

Supplementation of ewes on pasture to provide an epidemiological benefit for gastrointestinal parasitism

RD Tambunan^{1,2*}, CM Logan¹, AC Bywater¹ and AW Greer¹

¹*Faculty of Agriculture and Life Sciences, Lincoln University, PO Box 85084, Lincoln 7647, Christchurch, New Zealand*

²*Indonesian Agency for Agricultural Research and Development, Ministry of Agriculture of Indonesia*

*Corresponding author: Email: RenyDebora.Tambunan@lincolnuni.ac.nz

Abstract

The potential epidemiological benefit of reducing the peri-parturient relaxation in immunity to gastro-intestinal nematode parasites through supplementing twin-bearing ewes during the first four weeks of lactation was evaluated in a replicated farmlet study. In two sequential years, ewes either grazed pasture alone or grazed pasture while supplemented with 0.5 kgDM/d of a high-protein pellet. Supplementation did not affect ewe live weight or body condition score or weight of lamb weaned per ewe ($P > 0.05$). Ewe faecal egg counts (FEC) showed a time x supplementation interaction ($P < 0.05$), being reduced by 50% from week six of lactation in both years, although this only resulted in transient and inconsistent reductions in pasture larval contamination. After weaning, there was no consistent parasitological benefit to lambs grazing areas where ewes had been supplemented that were reflected in either pasture larvae concentrations, lamb FEC, the requirement for anthelmintic treatment or lamb growth rate ($P > 0.05$ for all). Despite supplementation of ewes during the first four weeks of lactation successfully reducing ewe faecal egg count by 50%, this was not sufficient to provide a measureable and consistent epidemiological benefit to the lambs.

Keywords: supplementation; sheep; peri-parturient; epidemiological; nematode

Introduction

Nematode parasites are a major animal health impediment for grazing ruminants around the world. Sheep farmers in New Zealand consider nematode parasites as their most significant animal health issue (Lawrence et al. 2007), and can cause significant losses in productivity and welfare if not prevented. Many control methods have been used to reduce the effect of parasite infection, including the use of anthelmintics. However, due to the development of anthelmintic resistance, alternative control options are needed.

A potential method of control is the nutritional supplementation of the ewe during the peri-parturient period (Beasley et al. 2012; Donaldson et al. 1998, 2001; Houdijk 2008; Kahn 2003). Donaldson et al. (1998) suggested that manipulation of nutrient supply to breeding ewes could potentially reduce larval contamination of pasture, especially for prolific flocks, as they are more vulnerable to breakdown of immunity. Moreover, because of lower milk intake their lambs will be forced to graze at a younger age and hence receive a greater larval challenge while experiencing a lower protein intake. However, most of these studies have only investigated the effects in either indoor studies or for a relatively short term. Consequently, the potential for an epidemiological benefit through reduced larval contamination for grazing lambs throughout the remainder of the season because of lower nematode egg excretion by the ewes has not yet been explored. Thus, this experiment aimed to evaluate the benefits of supplementing lactating ewes on pasture to reduce the peri-parturient relaxation in immunity (PPRI) to gastro-intestinal nematode parasitism and its provision of an epidemiological benefit to grazing lambs after weaning.

Materials and methods

Experimental design

The studies were conducted at summer-safe unit of LincolnSheep, Lincoln University, Canterbury, New Zealand over two sequential years (2015/2016 and 2016/2017) with the approval of the Lincoln University Animal Ethics Committee (approval #635 and 2016-25). Prior to lambing in 2015, twin-bearing crossbred ewes ($n=140$ in 2015) were randomly allocated to one of two farmlet treatments, *viz*, supplemented or not, and set-stocked in each replicate paddock. In 2015 each treatment was replicated on two paddocks, with a total of four paddocks (29-38 ewes/paddock, based on pasture availability). Ewes remained in their farmlets across both years and in 2016 ($n=128$) each treatment was replicated on four paddocks, with a total of eight paddocks (12-20 ewes/paddock, based on pasture availability).

Ewes in the supplemented treatment were given access to a commercially available high-protein sheep pellet (Farmlands stock feeds Ltd) through an Advantage Feeder (NGF800, Advantage Feeders Ltd, NZ). The pellet contained barley, wheat, soybean meal, peas, canola, wheat by-products, maize, oats and molasses with 25% crude protein and 12.8 MJ/kg dry matter (DM) of estimated metabolizable energy, as per product label. The feeder was initially restricted to supply 50 g/d/ewe three weeks prior to lambing and subsequently increased to 500 g/d/ewe during the first four weeks of lactation at which point the supplementation ceased. The amount of supplement consumed for each paddock was recorded and calculated as mean supplement intake per ewe. Amounts of the supplement were calculated to supply an additional 100 g of metabolizable protein (MP)/head/d assuming that unsupplemented ewes may consume 2 kg DM/day

with a total MP intake of 160 g/d (AFRC 1993). Thus, assuming no substitution, supplementation of 0.5 kgDM/d was calculated to increase total MP supply to about 260 g/d and a total DM intake of 3.5% of body weight. These supplementation rates were selected based on indoor studies which have indicated this MP supply may reduce worm burden in peri-parturient ewes by up to 50% (Donaldson *et al.* 2001). All ewes were allowed to graze on ryegrass/white clover pasture. Ewes remained in their respective paddocks until weaning at approximately 12 weeks after mean lambing date.

Live weight (LW), body condition score (BCS) and faecal egg counts (FEC) of all ewes were monitored at set stocking, tailing (approximately four weeks after lambing) and fortnightly thereafter until weaning. Faecal eggs were counted using the modified McMaster method (MAFF 1986) with a sensitivity of 100 eggs/gram (epg). Pasture grab samples were collected using a W-shape pattern from each paddock for the measurement of pasture parasite larval concentration fortnightly from set-stocking and infective third-stage (L3) larvae were recovered from the pasture using a modified Baermann technique (MAFF 1986). L3 present were then counted and morphologically differentiated from free-living larvae under a microscope. Two readings were performed for each sample and expressed as L_3/kg DM.

At weaning all lambs were drenched to remove residual parasite contamination and then exposed to a targeted selective treatment regime while grazing the areas in which ewes had been or not supplemented to determine if any epidemiological benefit of supplementation existed. To account for any potential carry-over effect, lambs (60 lambs/replicate in 2015 and 35 lambs/replicate in 2016) originating from each treatment were stratified across the treatment area, with each replicate ($n=2$, for 2015 and $n=3$, for 2016) consisting of 50% of lambs originating from a supplemented ewes and 50% of lambs originating from unsupplemented ewes. For each lamb replicate the potential for growth was assessed using sentinel lambs ($n=10$ in 2015 and $n=6$ in 2016) that were treated with a long-acting anthelmintic (1 ml/20 kg LW; Cydectin, Pfizer Animal Health, Auckland, NZ). Selection of these was based on placement when ranked hierarchically by LW. The remaining lambs in each replicate were subjected to a targeted selective treatment (TST) regime where the need for anthelmintic was based on animals achieving acceptable growth rates. In 2015, treatment thresholds were determined using Happy Factor Model (Greer *et al.* 2009) with the treatment threshold set to an efficiency of 0.74 (Greer *et al.* 2010). In 2016, treatment thresholds were set at 80% of the mean growth rate of sentinel lambs. Within each treatment and replicate lambs and ewes were rotationally grazed for the remainder of the grazing season with ewes following the lambs. The ewes were moved into a paddock on the day the lambs were moved out. To simulate on-farm conditions where lambs may be sent to slaughter, lambs were removed from the study once their body weight

exceeded 40 kg. Lambs were weighed fortnightly with the use of a swing-gate autodrafter (Prattely Industries Ltd) fitted with a tag reader. Any individual failing to reach their minimum target liveweight gain (LWG) was automatically drafted to one side, treated with anthelmintic and returned to graze with the remainder of the group. Faecal samples per rectum from the sentinel lambs and six TST lambs from each replicate were collected fortnightly.

After the completion of the first year, the paddocks were then grazed by the ewes for one more rotation before they were removed from the pastures and grazed on winter crops until being set-stocked for the 2016 study.

Statistical analysis

The LW and BCS of ewes, FEC and pasture larval counts data were analysed using the Restricted Maximum Likelihood (REML) using GENSTAT statistical package (16th Edition ver.16.1.10916, VSN International Ltd, UK). Other parameters were analysed using one-way analysis of variance (ANOVA) by Minitab statistical package (16Th Ed.). FEC and pasture larval counts were log transformed ($\log_{10}(\text{count} + 1)$) before analysis to obtain a normal distribution, and presented as back-transformed means. Where the F-test for treatment was significant ($P<0.05$), treatments were compared with a least significant differences test with a significance value of 5%. Due to the weight of conceptus at the start of lambing, change in ewe LW from four weeks after lambing only was assessed. The LWG, number of drench and LWG/drench of sentinel and TST lambs after weaning were analysed using one-way analysis of variance (ANOVA) by Minitab statistical package (16th Ed.).

Results

Ewes and lambs performance throughout lactation

The performance of ewes and lambs during lactation period of 2015 and 2016 is given in Table 1. Supplementation had no effect ($P>0.05$) on ewe BCS in both years and only on ewe mean LW in 2016 where unsupplemented ewes

Table 1 Effect of supplementation on ewe live weight, BCS, number of lambs weaned per ewe and weight of lamb weaned per ewe throughout lactation. Data represents mean \pm SEM.

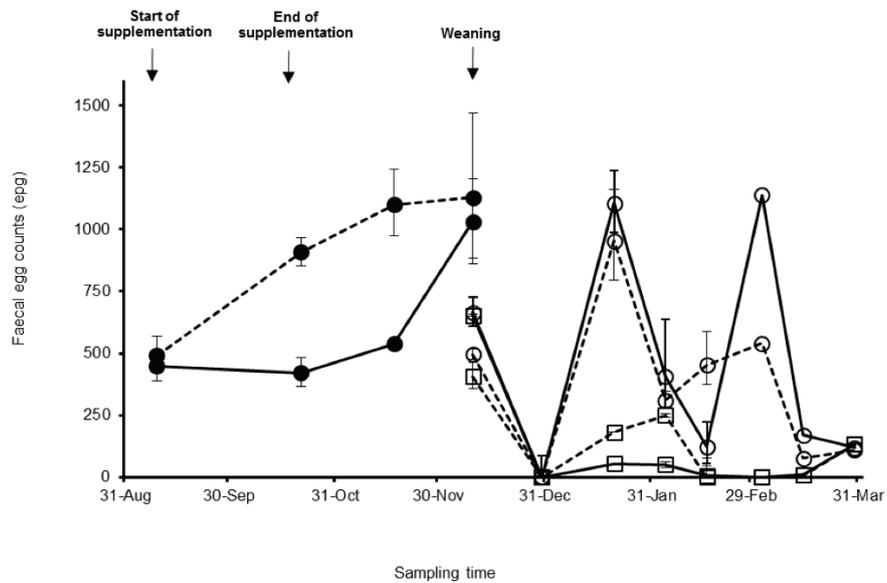
Parameter	Supplemented	Unsupplemented	P value
Mean live weight (kg)			
2015	60.30 \pm 0.59	62.20 \pm 0.77	0.18
2016	63.83 \pm 1.72	67.92 \pm 1.72	0.02
BCS			
2015	2.73 \pm 0.06	2.83 \pm 0.07	0.41
2016	3.06 \pm 0.08	3.11 \pm 0.08	0.56
Number of lambs weaned per ewe			
2015	1.69 \pm 0.14	1.56 \pm 0.05	0.47
2016	1.96 \pm 0.13	1.84 \pm 0.09	0.48
Weight of lamb weaned per ewe (kg)			
2015	34.88 \pm 3.98	31.77 \pm 0.89	0.53
2016	48.01 \pm 1.97	50.02 \pm 1.14	0.41

Table 2 The effect of supplementing ewes on pasture on the performance of their offspring after weaning. Data represents mean ± SEM.

Parameter	Sentinel lambs			TST lambs		
	Supplemented	Unsupplemented	P value	Supplemented	Unsupplemented	P value
LWG (g/d)						
2015	139.50±13.50	180.77±4.46	0.10	146.76±2.04	165.30±0.94	0.01
2016	142.39±3.66	135.78±5.86	0.39	150.74±4.08	147.71±4.27	0.64
Number of drench						
2015	-	-	-	3.03±0.01	3.20±0.04	0.04
2016	-	-	-	2.22±0.06	2.35±0.12	0.40
LWG/drench (g/d)						
2015	-	-	-	48.44±0.83	51.61±0.88	0.12
2016	-	-	-	81.97±6.08	77.78±4.00	0.59

Figure 1 Log 10-back-transformed means of faecal egg count (FEC) for ewes (closed symbols) that were supplemented (solid line) or not (dashed line) during the first four weeks of lactation and for lambs (open symbols) that were suppressively drench (squares) or exposed to a targeted selective treatment anthelmintic regime (circles) that subsequently grazed areas where ewes had been supplemented (solid line) or unsupplemented (dashed line) in: a) 2015; b) 2016.

a) 2015



b) 2016

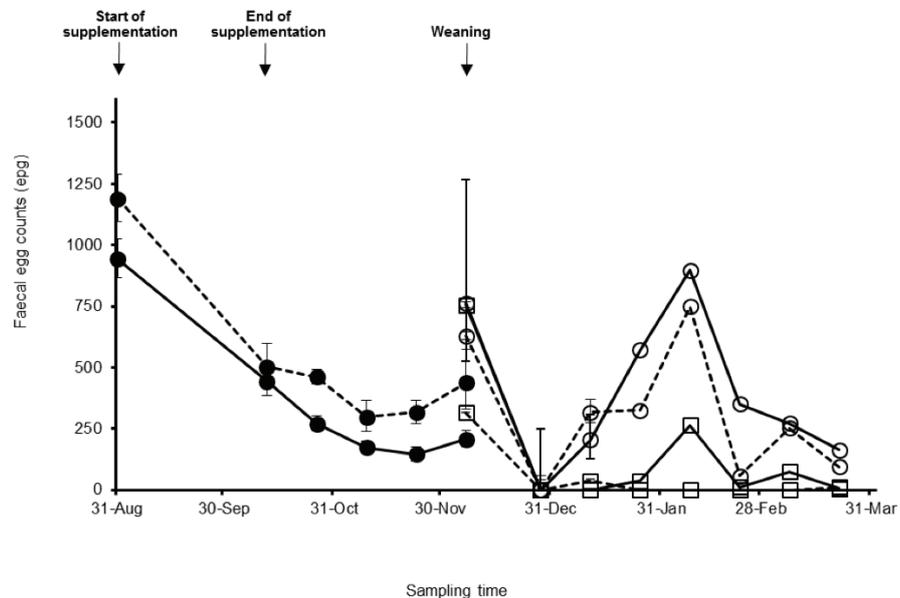
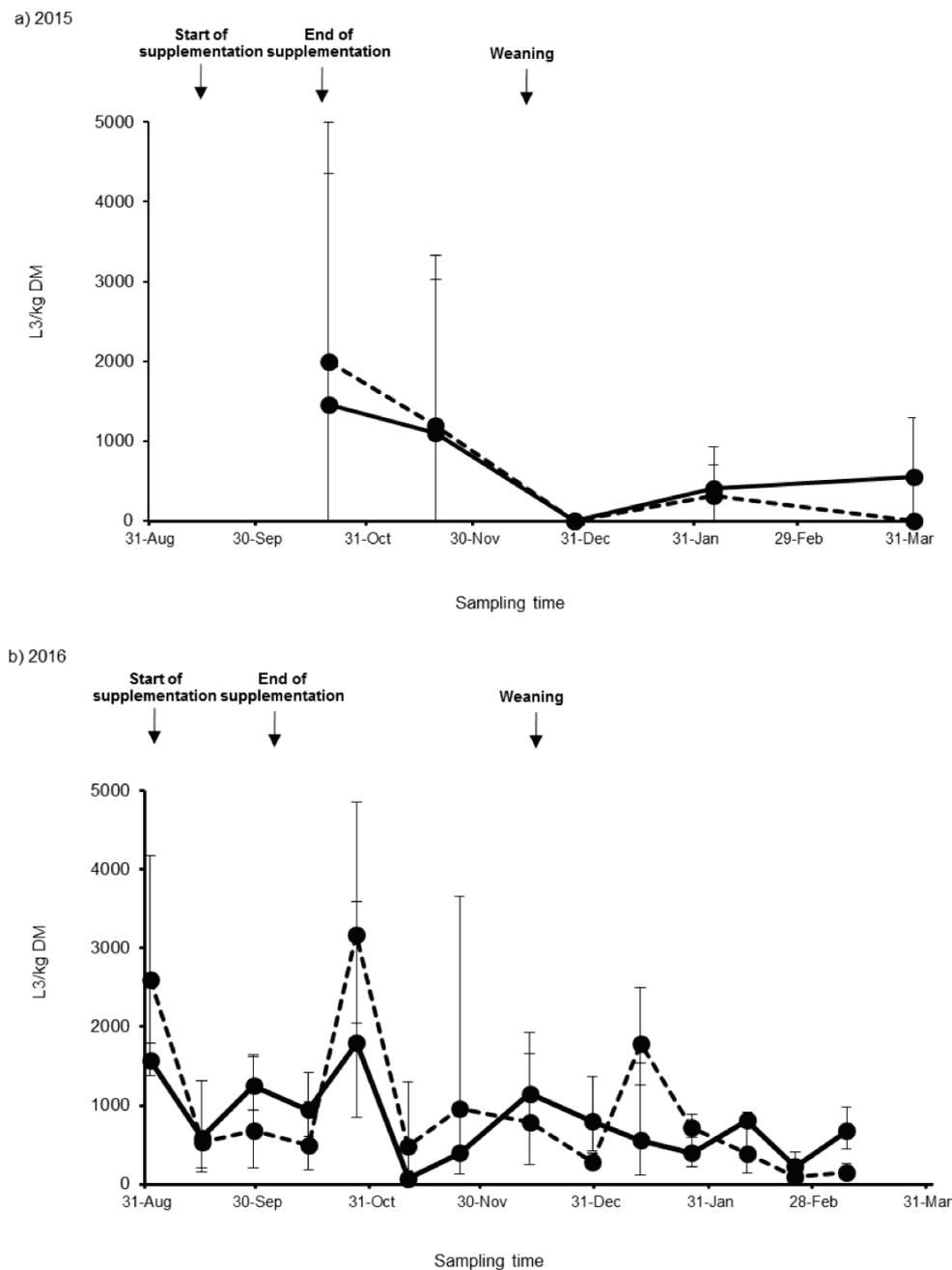


Figure 2 Log 10-back-transformed means of number of L3 larvae of strongyle nematodes per kgDM in paddocks grazed by the ewes and the lambs where ewes had been supplemented (solid line) or remained unsupplemented (dashed line) during the first four weeks of lactation in: a) 2015; b) 2016.



were heavier ($P=0.02$) than their counterparts. Additionally, supplementation had no effect on either the number of lambs weaned per ewe or the weight of lamb weaned per ewe during both years ($P>0.05$ for all).

Lamb performance after weaning

The performance of lambs after weaning that grazed areas where ewes had been supplemented or remained unsupplemented in 2015 and 2016, is given in Table 2. Overall, there were no differences ($P>0.05$) between the two treatment groups in all years, except for a greater LWG and a greater number of drenches administered per lamb of targeted selective treatment (TST) lambs in areas where ewes had not been supplemented in 2015.

Parasitological measurements: FEC and pasture larvae (L3)

There were no differences in parasitic load between farmlets and animals at the beginning of the study. Mean faecal egg counts of ewes and lambs in both years are given in Figure 1. For ewes in 2015, there was a time \times supplementation interaction ($P=0.04$), reflecting similar FEC at the start of lactation, that increased in unsupplemented, but not supplemented ewes in weeks six and eight ($P=0.03$) although were not different at weaning (week 12). In 2016 similar reductions in ewe FEC were observed, however the reduction continued until weaning. For lambs there were no effects of treatment ($P>0.05$) in FEC after weaning in both years.

The number of L3 larvae of strongyle nematodes (*Nematodirus spp* and other strongyles) present on pasture grazed by ewes and lambs before and after weaning are presented in Figure 2. Overall, there were no differences ($P>0.05$) in numbers of L3 larvae/kgDM on pastures grazed by the ewes and the lambs before and after weaning in both years.

Discussion

The primary objective was to evaluate the benefits of supplementing lactating ewes on pasture and its provision of an epidemiological parasite benefit to grazing lambs after weaning. The short period of targeted supplementation was chosen in an attempt to provide a cost-effective supplementation regime which provided additional nutrient supply to the ewes at the time of greatest need, such as until peak lactation. Overall, supplementation of lactating ewes in both years was successful in reducing ewe FEC, being a 50% reduction during periods of lactation. Although MP supply was not measured in this study, the observation of a 50% reduction in FEC is comparable with the 50% reduction in worm burden observed in parasitised lactating ewes relative to MP supply shown by Donaldson et al. (2001). However, in 2015 this effect was transient, as FEC of supplemented ewes increased and was not different at week 12 of lactation. In part, this increase in FEC by weaning was not expected as the demand for nutrients for lactation may be expected to be relatively low. However, climatic conditions were not favourable for pasture growth during this year, resulting in low pasture availability, which may have resulted in unintentional nutritional stress, with mean pasture mass declining to less than 700 kg DM/ha in all paddocks, below recommended levels (Corner-Thomas et al. 2015). In 2016, the reduction in ewe FEC in supplemented animals was more consistent throughout lactation indicating a longer-term benefit to the ewe as this extended beyond the pre-patent period of any larvae that would have been ingested post-supplementation. Nevertheless, while supplementation appeared to offer some benefit in reducing the peri-parturient relaxation in immunity to nematode parasites, the FEC in neither treatment in either year was reduced to low levels by weaning, possibly indicating that the nutritional stress caused by lactation extends well beyond peak lactation.

Given the only measured difference between the ewes groups and their performance was the reduction in FEC, it seems reasonable to suggest this reflects diversion of nutrients into immune function rather than increases in ewe body weight gain or lactation performance. This interpretation would be in-line with the nutrient partitioning framework suggested by Coop and Kyriazakis (1999), as the additional nutrients supplied by supplementation appeared to be utilised by the ewes for maintenance of immunity. This possibly reflects the nutritional cost of the immune response in lactating ewes and is supported by a lack of effect of supplementation on change in ewe live weight, ewe BCS or lamb production. The lack of an effect on

ewe LW or BCS or lamb weaning weight per ewe observed here is comparable to what has been reported in farm studies with interventions aimed at breaking the PPRI with long-acting drenches (Garland & Leathwick 2015; Miller et al. 2015). However, the design of the aforementioned studies was such that it did not allow an evaluation of the epidemiological benefit which may have accrued through interrupting the PPRI.

After weaning, supplementation of lactating ewes did not provide a clear benefit to lambs grazing the areas where ewes had been supplemented. Interpretation of the lamb LWG may be influenced by the potential differences between the pastures of each paddock, as such, comparisons between the TST and sentinel animals are preferred as these grazed the same areas. This aside, those lambs grazing areas on which ewes were supplemented did appear to have a lesser need for drench, although this only occurred in 2015. This indicates a small epidemiological advantage may have been conferred through the supplementation of ewes, although such effects were transient and were not great enough to result in a consistent difference in pasture larval contamination. Further, this apparent benefit is relatively low given the extent of the difference in FEC in the ewes for much of the lactation period. In part, this may reflect the design of the study, whereby the ewes followed the lambs, to mimic grazing practices on-farm. This may have contributed to the lack of effect due to the net removal of parasites by grazing non-lactating ewes (Leathwick et al. 2008). This may have been further exacerbated due to the low pasture availability in 2015 resulting in ewes grazing further down in the sward where a majority of the parasite population is believed to exist (Vlassoff 1982; Gazda et al. 2009). Alternatively, the lack of benefit may reflect relatively low transmission of disease from contamination supplied by lactating ewes. In the current study, pasture larval concentrations generally reduced during lactation despite the ewe FEC indicating a reasonable number of nematode eggs were being deposited. Nematode egg viability has been shown to be influenced by immune mechanisms of the host, with eggs from peri-parturient animals having a lower viability (Jørgensen et al. 1998), an effect which may be compounded by the relatively low egg development that has been reported during cooler periods of winter and early spring (Leathwick et al. 2011; Waghorn et al. 2011). Nevertheless, the results of the current study indicate that a 50% reduction in the FEC of lactating ewes is not sufficient under these conditions to result in an epidemiological advantage to the grazing lamb.

Conclusion

Supplementation of ewes during the first four weeks of lactation had no effect on ewe performance but was successful in temporarily reducing faecal egg by 50%, presumably reflecting better maintenance of immune function through greater nutrient supply. However, this reduction in parasite contamination was not sufficient to provide a measureable and consistent epidemiological

benefit to the grazing lambs that may assist with parasite control. It is suggested that for strategies to help break the parasite lifecycle through targeting the relaxation in immunity in the peri-parturient ewe, a reduction in ewe faecal egg count by more than 50% is required.

Acknowledgements

The authors acknowledge the technical support given by the staff of the Johnstone Memorial Laboratory (JML), Lincoln University. In addition, the authors thank Farmlands for financial support and supply of the high-protein pellet.

References

- Agricultural and Food Research Council 1993. Energy and protein requirements of ruminants. An advisory manual prepared by the AFRC Technical Committee on Responses to Nutrients. CAB International, Wallingford.
- Beasley AM, Kahn LP, Windon RG 2012. The influence of reproductive physiology and nutrient supply on the peri-parturient relaxation of immunity to the gastrointestinal nematode *Trichostrongylus colubriformis* in Merino ewes. *Veterinary Parasitology* 188: 306-324.
- Coop RL, Kyriazakis I 1999. Nutrition–parasite interaction. *Veterinary Parasitology* 84: 187-204. doi 10.1016/S0304-4017(99)00070-9.
- Corner-Thomas RA, Hickson RE, Morris ST, Back PJ, Ridler AL, Stafford KJ, Kenyon PR 2015. Effects of body condition score and nutrition in lactation on twin-bearing ewe and lamb performance to weaning. *New Zealand Journal of Agricultural Research* 58(2): 156-169, doi: 10.1080/00288233.2014.987401.
- Donaldson J, van Houtert MFJ, Sykes AR 1998. The effect of nutrition on the peri-parturient parasite status of mature ewes. *Animal Science* 67: 523-533.
- Donaldson J, van Houtert MFJ, Sykes AR 2001. The effect of dietary fish-meal supplementation on parasite burdens of peri-parturient sheep. *Animal Science* 72: 149-158.
- Garland CB, Leathwick DM 2015. A cost-benefit analysis of pre- and post-lambing drench treatments to twin-bearing ewes on commercial farms in the southern North Island of New Zealand. *New Zealand Veterinary Journal* 63(4): 220-226. doi 10.1080/00480169.2015.1012133.
- Gazda TL, Piazzetta RG, Dittrich JR, Monteiro ALG, Thomas-Soccol V 2009. Distribution of nematode larvae of sheep in tropical pasture plants. *Small Ruminant Research* 82: 94-98. doi: 10.1016/j.smallrumres.2009.02.004.
- Greer AW, Kenyon F, Bartley DJ, Jackson EB, Gordon Y, Donnan AA, McBean DW, Jackson F 2009. Development and field evaluation of a decision support model for anthelmintic treatments as part of a targeted selective treatment (TST) regime in lambs. *Veterinary Parasitology* 164(1): 12-20.
- Greer AW, McAnulty RW, Logan CM, Hoskin SO 2010. Suitability of the Happy Factor decision support model as part of targeted selective anthelmintic treatment in Coopworth sheep. *Proceedings of the New Zealand Society of Animal Production* 70: 212-216.
- Houdijk JGM 2008. Influence of peri-parturient nutritional demand on resistance to parasites in livestock. *Parasite Immunology* 30(2): 113-121.
- Jørgensen LT, Leathwick DM, Charleston WAG, Godfrey PL, Vlassoff A, Sutherland IA 1998. Variation between hosts in the developmental success of the free-living stages of trichostrongyle infections of sheep. *International Journal of Parasitology*: 1347-1352.
- Kahn L 2003. Regulation of the resistance and resilience of peri-parturient ewes to infection with gastrointestinal nematode parasites by dietary supplementation. *Australian Journal of Experimental Agriculture* 43(12): 1477-1485.
- Lawrence KE, Leathwick DM, Rhodes AP, Jackson R, Heuer C, Pomroy WE, West DM, Waghorn TS, Moffat JR 2007. Management of gastrointestinal nematode parasites on sheep farms in New Zealand. *New Zealand Veterinary Journal* 55: 28-234.
- Leathwick DM, Miller CM, Atkinson DS, Haack NA, Waghorn TS, Oliver AM 2008. Managing anthelmintic resistance: untreated adult ewes as a source of unselected parasites, and their role in reducing parasite populations. *New Zealand Veterinary Journal* 56: 184-195.
- Leathwick DM, Miller CM, Waghorn TS 2011. Development and spatial distribution of the free-living stages of *Teladorsagia circumcincta* and *Trichostrongylus colubriformis* on pasture: A pilot study. *New Zealand Veterinary Journal* 59: 272-278.
- MAFF 1986. Manual of veterinary parasitological laboratory techniques (3rded.). London, UK: Her Majesty's Stationary Office (HMSO). Pg. 7-9.
- Miller CM, Ganesh S, Garland CB, Leathwick DM 2015. Production benefits from pre- and post-lambing anthelmintic treatment of ewes on commercial farm in the southern North Island of New Zealand. *New Zealand Veterinary Journal* 63(4): 211-219. doi 10.1080/00480169.2015.1007108.
- Vlassoff A 1982. Biology and population dynamics of the free living stages of gastrointestinal nematodes of sheep. In: Ross AD (ed). *Control of Internal Parasites of Sheep- an Animal Industries Workshop*. Lincoln College, Lincoln. Pg. 8-12.
- Waghorn TS, Reynecke DP, Oliver A-MB, Miller CM, Vlassoff A, Koolaard JP, Leathwick DM 2011. Dynamics of the free-living stages of sheep intestinal parasites on pasture in the North Island of New Zealand. 1. Patterns of seasonal development. *New Zealand Veterinary Journal* 59: 279-286.