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The Effect of Management Strategies on Haemonchus Contortus Infections of Sheep and Goats on Intensively Grazed Pasture

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THE EFFECT OF MANAGEMENT STRATEGIES ON HAEMONCHUS CONTORTUS INFECTIONS OF SHEEP AND GOATS ON INTESIVELY GRAZED PASTURE

by

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The Effect of Management Strategies on *Haemonchus contortus* Infections of Sheep and Goats on Intensively Grazed Pasture

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Abstract

The purpose of this study was to determine how the management strategies used by producers on sheep and goat farms in Maine, New Hampshire, and Vermont could affect the fecal egg counts of the parasite called *Haemonchus contortus* in their animals. Surveys about management practices and fecal samples from the juvenile and adult populations were collected during the months of June, July, and August in the summer of 2015. Next, the samples were analyzed and fecal egg counts were determined using the McMaster method and a fluorescence-based *H. contortus* speciation technique. The fecal egg counts and survey answers were compiled and Analysis of Variance tests were used to determine significance between groups.

The results of this study suggest that particular management practices are more effective than others at controlling the proliferation of *H. contortus* on farms. Specifically, the use of chemical dewormers rather than natural dewormers led to lower fecal egg counts (P=0.078). In addition, farms with larger numbers of animals tended to have lower fecal egg counts (P=0.091). Other factors, such as the use of FAMACHA scores for the replacement of animals or the geographic locations of the farms did not prove to have a significant effect (P=0.75 and 0.97, respectively). The results of this study can be used to teach producers about realistic ways to control *H. contortus* in their flocks and lead to healthier, more productive populations.

Acknowledgements

First of all, the author would like to thank Dr. Jim Weber for his assistance with the project idea, the collection of data, and the writing of this paper. Ann Bryant must also be thanked for her tireless efforts in determining the fecal egg counts and recording them in preparation for this project. The author would also like to thank the producers throughout Maine, New Hampshire, and Vermont who volunteered to fill out the survey and send in the samples. Without their help, this study could never have been conducted. It is also important to thank the members of the Cooperative Extension who helped with the distribution of the surveys.

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Introduction

Infection by *Haemonchus contortus*, a parasitic nematode that resides in the abomasum, can have a devastating effect on sheep and goat populations in the United States. *Haemonchus contortus* attaches to the wall of the abomasum and causes hemorrhage, which can then lead to anemia and hypoproteinemia (Ortolani et al., 2013). This dangerous parasite is estimated to cost countries several million dollars in losses per year, and although it is normally a sub-tropical species, it is increasingly becoming a major problem in the northern United States. After its introduction to colder regions, *H. contortus* has been able to adapt and become endemic in northern climates despite the harsh winters (Assenza et al., 2014).

One of the most concerning factors about *H. contortus* pathogenicity is the fact that the parasite is able to quickly develop resistance to anthelmintics. Anthelmintics are a type of medicine used to expel or destroy parasitic worms, particularly those of the intestine. The first reports of resistance in *H. contortus* to anthelmintics occurred due to the increased use of phenothiazine from the 1950s to the 1960s. In 1961, thiabendazole was introduced, which was originally heralded for its efficiency and wide range of effectiveness. However, after just a few years the same pattern repeated itself and reports of *H. contortus* resistance began to appear (Kaplan, 2004).

Therefore, research is beginning to move towards the development of non-chemical methods for preventing the spread and infection of *H. contortus* in small ruminant populations throughout the country (Kaplan, 2004). Most of these methods

involve a reliance on environmental factors such as pasture rotation (Ortolani et al., 2013). If a producer practices pasture rotation, this means that he or she moves the sheep or goats between different pastures throughout the summer to prevent the animals from staying on infected pasture for too long. Management strategies for pasture rotation would ideally involve several grazing areas. However, it is often difficult to properly time the rotation between pastures for optimal parasite control and forage quality. Although long periods of rest are ideal for ridding the fields of parasites, this will often allow the grass or forage to grow too long (Tritschler). On many New England farms, the pastures take about 2 to 6 weeks to fully recover from intensive grazing (Livestock Grazing and Stocking Rates). Therefore, one proposed system involves a grazing period, a rest period, a hay harvest, another rest period, and then back to the beginning with a grazing period. Unfortunately, most small ruminant producers in New England do not have the resources for this system. It is also important to note that sheep often prefer pasture with approximately 4-inch high grass. However, parasites tend to thrive on the moisture from the lower part of the forage, and therefore if the sheep are forced to graze on taller, woodier plants, they may ingest fewer parasites (Tritschler).

Recent studies have shown that implementing a rotational grazing system can decrease the levels of nematode infections in sheep by breaking the life cycle of the parasites at the L3 stage (Colvin et al., 2008). The third-stage larvae (L3) of *H. contortus* is the larval form that persists on pasture grass and is capable of infecting sheep during grazing. When ingested, it soon travels to the abomasum of the host, growing into the fourth larval stage (L4) and then adult stage. Warm conditions tend to promote better *H. contortus* growth, but the L4 larvae can also go through an "arrested development" period

within the sheep abomasum during droughts or cold weather. This process, called hypobiosis, causes serious problems for sheep producers during lambing season in the spring, when the *H. contortus* complete their development and begin to cause anemia in the ewes (Laing et al., 2013).

In a study conducted in 2009, Katahdin lambs were rotated through several grazing pastures containing either rotational bermudagrass (no grass height specified) rotational short bermudagrass (forage specifically measured at below 10 cm), or continuous bermudagrass (no height specified, no rotation between pastures). Although the results were clouded by the multitude of variables in the study, the lambs that grazed on continuous bermudagrass required more deworming than those that grazed on rotational bermudagrass (Burke et al., 2009). Another study demonstrated that rotational grazing on pastures led to lower *H. contortus* egg counts, and fewer sheep required chemical dewormers during the course of this study. Host resistance in the sheep was also explored during the trial and although the results demonstrate that rotational grazing led to lower larvae levels in the grass, it also decreased the resistance of the sheep due to lessened exposure to the parasite (Walkden-Brown et al., 2013).

In the past, the primary treatment for *H. contortus* infections was dosing all of the animals in a flock with non-specific anthelmintics every three to four weeks to prevent this life cycle from continuing. However, when widespread resistance began to occur over the past ten years, new methods were developed for parasite control. One of the most prominent techniques is FAMACHA scoring, which relies on a careful examination of the color of the mucosal surface of a sheep's lower eyelid to determine its anemia status. Although FAMACHA testing requires significant training, it is becoming quite

popular in the United States (Terrill et al., 2012). Other trials have tested the efficiency of feeding "nematode-trapping fungi" such as *Duddingtonia flagrans* in an effort to eliminate larvae and prevent the spread of *H. contortus* parasites (Fontenot et al., 2003).

Another new method of parasite control is the feeding of copper oxide wire particles (COWP). Male lambs treated with COWP had decreased fecal egg counts (from a mean of 8000 eggs/g to 250 eggs/g, $P < 0.001$). In addition, most of the nematodes that remained in the abomasum were males, which reduced their ability to reproduce. Although the copper levels in the liver of these lambs remained within the normal range during the trial, COWP might not be a viable option in locations where the soil already contains high levels of copper due to its toxicity in sheep (Burke et al., 2004). The possibility of developing vaccines to protect against *H. contortus* infections has also been considered and numerous experiments have been conducted on this topic, but the results are inconsistent (Terrill et al., 2012).

Therefore, the purpose of this study was to use survey and fecal egg count data to determine the most effective non-anthelmintic management strategies for sheep producers in order to mitigate the harmful effects of *H. contortus* on flocks. A similar study was conducted in Canada from 2009 to 2012, which used field studies, surveys, and egg counts to determine that almost all of the sheep flocks of Ontario possessed high levels of resistance to Ivermectin and Fenbendazole (Peregine et al., 2012). When the survey results were compared to the fecal egg counts, researchers observed mixed results regarding FAMACHA, drench tactics, and grazing procedures for the sheep of Ontario (Falzon et al., 2013). Based on the results of this New England survey, recommendations can be made to sheep producers in Maine and thousands of sheep can potentially be

saved through the implementation of the most effective methods of parasite control.

Materials and Methods

During the summer of 2015, New England sheep and goat producers were invited to participate in a study on parasitology in small ruminants. The invitations for the study were issued using the Cooperative Extension databases in Maine, New Hampshire, and Vermont. There was no selection criteria based on the size of the farm or the breed of sheep or goat produced. The study required two-part involvement by the producers, and participation was voluntary. Producers were first asked to fill out an extensive survey with various questions on pasture management, veterinarian influence, deworming methods, and handling practices (Appendix 1). The surveys were then returned to the university and the survey answers were transferred to an electronic database.

Next, the sheep and goat breeders were asked to obtain fresh fecal samples from their animals. The specifications requested that they attempt to make them representative samples by selecting pellets from a wide variety of their sheep and goats (five to ten animals). The processing of the pellets for shipment followed the procedure developed by Wadsworth (2015). The producers were asked to cut a square foot of plastic wrap and use this to tightly package the feces, allowing as little air as possible to remain in the bundle. The ends of the plastic wrap were twisted and tied to prevent airflow because the maintenance of an anaerobic environment has been shown to inhibit the hatching of the nematodes. The producers were next instructed to wrap paper towels around the plastic wrap and place the package in a re-sealable sandwich bag. They were also asked to mail

the samples immediately and refrain from refrigerating the samples (Wadsworth, 2015).

The samples were received by Dr. Weber's parasitology lab at the University of Maine. A modified McMaster egg count technique and a fluorescence-based *H. contortus* speciation technique were used to analyze the samples to determine the fecal egg counts (FEC) (Wadsworth, 2015) (Appendices 2 and 3). Although abomasal worms such as *H. contortus* were the main focus of this study, eggs from various other parasites were also identified and enumerated during the processing of the samples. The egg counts from all of the samples were also organized into a separate electronic database. Data from these farms was used in subsequent analyses.

Producers were asked to send fecal samples from early summer, mid-summer, and late summer. For the June sample, the first sample was received on June 8, 2015, and the last sample was received on July 2, 2015. For the July sample, the first sample was received on July 15, 2015, and the last sample was received on July 31, 2015. For the August sample, the first sample was received on August 25, 2015, and the last sample was received on September 15, 2015. The samples were analyzed in the same manner, so each participating farm should have had three data sets for fecal egg counts for the summer.

The next step of the analysis, and the first part of the current project, was to organize the three fecal egg counts and survey data for each farm into a Microsoft Excel (2011) database. In total, 130 farms in the Northeast completed the survey. Each of the farms was given its own row, with separate columns for each of the survey answers (Rows A-X). The *H. contortus* fecal egg counts were then organized into two groups for

each farm. Since most farms sent multiple pooled fecal samples from groups of adult ewes, adult rams, yearling lambs, newborn lambs, mixed lambs and ewes, etc. and some farms had both sheep and goats, efforts were made to have the greatest amount of consistent data. In general, the first fecal egg count group (Group 1) was from the largest population of adult small ruminants on the farm, usually the adult sheep (males and females combined) or just the adult ewes. In cases where there were both sheep and goat populations, the population with the largest amount of animals was selected and their fecal egg counts were recorded for Group 1. The Group 1 fecal egg counts were recorded separately for each farm for June, July, and August, and any month that did not have results was left blank. The second group (Group 2) from each farm consisted of the largest group of juveniles (whether lambs or kids). The same process was repeated for June, July, and August with Group 2.

After the fecal egg count data was entered, 39 farms had completed the survey but had not submitted any fecal matter for analysis. These farms were removed from the analyzed data pool. Next, ten more farms were removed from the data pool due to concerns about the accuracy and consistency of the data. Most commonly, the farms were removed because their fecal samples were taken from changing groups throughout the summer, so the populations and their fecal egg count levels could not be accurately compared on a monthly basis. In total, 81 farms had complete survey results and fecal egg count results for at least one month of the summer.

Next, the fecal egg counts were compared to the survey results to determine any correlation between the *H. contortus* fecal egg counts and management practices in northern New England. Analysis of Variance tests were completed for the data, as well as

a comparison of means and standard deviation. These tests were conducted with the use of the StatPlus program for Mac, as well as Minitab Express (2015). The test results were organized with the use of data tables, written descriptions, and various graphs.

After the Analysis of Variance tests were conducted on the data, the mean values for each of the groups were calculated and compared. The *P*-values were also calculated, and these were analyzed to determine the level of significance. *P*-values below 0.05 indicated a rejection of the null hypothesis and were found to be statistically significant at the critical alpha level. An ANOVA test with a *P*-value of greater than 0.05 indicated that any difference between the groups was most likely due to random error, and therefore the null hypothesis was not rejected and no statistical significance was found.

Results and Discussion

Overall statistics

Although producers from Maine, New Hampshire, and Vermont were all contacted and invited to participate in the study, the data showed that nearly half of the analyzed data came from Maine farms (Figure I). This could be due to the fact that there is a lot more acreage in Maine than in the neighboring states, and therefore there are more sheep and goat farms. It could also be due to the fact that producers in Maine might have been more likely to hear about the research study and be more inclined to participate due to their closer proximity to the University of Maine campus.

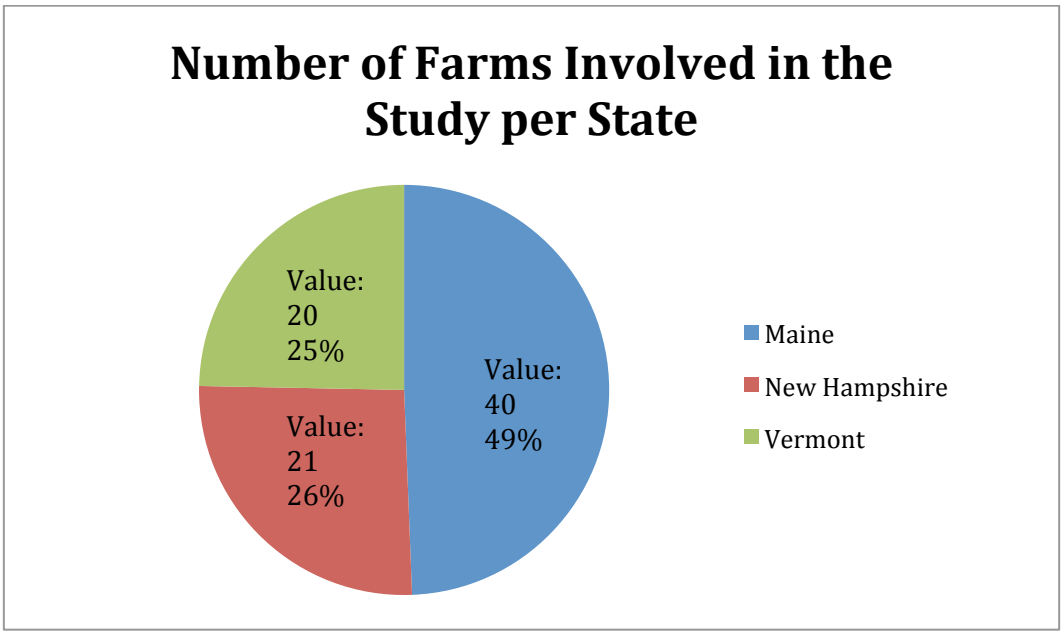

FIGURE 1. A comparison of the number of surveyed farms in Maine, New Hampshire, and Vermont for the summer of 2015.

The total combined fecal egg counts differed for the months of June and July (*P*=0.028) (Table I). Although both months had an almost identical sample size (the exception was one farm which did not submit a fecal sample in June), their means were different, with averages of 535 fecal eggs for June (+/- SD of 915) and 970 fecal eggs for July (+/- SD of 1504) (Table I). Based on the dependence of larval development on an average daily temperature of greater than 20 °C, the eggs which were deposited on the fields in the spring likely developed into larvae during mid-June and had not yet reached the infective L3 stage during the first month of sampling. In July, the fecal egg counts increased, which was probably the result of the newly formed adults beginning to produce their own eggs to re-infect the pastures.

The combined fecal egg count for August was numerically lower than in July, but was not statistically different (P=0.47) (Table I and Figure 2). If the new adults continued to produce eggs after infecting the sheep in July, the fecal egg count should have increased during August. The fecal egg count instead decreased in August, which may have been due to the deworming strategies of many producers. By August, the *H. contortus* parasites would have had approximately a month to affect the body systems of the host. Therefore, the animals may have started to show serious signs of anemia, bottle jaw, and other ailments, causing producers to deworm them. This could have caused the slight reduction in FEC that is present in the data, even if there was widespread resistance in the flock to the chosen dewormer. In the future, survey questions about the specific timing of the dewormer's administration may help with our understanding of the data.

Another possible reason for the fecal egg count decline during the month of August would be that the worms were beginning to enter hypobiosis due to the decreasing photoperiod. June 21, 2015 was the longest day of the year, and by mid-August the photoperiod was drastically shortened. This may have triggered the larvae to enter hypobiosis, the dormant stage of their life cycle inside the abomasum of the sheep and goats. While they are in hypobiosis, the larvae are not producing eggs, and therefore the fecal egg count levels may seem to have declined even though the larvae were still present inside the host (Machen et al.). In addition, the summer of 2015 in Maine was exceptionally dry, especially in comparison to preceding years (Birkel, 2015). Since *H. contortus* larvae tend to thrive in moist environments, the dry weather and lack of rainfall may have contributed to the death of the parasite in the late summer and early fall.

TABLE I. A comparison of the combined fecal egg counts for *Haemonchus contortus* from adult and juvenile small ruminants on the surveyed farms for the months of June, July, and August.

	Sample Size	Sum	Mean	Standard Deviation	*P*-value
Combined FEC for June	80	42,815	535	915	**0.028**
Combined FEC for July	81	78,600	970	1504	**0.47**
Combined FEC for August	81	66,250	818	1129	

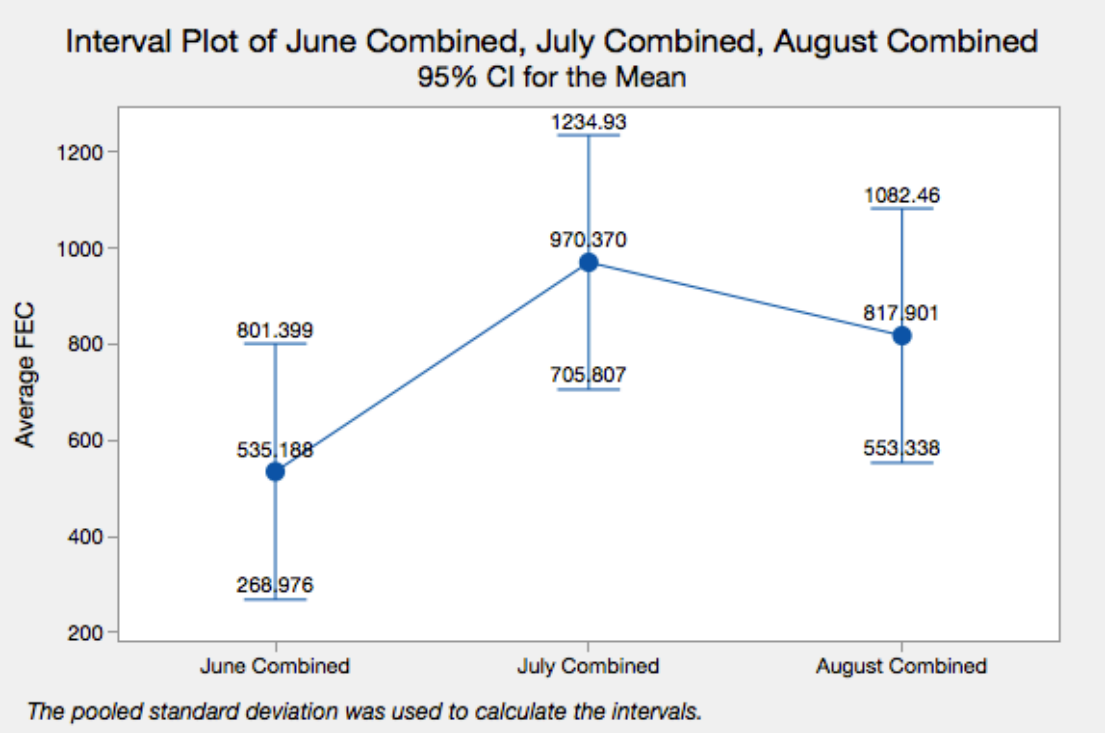

FIGURE 2. A graph of the combined fecal egg counts for *Haemonchus contortus* from adult and juvenile small ruminants on the surveyed farms for the months of June, July, and August.

The *P*-value for the combined FEC comparison from June to August demonstrated a tendency toward a difference between the groups (*P*=0.083) (Table II). Even though there was not a significant difference, the lower mean in June was likely due to the fact that in June, *H. contortus* was still in its juvenile stage and was not producing large amounts of eggs. In August, on the other hand, *H. contortus* was actively creating eggs (even when confronted with an anthelmintic and hypobiosis).

TABLE II. A comparison of the combined fecal egg counts for *Haemonchus contortus* from adult and juvenile small ruminants on the surveyed farms for the months of June and August.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Combined FEC for June	80	42,815	535	915	
Combined FEC for August	81	66,250	818	1129	**0.083**

The FEC of the adult and juvenile sheep and goats differed for June (*P*=0.042), July (*P*=0.035) and August (*P*=0.0042) (Table III). However, the FEC levels of the juveniles were actually lower than the adults in June (mean of 393 +/- SD 549 vs. mean of 173 +/- SD 758) (Table III). In June, the larvae had not yet developed to the infective L3 stage, and therefore the fecal egg counts in both the adults and juveniles were caused by the larvae from the previous year. The juvenile population was composed of both yearlings and lambs/kids. Since the yearlings were younger than the adults, they likely had lower numbers of carryover larvae from the summer and fewer larvae waking up from hypobiosis the following spring. In addition, the lambs in this group would have had no exposure *H. contortus* the previous year, further lowering the FEC.

In August and July, the mean FEC of the adults were lower than that of the juveniles (Table III). Although the large standard deviation makes it difficult to draw a definite conclusion, this is probably due to the increased susceptibility of the younger sheep and goats to the parasites. While the older animals may have built up some tolerance and immunity to *H. contortus,* the younger animal struggled to deal with the parasite loads. Producers who are concerned about the growth rate and physical condition of their lambs should monitor lamb and kid FEC closely to ensure that parasitism is not playing a large role in reducing the growth rate.

TABLE III. A comparison of the adult versus juvenile fecal egg counts for *Haemonchus contortus* from the months of June, July, and August.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
FEC for June: Adults	80	31,440	393	549	
FEC for June: Juveniles	66	11,425	173	758	**0.042**
FEC for July: Adults	72	31,100	432	741	
FEC for July: Juveniles	59	47,500	805	1243	**0.035**
FEC for August: Adults	68	23,750	349	454	
FEC for August: Juveniles	55	42,500	773	1087	**0.0042**

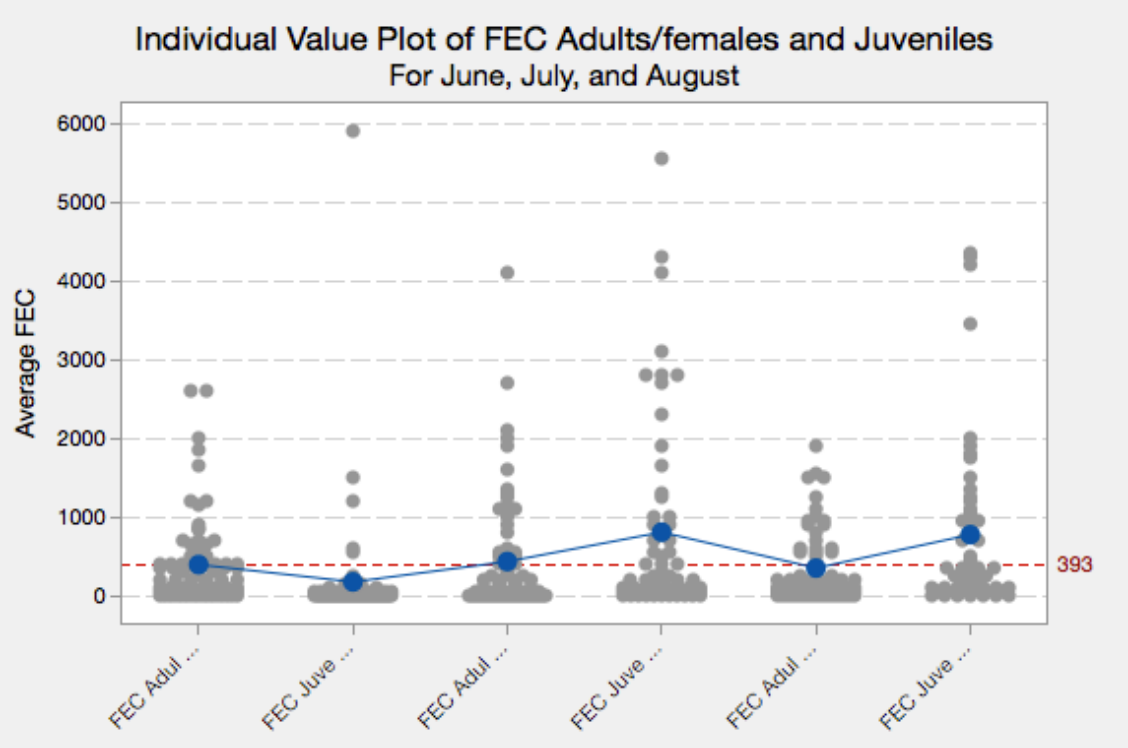

FIGURE 3. A comparison of the adult versus juvenile fecal egg counts for *Haemonchus contortus* from the months of June, July, and August.

The mean overall FEC were quite similar for the farms from three different states in New England, although New Hampshire had the highest counts and Maine had the lowest by small margins. However, the *P*-value of 0.97 suggests that there were no significant differences between the states in terms of FEC (Table IV). This supports the data collected by Wadsworth, 2015, which also found no significant difference in FEC for the same states. It is unfortunate that the number of samples in New Hampshire and Vermont was only half the number analyzed in Maine, which may have affected the statistical analysis. Still, the results suggest that producers throughout New England may be struggling with similar FEC levels or are using the same management strategies to deal with them.

TABLE IV. A comparison of the average fecal egg counts of *Haemonchus contortus* for the surveyed farms in Maine, New Hampshire, and Vermont for the summer of 2015.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Maine Overall FEC	40	31,271.7	671	942	**0.97**
New Hampshire Overall FEC	21	17,633.3	962	633	
Vermont Overall FEC	20	16,300.0	815	933	

The average fecal egg counts for farms inhabited by both sheep and goats were about 40% higher than those of the farms with one species (Table V and Figure 4). The different sample sizes may have been one of the major causes of the *P*-value of 0.40. Normally, mixed species grazing actually helps to reduce the parasite load in animals due to the fact that the life cycle of the parasite is interrupted by a wide variety of hosts. However, due to the fact that sheep and goats are both small ruminants, they can co-infect each other. In this case, the higher levels of *H. contortus* FEC on farms with both sheep and goats were due to the fact that the sheep and goats were mixing *H. contortus* parasites that may have had varying levels of resistance to particular dewormers or other treatments. Goats and sheep often have very different responses to infection as well. In general, goats tend to have less effective immune responses to nematodes than sheep. Therefore, it is often difficult for the producer to determine how or when to treat the sick animals.

If a producer was looking for another species to disrupt the *H. contortus* life cycle while grazing on pastures, a better choice would be cattle or horses due to the fact that there are fewer parasite similarities between these species and small ruminants. A producer may also have success by allowing sheep to infect a pasture that contains both tall and short forages, and then moving the sheep and allowing the goats to graze. If given a choice, goats will often choose to graze on taller forages, and therefore they would probably consume fewer parasites and allow more of the larvae to remain in the shorter grass and eventually die (Coffey, 2001).

TABLE V. A comparison of the average fecal egg counts of *Haemonchus contortus* for the surveyed farms with only sheep, only goats, and sheep and goats for the summer of 2015.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Sheep Only	53	39,888.3	753	778	
Goats Only	16	13,925.0	870	841	**0.40**
Sheep + Goats	12	13,450.0	1,121	1162	

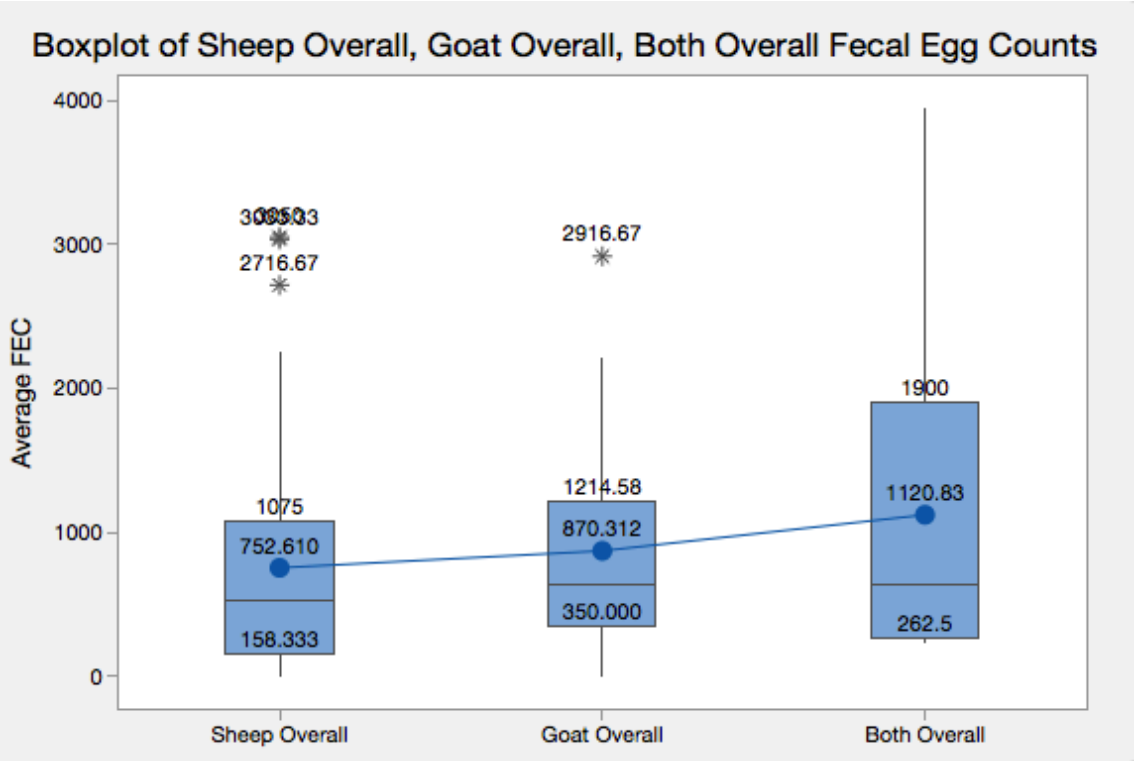

FIGURE 4. A comparison of the average fecal egg counts of *Haemonchus contortus* for the surveyed farms with only sheep, only goats, and sheep and goats for the summer of 2015.

Pasture grazing

There was no difference in fecal egg counts for the month of June between farms that had pasture with less than or equal to 10 acres and farms with over 10 acres of pasture (Table VI). The tests for July and August were also quite similar (data not shown). Overall pasture size had no significant effect on the FEC values.

TABLE VI. A comparison of the combined fecal egg counts of *Haemonchus contortus* for the month of June with dependence upon the acres of available pasture. The first group was for farms with less than or equal to 10 acres of pasture available for grazing, and the second group was for farms with greater than 10 acres of pasture available for grazing.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Less Than or Equal to 10 Acres	52	25,975	500	717	**0.70**
Greater Than 10 Acres	29	16,890	582	1196	

Several more trials were conducted on this topic. The first trial focused on the amount of pasture available to each animal. For example, an available pasture of 0.8 means that 0.8 acres of pasture were available per sheep or goat. The second trial focused on the stocking density of the animals on the pasture. For example, a stocking density of 2.0 would mean that there were 2.0 animals per acre of pasture. Although these trials are very similar, both analyses were conducted in order to break the data up into different groups that might indicate a significant difference.

The means were numerically different for the three groups regarding availability of pasture, but the difference between them was not statistically significant, most likely due to the high variability and the small sample sizes (P=0.45) (Table VII and Figure 5). Even when the first group (mean = 671 +/- SD 504) and the third group (mean = 962 +/- SD 829) were compared, the results were not significant (P=0.13), although the P-value was closer to 0.05 (Table VIII).

TABLE VII. A comparison of the average fecal egg counts of *Haemonchus contortus* per farm for the summer with dependence upon the available pasture per animal for grazing.

	Sample Size	Sum	Mean	Standard Deviation	*P*-value
Less Than 0.4 acres/animal	28	18,783.3	671	504	**0.45**
Between 0.4 and 0.79 acres/animal	31	27,321.7	881	1096	
Greater Than 0.8 acres/animal	22	21,158.3	962	829	

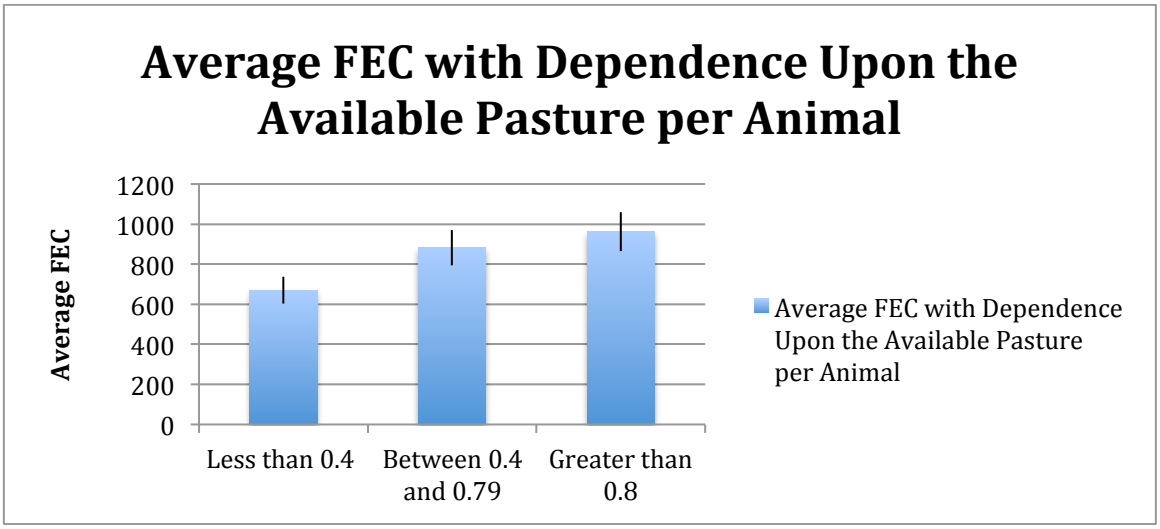

FIGURE 5. A comparison of the average fecal egg counts of *Haemonchus contortus* per farm for the summer with dependence upon the available pasture per animal for grazing. 10% error bars were also used for this figure.

Based on the assumption that more pasture per sheep should decrease the parasite load due to spreading the parasites out further and reducing their concentration on the pasture, the fecal egg counts should be higher if the sheep have less pasture. However, the highest fecal egg counts came from the farms that actually had the highest ratio of available pasture to animals (Table VIII). One possibility is that the farms with less

pasture space had to do a better job of maintaining the pastures and keeping up with other management techniques in order to keep their animals healthy. If producers were rotating their animals through pastures, this could also change the average FEC for each farm.

TABLE VIII. A more specific comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the available pasture per animal.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Less Than 0.4 acres/animal	28	18,783.3	671	504	**0.13**
Greater Than 0.8 acres/animal	22	21,158.3	962	829	

The stocking density of each farm was also calculated and analyzed. Farms with no pasture available for grazing were excluded from this test. Stocking density was defined as the number of animals per acre of pasture. Although the mean FEC was slightly higher for farms with a stocking density of greater than 2.0 animals/acre, the P-value of 0.83 could not be considered statistically significant (Table IX). However, when a scatterplot was created from the data and a linear regression line was calculated for the graph, a clear trend was shown (Figure 6). The linear regression line had an R-squared value of 27.33% and an equation of (Average FEC per farm) = (209.64 x Stocking Rate). The linear regression line indicated that as the stocking density of the pasture increased (more animals on the same amount of land), the fecal egg counts also increased. These results were most likely due to the concept of overcrowding the pastures and increasing

the fecal egg concentration on the pasture, which would then have lead to higher infection levels in the small ruminants.

TABLE IX. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the stocking density of the pastures.

	Sample Size	Sum	Mean	Standard Deviation	*P*-value
Stocking Density: < or = 2.0 Animals/Acre	30	31,341.7	825	851	**0.83**
Stocking Density: > 2.0 Animals/Acre	40	34,688.3	867	890	

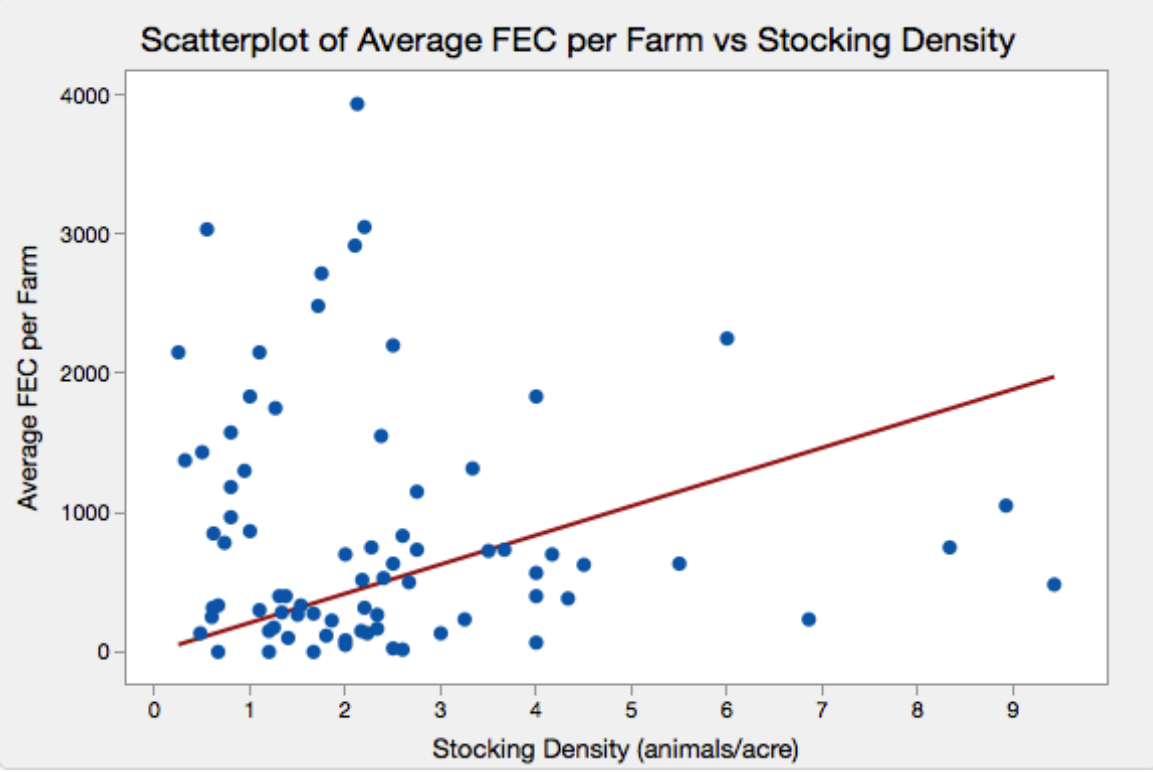

FIGURE 6: A comparison of the average fecal egg counts of *Haemonchus contortus* per farm for the summer with dependence upon the stocking density of the pastures.

There was no detectable difference in the combined fecal egg counts of the animals for the summer based on the turnout dates of before May 1st or on/after May 1st (*P*=0.75) (Table X). We had hypothesized that the animals which were first let out on pasture during and after May would have lower levels of *H. contortus* infection later in the summer than those that were allowed to graze on pasture during the late winter and early spring. This theory suggested that for the later turnout group, *H. contortus* would begin producing fecal eggs after the hypobiotic period in the spring, but the animals would not consume the eggs due to the lack of pasture and the parasites would perish. The lack of difference in these results was probably due to the fact that the *H. contortus* larvae were able to keep producing eggs through April or May, when the sheep were able to graze on the pasture. At this point, the larvae were able to develop into the L3 larvae on grass, infect the sheep, and still have plenty of time to begin laying new eggs by July.

TABLE X. A comparison of the average fecal egg counts of *Haemonchus contortus* for each farm for the summer with dependence upon when the small ruminants were first allowed to graze on pasture. The first group represents the farms that claimed to have turned their animals out on pasture for the first time before or during April 2015. The second group represents the farms that claimed to have turned their animals out on pasture for the first time after or during May 2015.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Before/During April	29	24,383.3	841	723	**0.75**
After/During May	43	39,130.0	910	985	

There was no significant difference in the fecal egg counts for adults based on turnout time for the months of June, July and August, with *P*-values between 0.2 and 0.7 (Table XI). The month of June showed the largest difference, with a *P*-value of 0.27 and higher FEC levels for animals that were turned out on pasture before May (Figure 7).

TABLE XI. A comparison of the average fecal egg counts of *Haemonchus contortus* for adult sheep and goats with dependence upon when the small ruminants were first allowed to graze on pasture during 2015. The first group was the farms that began turning their animals out before May 1st, and the second group was the farm that began turning their animals out during or after May 1st.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Adults, June, Before May 1st	28	13,800.0	493	610	**0.27**
Adults, June, Began On/After May 1st	49	16,990.0	347	520	
Adults, July, Before May 1st	27	10,550.0	391	558	**0.67**
Adults, July, Began On/After May 1st	42	19,800.0	471	863	
Adults, August, Before May 1st	24	7,400.0	308	397	**0.55**
Adults, August, Began On/After May 1st	41	15,550.0	379	491	

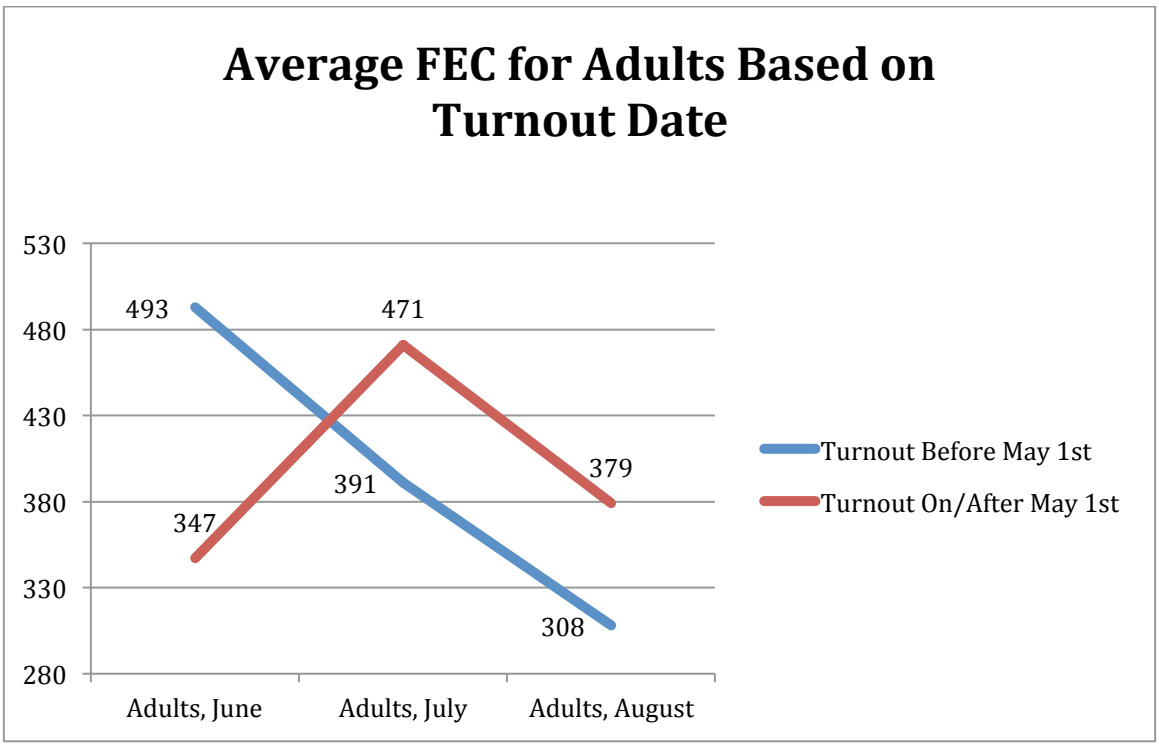

FIGURE 7. A comparison of the average fecal egg counts of *Haemonchus contortus* for adult sheep and goats with dependence upon when the small ruminants were first allowed to graze on pasture during 2015. The first group was the farms that began turning their animals out before May 1st, and the second group was the farm that began turning their animals out during or after May 1st.

There was no significant difference in the fecal egg counts for the juvenile populations for the months of June, July, and August based on turnout date, with *P*-values of between 0.4 and 0.99 (Table XII and Figure 8). The majority of the eggs were probably laid on pasture during the mud season, from April to May, and then hatched and developed over the course of the summer. The problem with this data was that the two groups did not have enough separation between them. The parasites were able to lay eggs in April or May on the pasture and have the larvae consumed by the sheep in time to produce large amounts of fecal eggs during July, and therefore there it made no statistical difference whether the animals were turned out before or during May. It is also possible that a cold snap might have killed some of the eggs that were laid on pasture in April,

which would further reduce the fecal egg counts for the early turnout group. A controlled experiment with one group of sheep released onto pastures in April/May and one group of sheep turned out in June may lead to a greater level of significance. Unfortunately, only 6 of the 81 of the farms turned their sheep out on pasture in June or after, which is not a large enough sample size to form any definite conclusions or run any significant tests.

TABLE XII. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer for juvenile sheep and goats with dependence upon when the small ruminants were first allowed to graze on pasture during 2015. The first group was the farms that began turning their animals out before May 1st, and the second group was the farm that began turning their animals out during or after May 1st.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Juveniles, June, Before May 1st	24	2,075.0	87	162	**0.46**
Juveniles, June, On/After May 1st	40	9,350.0	234	965	
Juveniles, July, Before May 1st	21	16,900.0	805	1181	**0.99**
Juveniles, July, On/After May 1st	36	28,800.0	800	1324	
Juveniles, August, Before May 1st	20	14,500.0	725	1173	**0.73**
Juveniles, August, On/After May 1st	33	27,500.0	833	1074	

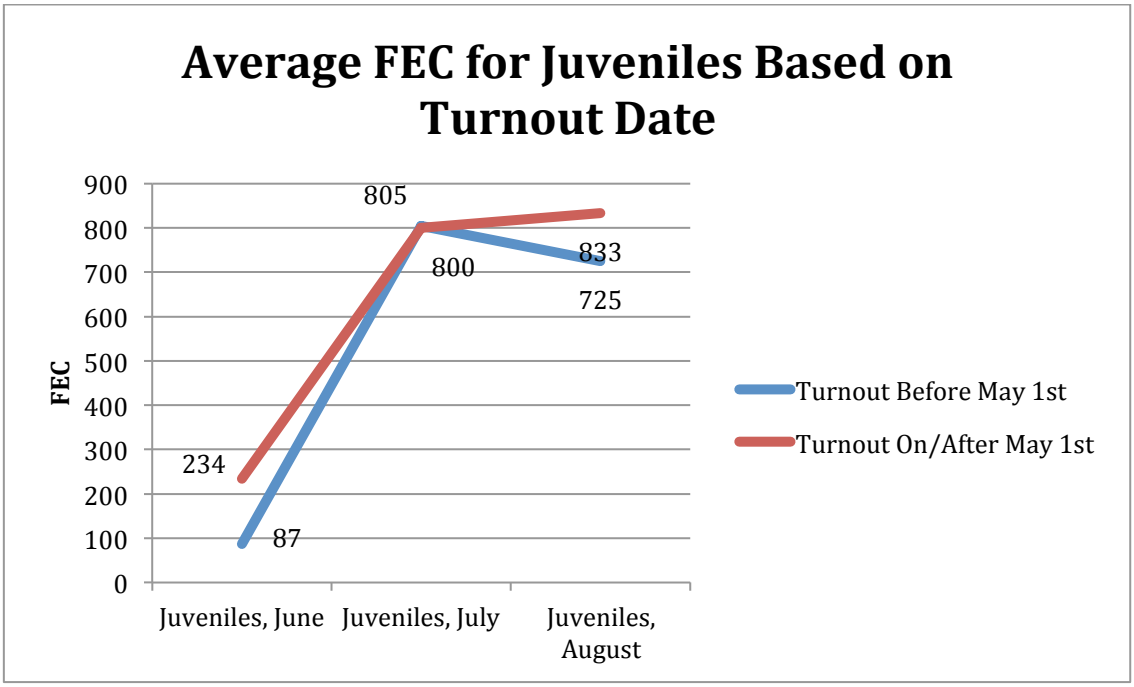

FIGURE 8: A comparison of the average fecal egg counts of *Haemonchus contortus* for juvenile sheep and goats with dependence upon when the small ruminants were first allowed to graze on pasture during 2015. The first group was the farms that began turning their animals out before May 1st, and the second group was the farm that began turning their animals out during or after May 1st.

Vet care and management practices

At first, farms with less frequent veterinary care seemed to have lower fecal egg counts. The farms that claimed that they "Never" used a veterinarian had very low FEC, with an average of around 261 FEC per farm (Table XIII). The other three categories all exhibited FEC in the same range, which was about three times the FEC of the farms that do not use the vet (Figure 9). However, when the results are viewed more closely, it becomes clear that there are some other factors to consider. The first factor is that the pool of "Never" respondents consisted of only 7 farms. This is such a small subsection of the 81 farms that the results are unlikely to be very accurate. In addition, the fact that the farms with high FEC used a veterinarian can be interpreted to mean that the producers

were focused on solving the problem and were enlisting the help of a veterinarian to do so.

TABLE XIII. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the frequency of vet care on the surveyed farms. The four groups represent vet care on an as-needed basis, vet care multiple times per year, no vet care, or vet care once per year.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Vet Care: As Needed	38	35,433.3	933	943	**0.30**
Multiple Times Per Year	17	14,975.0	881	757	
Never	7	1,825.0	261	246	
Once Per Year	19	15,030.0	791	861	

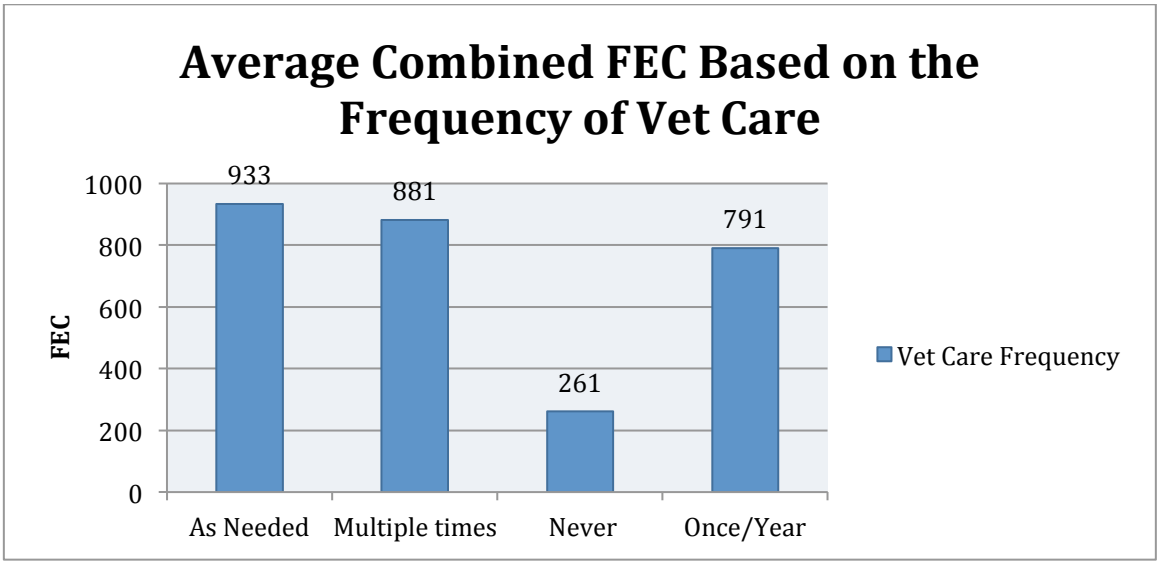

FIGURE 9. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the frequency of vet care on the surveyed farms. The four groups represent vet care on an as-needed basis, vet care multiple times per year, no vet care, or vet care once per year.

When results from the "As Needed" veterinary care group were compared with the "Never" group, although the *P*-value exceeded the required threshold of 0.05, the *P*-value of 0.07 implied that there was only a 7% chance that the difference in fecal egg counts was purely based on random error (Table XIV). Further studies should be conducted on this topic to determine why this correlation may exist.

TABLE XIV. A more specific comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the frequency of vet care.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Vet Care: As Needed	38	35,433.3	933	943	**0.07**
Vet Care: Never	7	1,825.0	261	246	

Whether or not a particular producer used FAMACHA scores in his or her deliberation over keeping or culling an animal seemed to have very little effect on the proliferation of *H. contortus* at the farms (*P*=0.75) (Table XV). It is likely that other factors provided more significant information than the use of FAMACHA scores for replacement.

TABLE XV. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon whether or not FAMACHA scores are used to determine which sheep or goats to replace.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
FAMACHA: Yes	24	21,641.7	902	994	**0.75**
FAMACHA: No	40	33,180.0	830	790	

There was no significant difference between well-established farms and newer farms for overall combined FEC for June (P=0.92) (Table XVI). The term "established" was left up to the producer to define, but the point of the question was to determine how long small ruminants had inhabited these farms and how this may affect the FEC. Further clarification on this question might have led to more accurate results. It was hypothesized that newer farms may have had less experience with treating *H. contortus* and therefore have higher infection levels. However, no significant difference between them could be found (mean = 526 +/- SD 1058 and mean = 547 +/-SD 750) (Table XVI).

TABLE XVI. A comparison of the average fecal egg counts of *Haemonchus contortus* for adults and juveniles for June depending on the number of years that the farm has been established. The first group was for farms that have been "established" for 10 or less years. The second group was for farms that have been "established" for more than 10 years.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
10 or Less - June	41	21,550	526	1058	**0.92**
More than 10 - June	39	21,315	547	750	

For the average FEC for July based on years of establishment, level of significance was less than the 0.05 threshold (P=0.20), but it was close enough to suggest that there may have been a difference, although future studies would be necessary to confirm it (Table XVII). The difference could be attributed to the ability of the larvae to develop into adults and produce their own eggs in the late spring. The fecal egg counts tend to be highest in July due to the fact that all of the new larvae have settled into the

small ruminant abomasums and are producing high levels of eggs. Newer producers may be less prepared for the sudden outbreak of fecal eggs or be less adept at recognizing the signs of high levels of infection, and this may contribute to higher FEC on their farms. It is also possible that the more established farms have had more time to work out a grazing management program and may be more skilled at pasture rotations, which would decrease the impact of the sudden surge in egg production. On the other hand, the scatterplot shows a trend towards more years of establishment causing higher FEC levels (Figure 10). The R-squared value for the linear regression line is 26.55% and the equation is (Average FEC/farm) = (33.5521 x Years Established). More research is required on this topic to determine why these results occurred.

TABLE XVII. A comparison of the average fecal egg counts of *Haemonchus contortus* for adults and juveniles for July depending on the number of years that the farm has been established. The first group was for farms that have been "established" for 10 or less years. The second group was for farms that have been "established for more than 10 years.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
10 or Less - July	37	48,800	1,319	1,876	**0.20**
More than 10 - July	35	29,800	851	1,096	

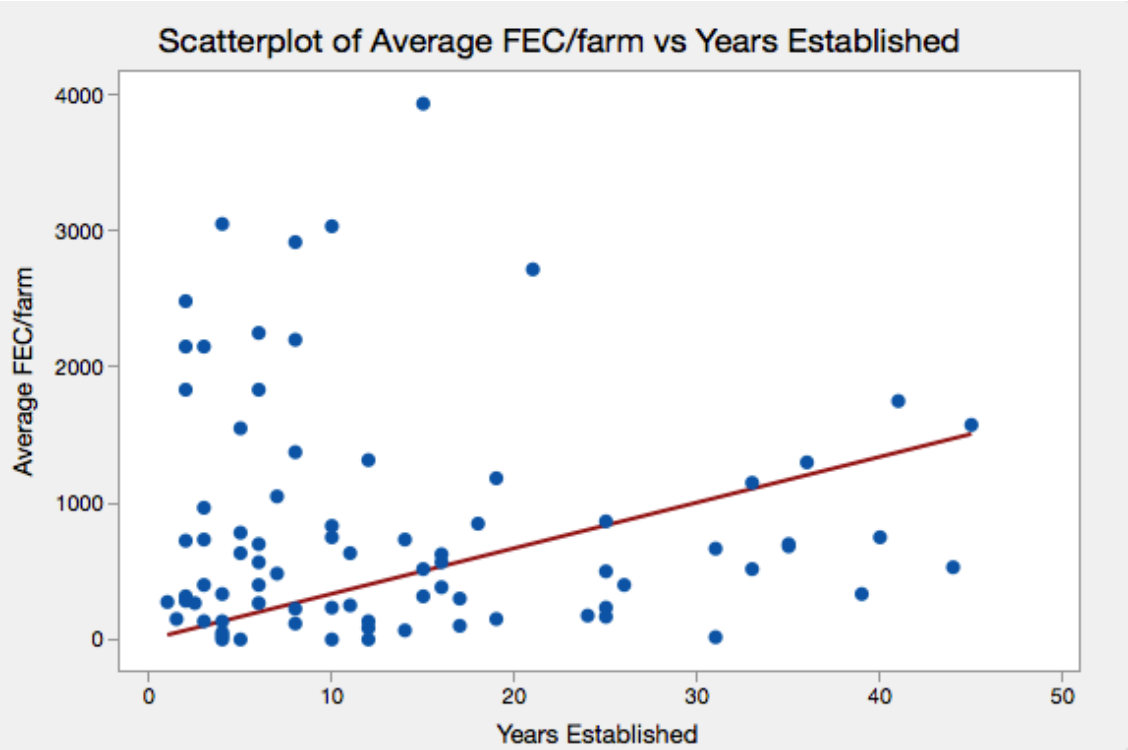

FIGURE 10. A comparison of the average fecal egg counts of *Haemonchus contortus* for adults and juveniles for July depending on the number of years that the farm has been established. The first group was for farms that have been "established" for 10 or less years. The second group was for farms that have been "established" for more than 10 years.

The farms were split into groups with a cutoff at 15 adult sheep. Larger farms with 16 or more sheep actually had lower FEC numbers (mean = 592 +/- SD 555 compared to mean = 964 +/- SD 989) and a trend was indicated (Table XVIII and Figure 11). A possible hypothesis for these results is that the farms with more sheep may have had a better organizational plan for monitoring infection levels in their animals. The larger farms were also more likely to be commercial businesses. They might have been able to invest more time and energy into eradicating parasites in their flock because their livelihood depended upon it. On the smaller farms, although the producers may know the sheep better on an individual basis, there may not have been an established protocol for

treating *H. contortus* infections or culling chronically infected animals.

TABLE XVIII. A comparison of the average combined fecal egg counts of *Haemonchus contortus* for adults and juveniles for the summer depending on the total number of adult sheep on the farm.

	Sample Size	Sum	Mean	Standard Deviation	*P*-value
Farms with 5-15 sheep	40	38,541.7	964	989	**0.091**
Farms with 16+ Sheep	25	14,796.7	592	555	

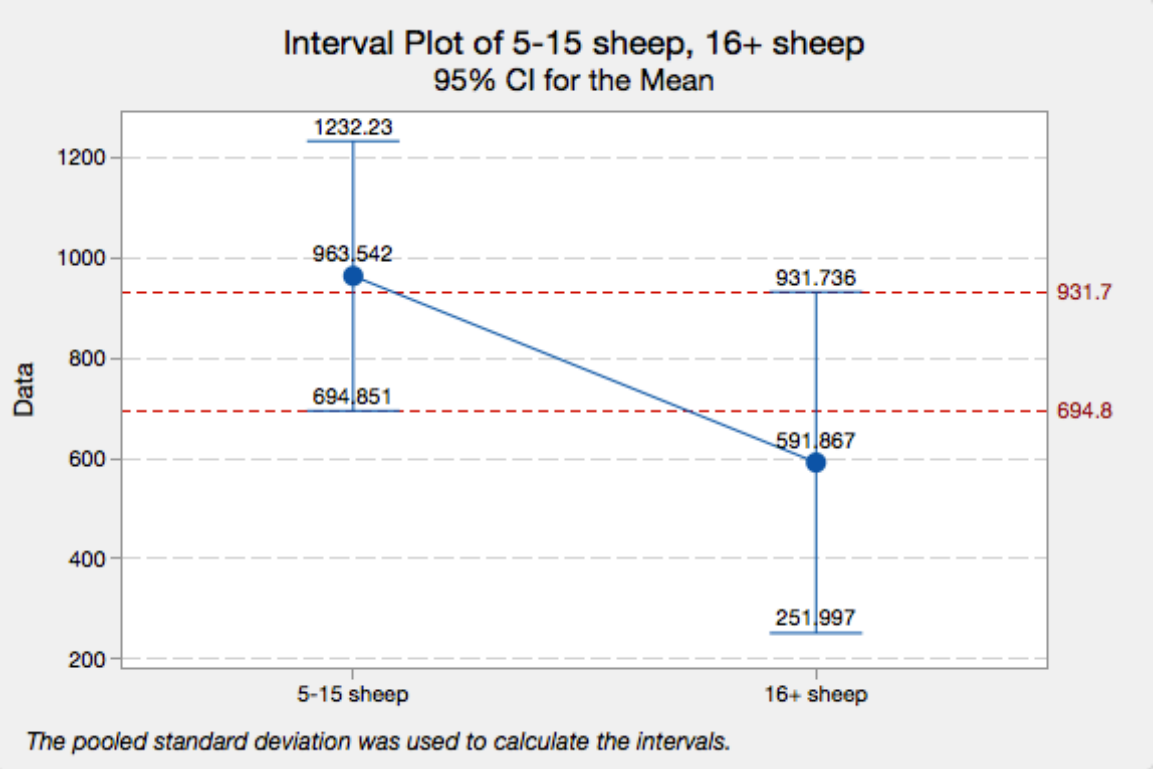

FIGURE 11. A comparison of the average combined fecal egg counts of *Haemonchus contortus* for adults and juveniles for the summer depending on the total number of adult sheep on the farm.

When producers were asked about the various ailments that they had noticed in their flocks, such as anemia, death, coccidiosis, coughing, etc., the farms with no

observable symptoms had the lowest FEC, which makes sense because lower infection levels will led to less signs of illness (*P*=0.62) (Table XIX and Figure 12).

TABLE XIX. A comparison of the average combined fecal egg counts of *Haemonchus contortus* for adults and juveniles for the summer depending on the number of parasite "symptoms" exhibited on the farms. For example, farms with "No Symptoms" had zero reported symptoms of a parasite infection in their flocks. Farms with "One Symptom" had only one reported symptom of a parasite infection on their farms.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
No Symptoms	40	29,441.7	736	832	**0.62**
One Symptom	18	17,083.3	949	882	
Two or More Syptoms	24	21,438.3	893	875	

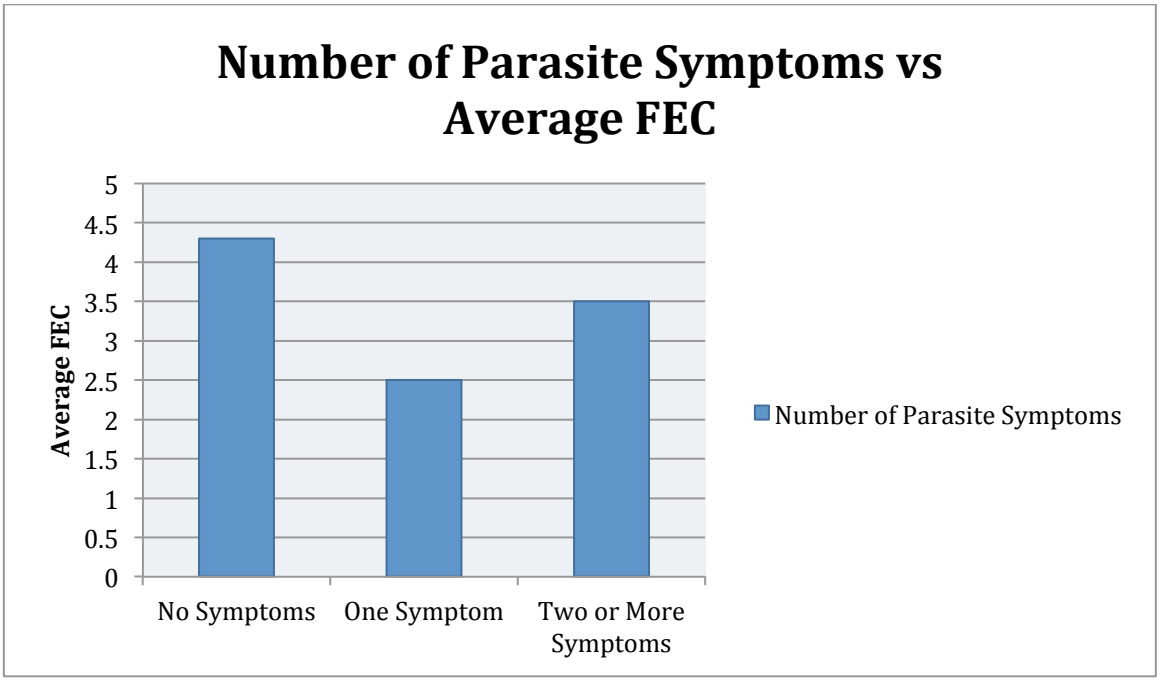

FIGURE 12. A comparison of the average combined fecal egg counts of *Haemonchus contortus* for adults and juveniles for the summer depending on the number of parasite "symptoms" exhibited on the farms. For example, farms with "No Symptoms" had zero reported symptoms of a parasite infection in their flocks. Farms with "One Symptom" had only one reported symptom of a parasite infection on their farms.

When “one symptom” was compared to “no symptoms” for the fecal egg count analysis, *P*=0.84, so the null hypothesis could not be rejected and there was no significant statistical difference between the two groups (Table XX). The data suggested that the number of signs of illness did not matter, and any observable symptoms in the sheep or goats indicated high levels of *H. contortus*.

TABLE XX. A more specific comparison of the average combined fecal egg counts of *Haemonchus contortus* for adults and juveniles for the summer with dependence upon the number of parasite “symptoms” exhibited on the farms. For example, farms with “No Symptoms” had zero reported symptoms of a parasite infection in their flocks. Farms with “One Symptom” had only one reported symptom of a parasite infection on their farms.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
One Symptom	18	17,083.3	949	882	**0.84**
Two or More Symptoms	24	21,438.3	893	875	

Dewormers

As stated in the introduction, anthelmintic resistance is a major concern in small ruminant populations in the Northeast. When the farms were split based on their deworming practices, there was a difference in FEC, although it was not statistically significant (Table XXI and Figure 13). The farms that did not use a dewormer had the lowest counts by a large margin, with an average FEC of 165 (+/- SD 136) (Table XXI). However, this made sense because farms with low levels of infection probably did not have animals that were exhibiting clinical signs and therefore did not need a dewormer.

The farms that did use a chemical dewormer had relatively high levels of infection with FEC of approximately 828 per farm (+/- SD 791). The farms that used organic and natural deworming methods had the highest infection levels, with over 1,264 FEC per farm (+/- SD 1350) (Table XXI).

There were two possible hypotheses for these results. The first is that higher FEC levels and an increased level of resistance to chemical dewormers forced producers with very high levels of infection to try alternative and organic methods to manage the *H. contortus* parasites. Whether the results of these treatment methods would appear in the data set would depend on when the remedies were administered. Another possible hypothesis to explain the data is that the organic and natural dewormers were simply less effective than the chemical dewormers, even with rising levels of anthelmintic resistance. If the treatments were less potent than the dewormers, this should have led to higher combined FEC for summer.

TABLE XXI. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the general type of dewormer used.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
No Dewormer	5	825	165	136	**0.078**
Chemical Dewormer	68	56,321.7	828	791	
Organic/Natural	8	10,116.7	1,265	1350	

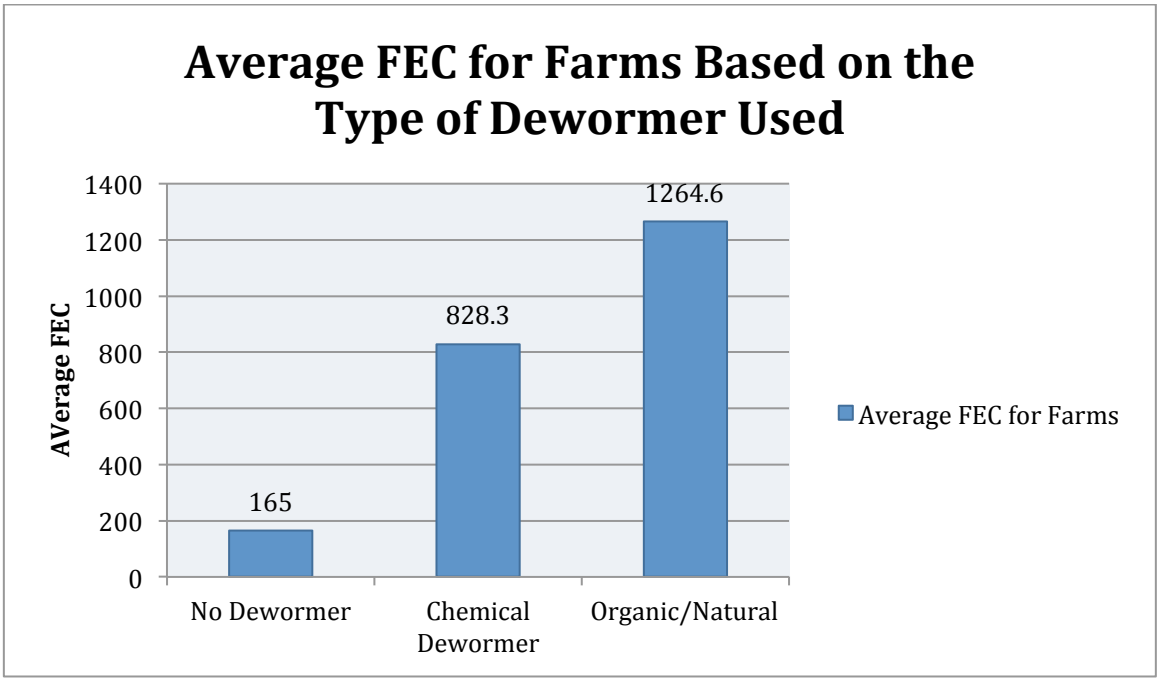

FIGURE 13. A comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the general type of dewormer used.

There was an observable graphical and statistical difference between the farms that used a combination of commercial and natural dewormers and those that used only commercial dewormers (P=0.014) (Table XXII and Figure 14). The average FEC for the first group were almost twice those of the second group (mean = 1,102 +/- SD 996 and mean = 634 +/- SD 525) (Table XXII). This implies that the combination of chemical and natural dewormers was not working successfully in the small ruminant populations, probably due to a variety of factors. These could include chemical interactions between the natural products and the ingredients in the commercial anthelmintics, or simply an inconsistency in treatment plans due to switching between different drugs. A more complete survey regarding the specific timing and pairing of the commercial and natural dewormers would help with the analysis.

TABLE XXII. A more specific comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the type of dewormer used. The "Commercial/Natural Dewormers" option implies a combination of commercial and natural dewormers used together or in sequence throughout the summer.

	Sample Size	Sum	Mean	Standard Deviation	*P*-value
Commercial/Natural Dewormers	25	27,550.0	1,102	996	**0.014**
Commercial Dewormers Only	42	26,608.3	634	525	

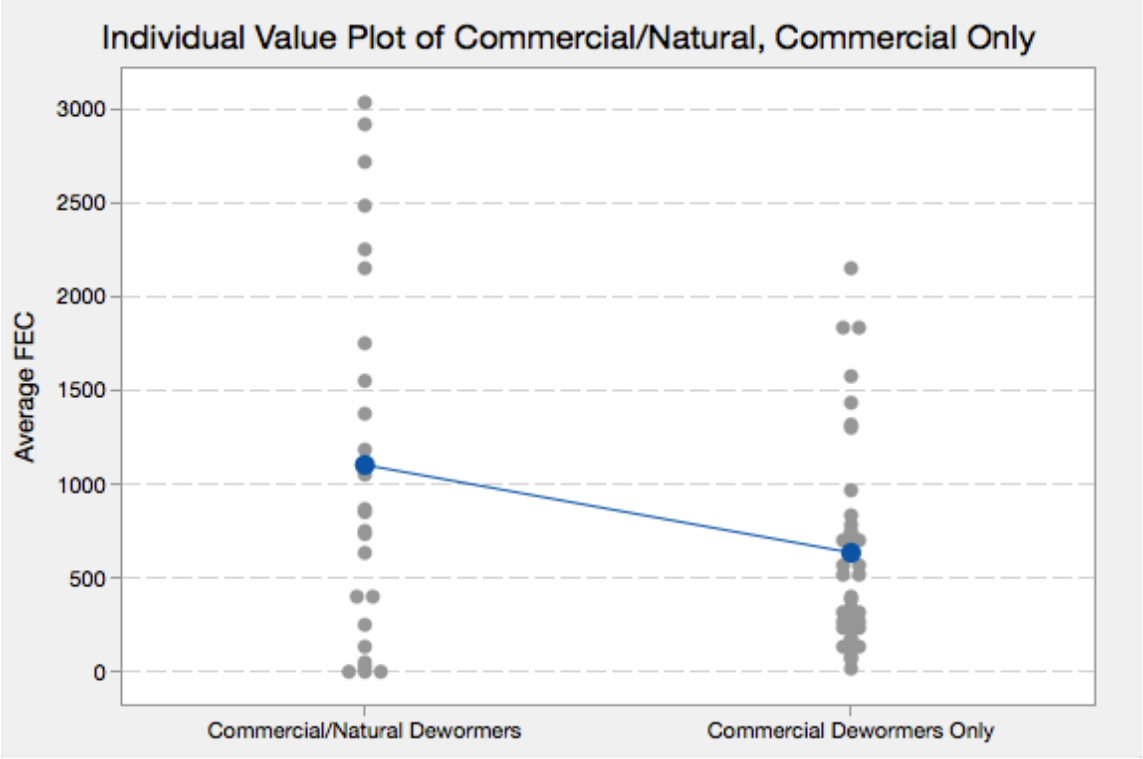

FIGURE 14. A more specific comparison of the average fecal egg counts of *Haemonchus contortus* for the summer with dependence upon the type of dewormer used. The "Commercial/Natural Dewormers" option implies a combination of commercial and natural dewormers used together or in sequence throughout the summer.

Farms were separated into two groups based on whether or not they reported Ivermectin use. Ivermectin (sometimes sold as Stromectol) is a chemical anthelmintic that was the most commonly used dewormer in the group of 81 farms that were sampled

in this study. However, due to the widespread use of Ivermectin in New England farms, there have been concerns about the level of resistance in *H. contortus* to this drug. The results did not support a correlation due to the *P*-value of 0.54 (Table XXIII). It was interesting to note that the farms that used Ivermectin had higher average FEC for the summer than farms that did not use Ivermectin. However, it was difficult to determine whether the widespread Ivermectin resistance caused the drug to be ineffective or whether the farms that already had lower FEC chose not to use Ivermectin because their problem was not severe. If the drug was ineffective, this could have led to higher FEC in the flocks by falsely tricking the producer into thinking that the problem had been addressed.

Similar results were also found when trials were conducted with Fenbendazole (Safeguard), another popular dewormer. This time, P=0.26, which suggested there may have been a possible trend between the use of Safeguard and higher FEC. Again, determining the timing of the dewormer administration would have led to more complete results.

In addition, very few of the farms used only one type of dewormer for the season. When the groups were made for these trials, any farm that utilized Ivermectin at some point during the summer was counted as a farm "using Ivermectin", and farms that did not report any Ivermectin use were categorized as "not using Ivermectin". Therefore, the interactions between the various dewormers were not taken into account because each dewormer was compared to FEC on an individual basis, even if it was used in combination with other products.

TABLE XXIII. A more specific comparison of the average combined fecal egg counts of *Haemonchus contortus* for the summer with dependence upon whether the farms used Ivermectin and Fenbendazole (Safeguard) dewormers.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Using Ivermectin	42	37,255.0	887	816	**0.54**
Not Using Ivermectin	39	30,008.3	769	902	
Using Fenbendazole	33	31,683.3	960	954	**0.26**
Not Using Fenbendazole	48	35,580.0	741	778	

There was no significant difference in the overall combined FEC for farms that used Cydectin over the summer and those that did not (P=0.71) (Table XXIV). Cydectin is a dewormer that was less popular in this study than Ivermectin and Safeguard. However, those farms that used Cydectin actually had lower FEC (mean = 774 +/- SD 770) than those farms that did not use Cydectin (mean = 853 +/- SD 892). The sample size of farms using Cydectin products was small (n=23) and the survey did not indicate how often the Cydectin was administered, so was difficult to draw any major conclusions from this data. However, the fact that fewer farms used Cydectin, which may have been more effective than the more popular brands such as Ivermectin and Safeguard, highlighted an area of potential future research. Future studies should be conducted to determine whether *H. contortus* parasites exhibit less resistance to Cydectin dewormers due to its limited use.

TABLE XXIV. A more specific comparison of the average combined fecal egg counts of *Haemonchus contortus* for the summer with dependence upon whether the farms used Cydectin dewormers.

	Sample Size	**Sum**	**Mean**	**Standard Deviation**	***P*-value**
Using Cydectin	23	17,813.3	774	770	**0.71**
Not Using Cydectin	58	49,450.0	853	892	

Conclusion

Summary of Results

Analyzing the fecal egg counts of *Haemonchus contortus* on farms can indicate the most successful ways to manage sheep and goat flocks in New England. The average fecal egg count per farm varied between the months of June, July, and August, with the highest contamination levels in July (mean = 970 +/- SD 1504). This is most likely due to the maturation of the L3 larvae into infective, egg-producing *H. contortus* adults. Egg counts then dropped for the month of August due to the transition of infective adults into a period of hypobiosis, as well as due to the dry summer. In addition, the results showed that juveniles produced much higher levels of *H. contortus* eggs than the adults for the months of July and August (Table III), but the adults had higher FEC during June (adult mean of 393 +/- SD 549 and juvenile mean of 173 +/- SD 758). The juveniles likely had lower infection levels in June due to possessing fewer infective larvae from the year before, but as the summer continued, their lack of exposure to the parasite made them

more susceptible.

In addition, the study found that there was no significant difference between the FEC infection levels in Maine, New Hampshire, and Vermont for the surveyed farms (*P*=0.97). This suggests that the producers may have been struggling with comparable infection levels or using similar management strategies. The farms were also compared on a basis of goats only, sheep only, and goats and sheep combined. Although the farms with sheep and goats had significantly higher average FEC than the single species farms (most likely due to parasite mixing and the different susceptibilities of the animals to infection) the vast difference in sample size probably contributed to the *P*-value of 0.40 (Table V).

Pasture management was another important topic for this study. There was no correlation between overall pasture available for grazing and FEC (*P*=0.70) (Table VI). For farms with less than 0.4 acres per animal available, the FEC were lower than the farms with greater than 0.8 acres per animal available (mean = 671 +/- SD 504 vs mean = 962 +/- SD 829), although the *P*-value was not low enough to meet the minimum threshold for significance (*P*=0.13) (Table VIII). The stocking density trial also showed a strong upward trend towards higher FEC for farms with higher stocking densities (Figure 6), but a split between farms with a stocking density of less than or equal to 2.0 animals/acre and greater than 2.0 animals/acre was not significant (*P*=0.83) (Table IX).

Another important consideration was the date when the flocks were first allowed out onto pastures in 2015. No significant difference was observed in the average FEC of flocks turned out before or during April and flocks turned out after or during May

(*P*=0.75) (Table X). A comparison of the adult population FEC for the months of June, July, and August with dependence upon the turnout date also did not indicate a significant correlation (Table XI). The same was true for the juvenile populations (Table XII). The reason for the lack of correlation between turnout dates and FEC was likely due to the fact that the split between the two groups occurred too early in the summer. Regardless of whether the sheep were turned out before or during/after May, the parasites were still able to infect the pastures with eggs, which were then able to develop into larvae and were ingested by the sheep and goats in time for the re-infection period in July.

Although the differences between farms that utilized a veterinarian "as needed", "multiple times per year", "never", and "once per year" were not statistically significant (*P*=0.29), the sample sizes were too unequal to draw any definite conclusions from the data (Table XIII). However, the farms that "Never" used a vet had the lowest average FEC (mean = 261 +/- SD 246), which might have been due to the fact that farms with lower infection levels did not need the services of a vet during the summer of 2015 (Table XIII). Producers were also asked whether they would consider FAMACHA scores when deciding whether to keep or cull an animal, and the average FEC for the "Yes" and "No" groups were not different enough to indicate a trend (*P*=0.75) (Table XV). Other factors should be considered when selecting animals for replacement if *H. contortus* is a major concern.

In addition, the data did not indicate a significant difference between the average FEC of farms that had been established for less than ten years versus ten or more years (*P*=0.92) (Table XVI). A better definition of the term "established" might have clarified

the survey question and led to different results. It was also possible that the age of a farm may simply have had no effect on the average FEC. The scatterplot seemed to suggest that more established farms tended to have higher FECs (Figure 10). To further complicate matters, the month of July had a *P*-value of 0.20, which was low enough to suggest that younger farms had a higher average combined FEC (mean = 1,319 +/- SD 1,876) than the older farms (mean = 851 +/- SD 1,096) (Table XVII). Newer producers may have been less skilled at observing the signs of infection during the *H. contortus* outbreak in July, and more established farms may have had better pasture management and rotation strategies in place as well.

Farms with adult sheep flocks of at least 16 sheep tended to have lower FEC than those with 15 or fewer sheep (*P*=0.091) (Table XVIII). Farms with greater numbers of sheep may have had a better system for treating *H. contortus* or they may have had more resources to invest in eradicating the parasite. When producers reported the number of parasite "symptoms" on their farms, no significant difference was found between farms with no symptoms, one symptom, and two or more symptoms (*P*=0.62), although the bar graph indicated that farms with one symptom had the lowest average FEC (Table XIX and Figure 12). Further analysis also showed no significant difference between the FEC of farms with one and two symptoms, and suggested that the number of observable symptoms in the animals did not have a correlation with the FEC (Table XX).

The farms that did not use a dewormer had much lower average combined FEC (mean = 165 +/- SD 136) than the farms that used chemical dewormers (mean = 828 +/- SD 791) or the farms that used organic or natural dewormers (mean = 1,265 +/- SD 1350) (*P*=0.078) (Table XXI). The farms with the lowest infection levels probably did not use a

dewormer because their animals were not showing major symptoms. The farms that used chemical dewormers might have had higher FEC levels than the organic/natural farms because the dewormers were still effective despite the high levels of antibiotic resistance, or because the resistance to chemical dewormers in past years caused producers with high levels of infection to turn to alternative methods.

A comparison between farms that used a combination of commercial and natural dewormers versus farms that only used commercial dewormers indicated a significant difference between the two groups (P=0.014) (Table XXII). Farms that used natural dewormers in combination with the commercial ones had much higher infection levels, probably due to chemical interactions between the combination of dewormers or inconsistency from switching between the two different categories. More information about how the dewormers were administered would have been helpful.

Finally, three different types of commercial dewormers were compared: Ivermectin (Stromectol), Fenbendazole (Safeguard), and Cydectin. For Ivermectin, there were no significant differences between farms that used these dewormers and those that did not (P=0.54) (Table XXIII). Fenbendazole had a low overall P-value and a lower mean for farms that did not use it (P=0.26) (Table XXIII), but the difference was not statistically significant. The problem with these dewormers could have been increased resistance to the anthelmintic or misplaced trust by producers in a drug that was not actually effective. However, interactions between various dewormers clouded the attempt to analyze the drugs on an individual basis. Cydectin led to lower FEC levels when it was used on a farm, but the P-value was 0.71 and the sample size was small (Table XXIV).

Overall, the most effective management techniques and characteristics of sheep and goat farms seem to involve the use of chemical versus natural dewormers, the size of the sheep flocks, and potentially the turnout date. Other factors such as the use of FAMACHA scores for replacement, and the location of the farms in Maine, New Hampshire, or Vermont, and the number of symptoms observed in the flock were not relevant to the *H. contortus* fecal egg count levels on the farms. It was difficult to determine whether resistance to chemical dewormers was prevalent in these flocks due to overlapping dewormers and the lack of information about when they were administered.

Comparison of Results

A summary of the conclusions gathered from this experiment would not be complete without a brief comparison to the data from 2014 that was collected and analyzed by Ariana Wadsworth (Wadsworth, 2015). Although the producers in the 2014 project were asked different questions and their samples were submitted at different times, the projects were similar enough for a comparison to occur.

First of all, it is important to note that due to a lack of samples, Wadsworth was unable collect enough data about the "unbred" populations (or "juvenile" populations, as they were called in this study) to make any definite statements about the effect of management strategies on *H. contortus* levels in the young animals. Therefore, the 2015 report and discussion were based on the samples from adult populations, while this 2016 analysis was mostly based on the combined pool of adult and juvenile samples. Wadsworth's FEC values will generally be lower than this study's values because the combined value in this study is the sum of the average adult and juvenile FEC for each

farm. Therefore, the values should not be directly compared.

Both Wadsworth's project and Pouliot's project compared the infection levels of Maine, New Hampshire, and Vermont farms. In Wadsworth's study, New Hampshire had the lowest *H. contortus* infection levels, with an average fecal egg count in eggs per gram of 208 ($P<0.30$) (Wadsworth, 2015). In the Pouliot study, New Hampshire actually had the highest FEC with a mean of 962 ($P=0.97$) (Table IV). This indicates that either New Hampshire had a drastic *H. contortus* problem that appeared in less than a year or that the participating farms for both years were quite different. However, in both studies the *P*-values were too high to assume a significant difference.

Wadsworth's project also compared the effect of species type on the FEC for the farms. Farms with only sheep were found to have the lowest average FEC (300.5 EPG), farms with only goats had a moderate FEC of 459.6 EPG, and farms with both sheep and goats had the highest average FEC of 592 ($P<0.009$) (Wadsworth, 2015). In the 2016 study, the farms with only sheep had the lowest average combined FEC of 753, farms with only goats had average combined FEC of 870, and the sheep and goat farms were once again the highest with FEC of 1,121 ($P=0.40$) (Table V). Although the FEC from year to year cannot be directly compared due to the fact that Wadsworth's data represents the average adult FEC and the Pouliot data represents the average combined FEC (adults + juveniles), the comparison seems to demonstrate a trend. Farms with only sheep tended to have lower fecal egg counts than farms with only goats, and both of these farms tended to have much lower FECs than the farms that tried to have both species.

Both Wadsworth's project and Pouliot's project compared the stocking rates on

the farms, although the two projects split the data up in very different ways. Wadsworth's data was split up into six different subsections and generally showed an exponential trend toward an increase in stocking rate leading to an increase in FEC (Wadsworth, 2015). In the 2016 study, however, the data was split into two subsections and there was no significant difference between the average FEC of both groups (P=0.83) (Table IX). However, the linear regression line that compared the stocking density and the average FEC per farm demonstrated the same trend that was seen in Wadsworth's study: an increase in the number of animals/acre led to an increase in the average FEC per farm (Figure 6).

Wadsworth also tested the effect of deworming methods on *H. contortus* infection levels. The results showed that farms with no dewormer had the highest FEC (mean = 550.9 EPG), and farms that used multiple dewormers had the lowest (mean = 188.7 EPG) (Wadsworth, 2015). The Pouliot study went in a slightly different direction, but did compare farms that used no dewormer with those that used a chemical dewormer. Surprisingly, the farms that did not use a dewormer had much lower FEC (mean = 165) than those that used a chemical dewormer (mean = 828). However, while Wadsworth's study had relatively equal group sizes (22 samples for the no dewormer category, 24 samples for the one dewormer category, and 20 samples for the multiple dewormer category), the samples for the 2016 study were drastically different (Wadsworth, 2015). Only 5 farms identified themselves as using "no dewormer", while 68 farms indicated that they used a "chemical dewormer" (P=0.078) (Table XXI). Therefore, the sample pool was likely quite different from last year to this year, and this could have contributed to the different results.

Finally, Wadsworth's project compared the effects of farm size and reached a very different conclusion from Pouliot's study. Although Wadsworth had predicted that larger farms would have lower FEC due to increased investments into animal welfare and greater knowledge of small ruminant parasitology, the data collected in 2014 actually showed that as the number of animals per farm increased, so did the average EPG (P=0.03). For 2016 study, the farms with 16 or more sheep actually had lower FEC than those with 5-15 sheep (P=0.091) (Table XVIII). Although Wadsworth's study compared animals in general and Pouliot's study compared only sheep, there was a drastic difference. This could also be due to the variable sample pools from year to year, as well as the varying time when the samples were taken and the fact that adult and juvenile populations were both used for this year.

Implications

These results demonstrated that certain management techniques led to lower *H. contortus* fecal egg count levels on farms throughout New England. If producers learn about the management practices that are more effective for sheep and goat flocks, they can implement these changes on their own animals. This will hopefully lead to decreased animal, meat, and wool losses. In addition, the animals will be healthier due to lower levels of parasites.

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Appendix 1: Producer Survey Questions (2015)

2015 Parasite Census for Sheep and Goat Producers

Northern New England Producer survey for 2015 grazing season - NE SARE Grant

Farm name*

Owner(s)*
Name(s)

City or Town*

Mailing Address*
Street address or PO box

Zip Code*

State*

Which of the following best describes your farm's parasite management practices?o*

- Certified Organic
- In the process of becoming Organic
- Use a combination of commercial and "Natural" products
- Use commercial dewormers
- Other:

Email address and farm web site (if applicable)*
Please enter the best email address to contact you at (ex. john.doe@gmail.com)

How many years has your farm been established?*

What is the total number of adult sheep on your farm?*
Animals greater than 1 year old

What small ruminent species do you keep?I*
Goats, sheep or both

- ○ Sheep
- ○ Goats
- ○ Both

What is the total number of adult goats on your farm?*
Animals greater than 1 year old

What are the major products of your herd?*
Check all that apply

- ☐ Fiber/Wool
- ☐ Dairy Products
- ☐ Meat Products
- ☐ Pelts
- ☐ Sales/Breeding Stock
- ☐ Other:

Do you graze your animals?*

- ○ Yes
- ○ No

If you graze, describe your rotational grazing practices (if used)
How often do you change fields? How large are the grazing blocks? Etc.

How many acres of pasture do you have?*

Do you use fecal egg counts as part of your parasite management?*

- ○ Yes
- ○ No

On average, how often does a veterinarian visit your farm?*

If you do not use a veterinarian, why not?

Choose the parasite management practice that best fits your farm.*

Please list all dewormer products you have used in the last year.*
For example: No dewormer, Safeguard Drench, injectable Ivomec, Copper Oxide boluses, etc.

Within the last year, have you had any of the following parasite related problems?*

- [ ] Bottle jaw
- [ ] Loss of lambs or kids to parasites
- [ ] Loss of adults to parasites
- [ ] Coccidiosis
- [ ] Persistent coughing
- [ ] None of the above
- [ ] Other:

Pick the best choice from the list below that describes your experience with FAMACHA.*
FAMACHA is a tool used to diagnose barber pole worm in sheep and goats.

- ( ) Not Familiar with FAMACHA
- ( ) Took FAMACHA training but don't use
- ( ) Took FAMACHA training and use
- ( ) Other:

If you use FAMACHA, how often do you check your animals?

- ( ) Weekly
- ( ) Monthly
- ( ) Once or twice a year
- ( ) Other:

Which of the following best describes your deworming strategy?*

- ( ) Selectively deworm based on FAMACHA or egg counts
- ( ) Deworm all stock after losses or sickness due to parasitism
- ( ) Routinely give dewormers at set times of the year
- ( ) Never deworm
- ( ) Other:

Do you consider FAMACHA scores when selecting replacement animals?
i.e. Would you cull an animal with consistently high FAMACHA scores?

- ( ) Yes
- ( ) No
- ( ) Other:

Describe how you catch your animals for examination.*
Chute system, catch by hand, catch pen, etc.

When did you first turn your animals out on pasture this year?*

- ○ Animals had access to pasture all winter
- ○ March
- ○ April
- ○ May
- ○ Other:

Where do you get most of your parasite/disease management information?*

- ☐ Veterinarian
- ☐ Extension Agent
- ☐ Web Sites
- ☐ Feed Store
- ☐ Other Farmers
- ☐ Other:

Appendix 2: SOP for McMaster Method

Modified McMaster Fecal Egg Count – Standard Sensitivity
(Weber Lab, May 2015)

Materials:

- disposable gloves
- saturated sodium nitrate (Fecasol soln.)
- graduated beaker
- tongue depressor or weigh spatula
- balance with sensitivity of 0.1 mg
- cheesecloth (gauze 3 x 3's work well)
- disposable plastic pipet
- McMasters egg counting slide **
- paper towels
- compound microscope.

Notes:

1. A fresh fecal sample should be collected.
2. For pooled samples, take small amounts from 10 animals, if possible. Keep sample tightly wrapped in plastic to exclude air. Mash and homogenize well before testing.
3. If a larval culture is also being completed, set it up before refrigeration.

Procedure:

1. Weigh out 2g of feces in a small beaker or paper cup.
2. Add 28 ml. of sodium nitrate flotation solution (q.s. to 30 ml) to feces, mix well.
3. Strain through 1 or 2 layers of cheesecloth (or tea strainer), mix well. Squeeze out as much fluid as possible from cheesecloth ball.
4. Immediately withdraw about 1 ml of the suspension with a pipet or syringe and fill both counting chambers of the McMaster slide. Work quickly, stirring as you draw up fluid. Let stand for 1-2 minutes to allow eggs to float to top. If visible air bubbles are present, remove the fluid and refill.

Hints:

- Steps 3 and 4 should be done at same time without letting sample sit between steps
- Once chambers are filled, step 3 can be started for the next sample
- Once filled, the chambers can set for 60 min before counting without causing problems. Longer than this and drying/crystal formation may begin
- Count all eggs inside of grid areas (greater than 2/3 of egg inside grid) using low power (10x) objective. Focus on the top layer, which contains the very small air bubbles (small black circles). Count both chambers. Count eggs from Trichistrongylids (oval shaped, ~ 80 microns long), Tapeworms (irregular to square, less than 70 microns), Strongyloides (oval with larvae w/I egg), ~ 50 microns long), and Nematodirus (oval, >150 microns).

Total egg count (both chambers of McMasters slide) x 50 = EPG (eggs per gram).

Each slide has two chambers, each of which holds a volume of 0.15 mL. Two chambers hold 0.3 ml of fecal mixture, which is 1/100th of the total volume of 30 ml. The number of eggs counted must be multiplied by 100 to calculate the total number of eggs in the sample. However, since you began with 2g of feces, you must multiply by 100 and then divide by 2 to yield eggs per gram.

** Chalex Corporation 5004 228th Ave. S.E., Issaquah, WA 98029-9224
(425) 391-1169; (425) 391-6669 (FAX)
web site: www.vetslides.com
Two-Chambered McMaster Standard Slide

Appendix 3: SOP for Florescently-labeled lectin binding for identification of Haemonchus ova (6/10/15)

Reagents:

- Fluorescein-isothiocyanate coupled peanut agglutinin (PNA-FITC: Sigma L7381-2MG) diluted into 100x concentration (0.5 milligrams per mL) in PBS – store frozen in 50 microliter aliquots
- Fecasol (Sodium Nitrate) solution
- 1 x Phosphate Buffered Saline (PBS) solution (refrigerated)
- 150 mM galactose (150 mM = 52 mg in 2 mL of PBS; 10x concentrate = 520 mg in 2 mL of PBS)

Materials:

- 20 micron mesh nylon cloth (4x4)
- 2- 3x3 gauze pads for each sample
- Small Funnel
- 150 mL Beakers
- 250 mL Erlenmeyer Flasks
- Centrifuge tubes and racks: 50 mL, 15 mL, and 1.5 mL
- Transfer Pipettes
- Pipettors and tips, 1000 uL, 200 uL and 10 uL
- Depression slide
- Aluminum foil

Equipment:

- Balance
- Centrifuge (that can hold 50 mL and 15 mL centrifuge tubes, capable of 1800 x g)
- Mini Centrifuge (14,000 x g-no brake)
- Microscope with incandescent and fluorescent bulb
- External hard drive

Isolation and Purification of Ova:

Ova can be isolated directly from feces or from the Fecasol leftover from a McMaster test:

Fecasol ova isolation from fecal sample:

1. Add 4-8 grams of fecal sample plus 20 mL of Fecasol to small beaker
2. Mash feces thoroughly once softened at room temperature
3. Add another 20 mL of Fecasol solution to fecal matter and blend
4. Set funnel on flask, open 2 gauze pads and arrange in funnel.
5. Filter slurry through gauze. Rinse with additional 10 mL of Fecasol and collect all fluid in beaker. Ball up and squeeze the gauze pad, toss gauze ball in trash when done.
6. Immediately pour liquid into a 50 mL centrifuge tube. Pair and balance tubes with Fecasol solution (use water in a balance tube if odd number of samples) Centrifuge at 1800 x g for 10 minutes. The brake should be set to 0. The ova are floating in this solution and the solids will be in a loose pellet at the bottom of the tube.

Post McMaster ova isolation:

1. Individual samples should have a minimum of 150 e/g.
2. For Witter samples, pool groups according to current study. To pool groups:
    a. Label tubes with the appropriate pool number.
    b. Combine the leftover Fecasol from two samples in a 50 mL tube.

c. Balance and centrifuge the tubes at 1800 x g with the brake off for 10 minutes. The ova are floating in the supernatant and the solids will be in a loose pellet at the bottom of the tube.
d. Pool the samples by pouring all the samples from one group through the same 20 mesh screen as described below.

Removing the ova from the Fecasol

7. Place a funnel on an Erlenmeyer flask, wet a piece of 20 micron mesh screen with tap water and fit into the funnel like a coffee filter carefully arranging the mesh so there are no low points. The mesh can be folded.
8. Slowly pour the top 30-40 mL of supernatant through the mesh without disturbing the pellet in the bottom. The eggs will pool in a cluster in the center of the mesh. Once most of the liquid has passed through, rinse screen using a syringe with 20-60 mL of cold 1x PBS. Rinse with PBS until brown color is gone taking care to unfold the mesh and rinse all the exposed surfaces.
9. Remove nylon from the funnel by picking up all four corners keeping the ova in the center of the mesh. Keep track of the "up" side.
10. Lower the mesh into a 150 mL beaker. Position the mesh so that two corners are in the bottom of the beaker with a transfer pipette tip or a scoopula.
11. Using a syringe with a needle filled with 13mL of PBS, gently spray across top surface of nylon screen so that ova will run down into the beaker.
12. Swirl and Pour this liquid into a 15 mL centrifuge tube. Then rinse the beaker 2 times with about 1 mL of PBS. Add this liquid to the 15 mL centrifuge tube.
13. Balance and centrifuge at 1800 x g for 10 minutes. Make sure centrifuge brake is set to 0. The ova will form a visible pellet in the bottom of the tube.
14. Discard all but 0.5 mL of supernatant. Resuspend ova in remaining liquid. Transfer this liquid to a microcentrifuge tube. Rinse 15 mL tube 2 x with 0.5 mL PBS and add to sample.
15. The Eggs may be preserved at this point by adding 1:1 buffered formalin for long term storage or may be immediately stained.

Note:

- Load the microcentrofuge tubes in with the hinge side out. The centrifuge should be set at 14 x 1000 min^{-1}. To prevent braking on the microcentrifuge, set the timer for 4 minutes, switch off machine at 2 minutes, and let rotor lose speed gradually. Manually set time to zero, turn machine back on and open the lid. The pellet should be plastered against the bottom of the tube on hinge side of the tube making it easier to visualize. Remove the supernatant by withdrawing the liquid from the latch side of the tube.
- Refrigerate any samples that need to sit in between steps to prevent the ova from hatching.

Staining eggs for fluorescent microscopy

1. If the eggs are in formalin, spin down ova in the micro tube and remove the supernatant (see note.) and rinse with 1500 uL PBS.
2. Centrifuge and remove supernatant. Add 500 uL PBS.
3. Add 10 uL of 100x PNA-FITC concentrate to tube. Invert to mix contents. Wrap tubes in foil and incubate in refrigerator for at least one hour or overnight.

Visualizing eggs

1. Centrifuge the ova sample, remove the supernatant and rinse pellet in 1500 microliters of PBS.
2. Centrifuge again and remove supernatant. Resuspend in 100 uL PBS.
3. Cover tubes with aluminum foil and transport to #262 with external drive, rack, PBS, depression slide, 100 uL pipettor and tips.
4. Sign in to use microscope.
5. Ignite fluorescent lamp. (Do not shut off within 15 minutes of turning on. If turned off, allow to sit 5 minutes before reignighting.)
6. Place about 50 microliters from the top of the sample in depression on glass slide. Then resuspend sample and carefully pipette into the middle of the liquid on the slide.
7. Position slide on microscope stage and visualize at 4x. Gently vibrate the slide to push the ova closer together if they are too spread out.
8. Open DP Controller program
9. On the right side of the microscope, under "Olympus Bx60" push the light slide up to the camera symbol
10. Change the objective to10x. (There should be at least 10 ova in the camera field.)
11. On computer, hit play button on DP controller
12. To take incandescent photo, open eyepiece shutter one stop for the camera and refocus the image. (Settings are ISO1600, Auto, image size 1360x1024)
13. To take fluorescent photo, flip the light source to the top (toggle is next to light slide on right side of scope,) pull out top light shutter, set the computer to Manual, adjust speed to about 200-400 ns, focus and take fluorescent photo.
14. Take matching fluorescent and incandescent light photos of at least 100 ova.
15. Save photos on external drive. Label the photos with the accession number, if it is fluorescent or not, and the picture number (ex: "0006-13 fluor 1" matches "0006-13 light 1")
16. Transfer the ova back into their micro tube, rinse the slide and wipe with a Kimwipe.
17. Repeat until all samples are visualized.
18. Shut off microscope, cover and clean up space when done.

Analyzing Photos:

1. Load photos onto iPhoto
2. Count the abomasal fecal ova.
    a. The fluorescent ova must have a green ring or a significant glow.
    b. ¼ of the egg should be in view with a match on the incandescent photo in order to count the egg.
3. Record raw counts on Fluorescent Assay tally sheet.
4. Record final results on the appropriate project Fluorescent summary sheet.

The negative control is 150 mM galactose. Add one part 10x galactose to 9 parts of incubation mixture in microcentrifuge tube just prior to adding PNA-FITC, then complete steps 2 and 3. Eggs that turned green in non-galactose tubes should now be colorless.

Author's Biography

Catherine Werker Pouliot was born in Rochester, New Hampshire, on April 1st, 1994. She grew up in South Berwick, Maine, where she spent most of her time riding dressage or working with the animals on her family's hobby farm. Kate grew up in the Marshwood school system, where she excelled not only as scholar but as a distinguished athlete as well. After graduating from Marshwood High School in 2012, Kate began her undergraduate career at the University of Maine.

During her time at UMaine, Kate maintained a competitive GPA while being an avid member of the Maine Animal Club, the Sophomore Eagles Honor Society, and the All Maine Women Honor Society. In addition, she was the president of the University of Maine Dressage Club and a founding member and president of the Ewe-Maine Icelandic Sheep Club. Kate spent countless hours at the university's Witter Farm, where she helped with milking cows, feeding sheep, and assisting with the lambing process. In May of 2016, Kate graduated from the University of Maine with a Bachelor's Degree in Animal and Veterinary Sciences and a concentration in Pre-Veterinary Medicine, Honors Course of Study. After graduation, Kate will be attending Virginia-Maryland College of Veterinary Medicine on the Virginia Tech campus, where she will be studying to become a doctor of veterinary medicine with a specialization in small animal medicine. Kate hopes to graduate from VMCVM in four years and begin serving the community through her work as a veterinarian.