treated with an injection of avermectin at a therapeutic dose to control internal parasites, showed larval mortality, mortality of immature adults, reduced egg production, and inhibited ovariole development for periods of 1-4 weeks following treatment (Ridsdillsmith, 1993). Wardhaugh et al. (2001) found that faeces passed by sheep in Australia treated with controlled-release capsules (CRCs) of ivermectin precluded successful breeding by dung beetles in faeces collected up to 39 days after capsule administration. Newly-emerged *O taurus* also suffered significant mortality whereas those that survived underwent delayed sexual maturation. Ivermectin residues had no effect on the survival of sexually mature beetles, but reduced the fecundity of *O. taurus*. These authors concluded that ivermectin CRCs have the potential to cause substantial declines in beetle numbers, particularly if treatment coincides with spring emergence.

Specific chemicals aside, one must consider that any product designed to harm, limit, or kill would have some impact on the ecosystem in general, and should be used judiciously. Before treating your animals for internal parasites be sure that parasites are the problem. An egg count can help determine parasite load and whether the symptoms you may be seeing in the form of low gains, weight loss, unthriftiness, etc., are truly being caused by parasites.

Cell grazing systems can increase dung beetle populations and varieties by concentrating the manure in smaller areas, thus reducing the time beetles must spend in search of food. Grazing cycles that match the reproductive cycle of the beetles are favourable, as the livestock return to a grazing cell at the same time that new adults are emerging from the soil.

#### X. Objectives against project outcomes

- To investigate the extent of sheep dung burial by dung beetles The project found that dung beetles in the New England region will bury approximately 1g/beetle
- 2. To determine what species are utilising sheep dung
- Species attacking sheep faeces in the New England were: native beetles O. atrox, O. australis, O. capella, O. chepara, O. dandalu and O. granulatus; introduced O. gazella, E. africanus, E. intermedius and O. pecuarius. Dung ball rollers Sisyphus sp. also removed sheep dung.
- 3. To investigate any correlation between faecal egg count numbers and dung beetle activity This objective was changed to studying the effect of dung beetles on parasite larvae developing in sheep dung. The project found that beetles apparently destroyed all larvae in the dung that they attack.
- 4. To investigate the effect of drenching on dung beetle communities A trial to determine drench effects on dung beetles was not carried out in the project but a search of the literature indicates that macrocyclic lactone containing drenches can kill dung beetles.
- 5. To introduce or redistribute suitable dung beetle species if results support taking this action. Introduction and/or redistribution of dung beetles will be carried out under the auspices of the Dung Beetle Express if required by members.

#### XI. Acknowledgements

The project "partners" Granite Borders Landcare Committee Incorporated, Southern New England Landcare Limited, GWYMAC Inc and GLENRAC Inc, Armidale Rural Lands Protection Board and then the Northern New England RLPB are gratefully thanked for their part in providing office facilities, equipment and technical support to the project. Troy Kalinowsky, CSIRO Livestock Industries for providing *H. contortus* infected sheep faeces. Ian Colditz for analysing the data. Terry Boyd who set up the rain gauges for the trial, Maggie and Brian Hutton who provided the soil used in the culture pots. Gerald Martin, MLA Southern Producer Research Co-ordinator, for his helpful advice and patience during the course of this project. The Australian Meat and Livestock Authority's PIRD scheme is greatly appreciated for funding the project.

#### XII. Field days

Saturday, 13<sup>th</sup> September 2006 Topic – Can dung beetles reduce your worm burdens?

Friday, 6th October 2006

Held at the McMaster Laboratory, CSIRO Armidale Topic – Dung beetles and integrated pest management

#### Wednesday, 30th September 2009

#### Topic – Project Results and recommendations

A field day will be held to communicate the results obtained from this project to farmers. The data will be presented in concert with the Dung Beetle Express members who will demonstrate how to encourage dung beetles on properties.

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