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A Study of the Prevalence of Gastrointestinal Nematodes in Goats Obtained from Northwest Arkansas

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Abstract

Parasitic helminths have always been an issue in small ruminant production; pathogens that pose great negative impact on goat health and productivity. Insufficient work has been done to document the prevalence of parasitic helminths in the United States, especially in the south and southeast, where the largest goat populations are found.

The aim of this study was to survey the prevalence of infections by gastrointestinal nematodes in goats in Northwestern Arkansas. Gastrointestinal tracts were examined from 41 goats of various locations around Northwest Arkansas. Worm species were identified and population burdens were determined. Coprology was correlated with the actual worm populations.

In descending order, the most prevalent adult nematodes were *Trichostrongylus colubriformis, Haemonchus contortus, Teladorsagia spp., Oesophagostomum spp., Trichuris spp., Nematodirus spathiger* and *Cooperia curticei*. Goats commonly harbored more than one species of nematode. Nematode burdens varied greatly between animals, and respective of management factors.

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1.0 Introduction

Since 1992, the meat goat industry has been a rapidly growing division of livestock production. The primary reason is due to the goat's popularity as livestock and the demand for their products ("Marketing of Meat Goats", Goat 2009). Goat production in the United States has increased rapidly due to the economic value of the goat's ability to convert low quality, undesirable forages into high quality meat, milk, and fiber (Barkley et al., 2012). Demand for goat production in the United States has not only increased due to the high quality of meat but also in response to the ethnic populations in the United States that prefer goat meat and other goat products ("Meat Goats", "Goats", "Meat Goat Ops-USDA APHIS" 2012). As of January 2015, goat and kid inventory in the United States totaled 2.68 million head ("Sheep and Goats" 2015). Eighty percent of the goats in the United States are classified as meat goats, 10% as dairy goats and the remaining 10% as fiber goats (Solaiman, 2007). In 2015, Arkansas' meat goat inventory totaled 38,000 head, unchanged from 2014 ('Arkansas Cattle, Goat and Sheep Report", 2015). Most goats, regardless of their initial use, eventually end up in the meat market ("Marketing of Meat Goats").

There are many important diseases of small ruminants, but none are as pervasive or as direct a threat to the overall health of goats than internal parasites (Kaplan, 2010). This makes control of intestinal parasites the most important health issue for goats of all ages (Barkley et al., 2012; Nye et al., 2004; SARE, 2011; Waller, 2006, Várady et al., 2011; Hoste et al., 2005 and Schoenian, 2009 a). Goats evolved as browsers. They consume a higher percentage of their diet as brush, forbs, leaves, etc. (less desirable plants) than do other ruminants. The majority of forage consumed by goats is located away from the ground, and this helps reduce the ingestion of internal parasites (Barkley et al., 2012 and Fleming et al., 2006). If animals are allowed to browse, their chances of acquiring parasitic larvae diminishes as the grazing distance from the ground increases (Fleming et al., 2006). Goats are generally more susceptible to internal

parasites than sheep because goats have a lower capacity to develop an immune response specific to helminths. The lower innate capacity for an “anti-worm” immune response is most likely the result of their evolution (Hoste et al., 2008; McKenna and Watson, 1987; Lloyd, 1987; Jambre, 1984; and Pomroy et al., 1986). Several studies have illustrated that both the acquisition and the expression of immune responses against nematode species are less efficient in goats than in sheep (Huntley et al., 1995; Pomroy et al.,1986; and Hoste et al., 2008) In today’s goat production, a large percentage of goats are raised as grazers or intermediary browsers. When goats are forced to graze on the same pastures as sheep, the shared helminths may devastate the goat population while sheep are less affected (Pomroy et al., 1986).

Prominent nematodes that infect goats and sheep include: *Haemonchus contortus, Trichostrongylus colubriformis, Teladorsagia circumcincta, Cooperia spp., Nematodirus spathiger, Oesophagostomum spp, Trichuris spp., Dictyocaulus filaria, and Strongyloides papillosus.* These nematodes represent a major group of pathogenic agents which contribute to the losses incurred by the goat industry. The proportions of each of these nematodes in small ruminant populations vary according to host, geographic location, production management, etc.; factors that dictate the overall extent of gastrointestinal parasitisms. Control of internal parasites is of primary concern in any small ruminant health management program and is critical to operational profitability. Naturally infected ruminants usually have mixed infections of different species of nematodes. Goats and sheep share the same species of helminth parasites, however, insufficient work has been conducted to determine the prevalence of parasitic infections of goats in the United States; an initial step in constructing control strategies.

A major problem that the goat industry faces today is that resistance has developed to all the classes of compounds used for worm control in small ruminants. Intestinal parasites have become harder to manage in small ruminants because of the parasites’ increasing resistance to all available chemical dewormers (SARE, 2011). Nematodes negatively impact the animal's

health, reduce productivity, reduce weight gain, reduce performance and increase costs due to poor health (SARE, 2011). Depending on the balance between the parasite populations and the host, parasitic infections can provoke clinical signs and mortality. Pathological importance is primarily related to major production losses in quantity and quality; all induced by the direct effect of worms.

Very few studies have been conducted in the United States, and no studies in Arkansas, to survey the prevalence of internal parasites in goats. According to the Proceeding of the International Symposium in 2006, research with goats is minimal because of the low economic impact that goat products provide and the lack of organization among goat farms. In 2013, Arkansas was ranked number 15 (out of 50) in goat production with 42,000 head (Pinkerton et al., 2013). The majority of the goats in the US are raised in the southern and southeastern states. The southeastern states, including Arkansas, have the most conducive climatic conditions for the growth and establishment of large nematode parasite populations in resident herbivores. The first report of complete failure of all classes of anthelmintics used in small ruminants was made by Kaplan in 2005 at a meat goat farm in Arkansas (Fleming et al.,2006 and Kaplan et al., 2005). Resistance to all three drug classes of anthelmintics is now displayed by all major nematode parasites of sheep and goats throughout the world (Waller, 2006 and Mortensen et al., 2003).

This project was conducted to determine the incidence and prevalence of gastrointestinal nematodes in goats residing throughout northwest Arkansas via coprologic and necropsy examinations. Specific aims of this research were to: a) identify the worm species and population burdens in the goats' intestinal content, and b) to conduct fecal egg counts and larval identifications for correlation with the actual worm populations.

1.1 Nematodes: “Round Worms”

Most nematodes of goats are dioecious and follow the typical *Trichostrongylus,* direct life cycle. The direct life cycle is completed with one host and consists of one egg, four larval stages and mature, reproductive adults. At all stages of development, the nematodes are cylindrical and elongate in appearance. Extreme variations in length are seen with genus, species, sex, and stage of development. Adult females are typically larger than adult males of the same species. The nematodes are very site specific within the gastrointestinal tract and maintain their position primarily by constant motility. Some nematodes attach to the mucosa by oral fixation or wrap themselves around intestinal villi. Some “inactive” larvae embed in the tissue (or crypts) for a varied length of time. All nematodes of small ruminants vary in their activities resulting in pathology and also have varied means by which they are successful in the environment and host.

Reproduction occurs in the gastrointestinal tract. The oviparous female produces eggs that are voided from the host via the feces into the environment. Embryonation occurs immediately if environmental conditions are suitable (temperature, moisture, oxygen). The first stage larvae (L1) hatch out of the egg in approximately 1-2 days. The L1 feeds on bacteria and organic material in the feces. After a few days, the L1 develops and molts into a second stage larvae (L2). The L2 continues to live off the bacteria and organic matter in the feces (Barger, 1999, Smart drenching and FAMACHA integrated training, 2008). After approximately three more days (one week after the egg is passed via feces) the L2 molts but does not ecdysis (cuticle detaches from the larva but the sheath is not shed). The larva is now an en-sheathed, infective third stage larvae (L3). The L3 stage migrates from the feces and migrates onto the forage (negative geotropism). A grazing goat then ingests the L3 on the forage, beginning prepatency.

Once inside the rumen of a host animal the L3 sheds its protective sheath. The L3 is carried to its predilection location in the GI tract (abomasum, small intestine or large intestine)

and starts subsequent development. Within 7 days post infection, the L3 undergo the third ecdysis and develop into a parasitic fourth stage larvae (L4). Approximately 7-20 days post infection, the fourth and final ecdysis occurs as the L4 develops into an early adult (parasitic fifth stage larvae (L5)). After about 7 more days, the nematode is mature. The prepatent period (infection to egg production) is typically around 21 days, but can range from 15-40 days, post infection. Natural death of nematodes typically occur 1 to 10 months after the adult stage is reached (Yazwinski and Tucker, 2006). Infection is replenished by ingestion of the infective L3 by a grazing goat on a daily basis.

1.2 Abomasum Nematodes

Teladorsagia circumcincta (brown stomach worm), Figures 6a, 6b and *Ostertagia trifurcata*, Figure 7, have males measuring 7.5-8.5 mm long and females measuring 9.5-12 mm long. These worms thrive in cool, wet ambient environments. These worms follow the general trichostrongyle life cycle. *T. circumcincta*, are “grazers” as they feed on the nutrients in the mucus. The primary symptom of infection is diarrhea, due to the damage done to the stomach lining (interfering with protein digestion and the host’s appetite). An infection with *T. circimcincta* is commonly considered a production disease because the animals do not grow very well. This worm enjoys its greatest populations in the northern tier of the US.

Haemonchus contortus (barberpole worm), Figure 8a and 8b, is the most important and problematic nematode found in small ruminants and is mostly found in significant numbers in the southern states (Kaplan, 2010). This large, voracious hematophagic worm measures 18-30 mm long and is readily visible on the surface of an opened abomasum. It is known as the barberpole worm due to the appearance of the female’s white ovaries that twist around the red, blood filled intestine. Females are very prolific egg producers (~3,000 eggs/day/female), making them the most fecund nematode in ruminants. *H. contortus* is found primarily in tropical and subtropical regions. They thrive under hot environmental conditions; being very successful in the southeast US. Due to global warming, *H. contortus* is being found more and more north in

the US. These worms follow the general trichostrongyle life cycle, with developmental inhibition occurring during the winter season (arrestment during the L4 stage). Transmission is the lowest during the winter, increases in the spring (spring and post-parturient rise) with the warmer temperature and moisture and peaks during the summer followed by a decrease in the fall. Animals with a *H. contortus* infection show symptoms associated with anemia (pale mucous membranes, bottle jaw, and hydrothorax). Blood loss can lead to death of the animal.

1.3 Small Intestine Nematodes

Trichostrongylus colubriformis (the bankrupt worm), Figures 9a and 9b, is the predominant small intestine worm of sheep and goats. These small, thread-like worms measure approximately 4.3-8.6 mm long and are found throughout the US. The males have a large bursa with unequal, dark brown spicules and the females have a slit shaped vulva without distinctive exterior lips. Both sexes have an excretory pore on the neck. These nematodes thrive under cool and wet conditions. In small ruminants, this worm is generally the next most common and important after *H. contortus*. *T. colubriformis* follows the general trichostrongyle life cycle. Once in the small intestine, *T. colubriformis* feeds on nutrients in the mucosa, thereby causing irritation to the mucosa and interference with digestion. Diarrhea, swelling of the intestinal wall and edema, are common with large infections. The worm is called the bankrupt worm because death of an animal is uncommon but the animal develops poor condition, leading to production and income loss.

Cooperia curticei, Figures 10, is rare and relatively unimportant in small ruminants in the US. These worms follow the general trichostrongyle life cycle and are mildly pathogenic, with no extensive tissue invasion. These true ‘grazers’ live in the small intestine and suck on the mucosa and villi. They will wind around the intestinal villi, causing villar constriction and rejection (thigmokinetic effect).

Nematodirus spathiger (the thread necked worm), Figure 11, is a large worm found in the small intestine and is found throughout the US, usually in small numbers. These worms

follow the general trichostrongyle life cycle, with some unique variations. The L1, L2, and L3 stages develop and stay inside the egg, conferring high environmental resistance. Within the egg, the larvae have the ability to exist on contaminated pastures for 2 years (Yazwinski, unpublished 2012). On pasture the L3s hatch out of the egg due to proper environmental conditions (time, temperature, moisture, etc.). In the small intestine, adults strangle and atrophy the villi, triggering the thigmokinetic effect (villus rejection); this can cause diarrhea that leads to production loss. *Nematodirus spp*. infections are limited to younger animals; a condition primarily due to the animal's age and not induced immunity.

Strongyloides papillosus (intestinal thread worm), Figure 12, is a unique worm found in the small intestine of sheep, goats and cattle around the US. This worm has the ability to adjust to its environment by alternating free-living and parasitic life cycles (heterogonic and homogonic cycles, respectively), with only females being parasitic. The cycle executed is dependent on the environment that the infective larvae encounter. If the free-living environment is good (wet) the heterogonic cycle will predominate. If the free-living environment is bad (dry) the homogonic cycle will predominate. Parthenogenetic females in the small intestine produce small, light colored embryonated eggs and the eggs pass out in the feces. In the homogonic cycle; environmental stages transverse to the filariform. Following the heterogonic cycle; environmental stages include heterogonic males and females (free-living adult males and females). Progeny of these adults are larvae that develop into infective L3s with the filariform esophagus. No matter the cycle, the infective L3s penetrate through the skin and migrate directly into the host's blood stream. Transmammary infection has also been demonstrated but is probably rare. Larvae in the blood stream break into the mammary glands of the lactating animal and infect the offspring. Larvae are carried to the lungs from the blood, coughed up, swallowed and are passed to the small intestine. Pathogenesis of these worms is small intestine enteritis and diarrhea.

1.4 Large Intestine/Cecum Nematodes

Oesophagostomum spp. (the nodular worm), Figure 12, are “large” worms and are found throughout the US in relatively low numbers. They follow the general trichostrongyle life cycle, with a few variations. The infective L3 will infect *per os* or penetrate through the skin. Those that infect transcutaneously go through a tracheal migration and end up in the small intestine. The L3 larvae penetrate deep into the mucosa of the small intestine and nodules form around the L4s. Animals will not develop nodules the first time they are infected with *Oesophogostomum spp*., only upon a challenge infection. L4s in the nodules will either die or break out of the nodules to migrate and reside in the large intestine as adults. Adults and L4 feed on the host blood and tissue which contributes to the overall anemia of the host. Females are 13-24 mm long and males are 11-16 mm long, both with cephalic vesicles. Conditions associated with an active *Oesophogostomum spp*. infection include bloody, tarry diarrhea (caused by the L4s leaving the nodules) emaciation and weakness.

Trichuris spp. (the whipworm), are usually found in relatively low numbers in the cecum. Males measuring 50-80 mm long have a coiled body and females measuring 30-70 mm long have a banana-shaped body. The anterior end of the whipworm is thread-like and is used to thread the worm into the mucosa; making them hard to clear from an animal with most anthelmintics. *Trichuris spp*. are haematophagic and a large population can contribute to the overall anemia of the host. Usually, however they are relatively non-pathogenic. They follow the general trichostrongyle life cycle, except that the infective larva is the second stage (L2) and it stays inside the egg until eaten, and hence very resistant to the environment.

1.5 Cestodes: “Tapeworms”

Moniezia expansa is the primary tapeworm that infects the small intestine of small ruminants in the United States. These tapeworms vary in length due to immunity, cestocial treatment, “worm pressure” and age of the worm. The appearance of the strobilar (adult) form is completely different from the metacestode (larval) form. This hermaphroditic worm completes an

indirect life cycle. The intermediate host (orbatid mite) ingests expelled tapeworm eggs and eventually harbors the cysticercoid stage. The infection is transmitted when the definitive host consumes the infected mite. Once inside the definitive host intestinal tract, the protoscolexes are released from the cysticercoids and they attach to the intestinal wall. The scolex (head) of the tapeworm actually attaches the worm to the wall of the small intestine via four suckers. The neck of the scolex grows the proglottids of the worm. The adult tapeworm maintains its position via the suckers, adhesion of the flat strobilus to the mucosa, and winding with the curves of the GI tract. The strobilus “feeds” via diffusion through its microtriche tegument (cuticle), thereby absorbing nutrients from the host’s digested feed. Many producers are alarmed by tapeworm infections in their animals because the white segments (proglottids) are visible on the feces of an infected host. In truth however, very little damage is caused by normal (small) tapeworm infections. Heavy infections may reduce growth rates in kids and may cause intestinal blockage but these conditions are rarely seen.

1.6 Trematodes “Flukes”

Fasciola hepatica is the liver fluke and causes fascioliasis in ruminants. According to Martinez-Moreno et. al goat fascioliasis is less frequent and less important than infections in sheep and cattle (Martinez-Moreno et. al., 1999). *F. hepatica,* infection is not a concern for small ruminant producers in the Northwest region of Arkansas due to fluke life cycle requirements. The pastureland in which animals are grazing must be partially aquatic for a good portion of the year; a circumstance more of a concern in the southeastern states of the US. Liver flukes vary in size due to immunity and age of the worm. This hermaphroditic worm completes an indirect life cycle by using an active, semi-aquatic snail as an intermediate host. Leaf shaped adults maintain their position in bile ducts via suckers, cuticular hooks, molding to the shape of the surrounding environment, and becoming larger in size than their current location. These parasites reside in and cause damage in the liver. Pre-adults have a continuum of growth until they mature and wedge into the collecting bile ducts of the liver. Infection by *F. hepatica* in goats

usually develops into a chronic disease. This was confirmed in a study by Martinez-Moreno et. al who showed that an immune response occurs in goats but the goats never develop complete resistance, resulting in unthriftiness of the host, weight loss and sometimes death.

1.7 Anthelmintics: Classes of Anthelmintics

A. Benzimidazoles:

Fenbendazole (Safeguard and Panacur) and albendazole (Valbazen) are the two most commonly used Benzimidazoles in goats. Fenbendazole has a wide margin of safety but albendazole can be embryo-toxic (teratogenic). The benzimidazoles are known as the “white wormers”, due to their white appearance.

Benzimidazoles kill helminths by disrupting microtubule formation. Currently in the United States, there are high levels of resistance to the benzimidazoles by both *H. contortus* and *T. colubriformis* populations (Howell et al., 2008). Producers should use benzimidazoles to control gastrointestinal nematodes only if their worm burdens have been shown to be drug responsive/susceptible (FECRT).

B. Imidazothiazole/tetrahydropyrimidine:

Levamisole (Levasol, Tramisol, and Prohibit) kills gastrointestinal nematodes by depolarizing nicotinic neuromuscular junctions. It also acts as a cholinergic agonist in mammals, which is the reason for its narrow therapeutic index (Williamson, 2013). It is very important that animals be properly weighed and dosed when using levamisole (as well as any other anthelmintic). Animals should not be fasted prior to administering levamisole because toxicity is a concern. There are some populations of *H. contortus* in the U.S. that are still susceptible to levamisole (Howell et al., 2008 and Williamson, 2013).

Morantel tartrate (Rumatel) is a tetrahydropyrimidine drug. It also acts as a cholinergic agonist, but at a less potent level and has a larger margin of safety.

C. Macrocyclic Lactones (ML)

This group is composed of 2 groups; avermectins (ivermectin, doramectin, eprinomectin) and milbemycins (moxidectin). The primary activity of the MLs is directed at the glutamate-gated ion exchange gates in the cellular membrane of the nerves and muscles of the nematodes. MLs cause flaccid paralysis of the nematode by interfering with neurotransmission and muscle cell junction. The antiparasitic effect is mediated through selective binding to glutamate-gated chloride ion channels. MLs are lipophilic and do not cross the blood brain barrier in most mammals. MLs have a wide safety margin in mammals. According to S Howell, *H. contortus* in populations are already resistant to ivermectin and in the process of becoming resistant to Moxidectin (Howell et al., 2008).

The number of FDA approved drugs for goats is very limited; morantel (rumatel), thiabendazole (omnizole, no longer marketed), fenbendazole (Safeguard and Panacur) and phenothiazine (feno-drench suspension) which is no longer available in the USA (Kaplan, 2010). Effective control of gastrointestinal nematodes in goats can usually only be accomplished by using drugs in an extra label manner and with the assistance of a licensed veterinarian. Unapproved drugs that can be effective for the treatment of gastrointestinal nematodes in goats include ivermectin, doramectin, moxidectin (Cydectin), albendazole and levamisole.

Goats metabolize (detoxify) drugs much more rapidly than other livestock, thereby requiring high dosing (Kaplan, 2010). Depending on the anthelmintic being used, goats require 1.5-2 times the dose recommended for effectiveness on the label for sheep (SARE, 2011).

Reasons for resistance to develop against anthelmintics include under dosing the animal, rotating drugs too rapidly, dosing animals too frequently, non-strategic dosing, etc. Nematode resistance is genetically conferred. The use of chemical anthelmintics selects for resistance in the nematode population over time. There is a need to balance chemical intervention with proper management. Anthelmintics should only be administered to animals that

need treatment. The animals in the herd that remain untreated harbor gastrointestinal nematodes that will stay more susceptible to anthelmintics (refugia), thereby helping prolong chemical effectiveness.

2.0 Materials and methods

The following materials and methods were used throughout the entirety of the survey for each study animal used.

2.1 Necropsy and intestinal helminth collection

Forty-one gastrointestinal tracts from goats were collected between October 2013 and March 2015. All tracts were collected immediately post slaughter at local processing plants or farms and transported to the University for immediate processing. Processing of all intestinal tracts were conducted according to the W.A.A.V.P. guidelines (Wood et al., 2010). Immediately after animal demise the omasal and pyloric ends of the abomasum, in addition to the ileocecal junction, were ligated using heavy cotton string; thereby preventing the movement of contents (and nematodes) from their proper locations within the gastrointestinal tract. The three relevant sections of the GI tract (abomasum, small intestine and large intestine/cecum) were separated and placed into separate basins. If available, a fecal sample was collected directly from the rectum for coprology (fecal egg counts, coproculture, larval harvest and identification).

The abomasum was opened longitudinally and the contents collected into a graduated bucket. The opened abomasum was thoroughly rinsed and washed between each fold by hand. The rinse water and contents were combined in the bucket and brought up to 2L using tap water for aliquot retrieval. The cleaned abomasum was then covered with water and placed in a refrigerator to soak overnight.

The mesentery around the small intestine and large intestine was removed. The small intestine was opened along its entire length and the contents collected into a graduated bucket. The small intestine was then rinsed and “stripped” by hand. The rinse water and contents were

combined in a bucket and brought to 4L by added water for aliquot retrieval. The cecum and one-third of the length of the large intestine (from the illeocecal junction) was processed the same way as the small intestine. The content and rinse water were brought to 2L by adding water for aliquot retrieval. Visible adult worms (*Trichuris* spp.) were detached from the cecum and added to the collected contents. No gall bladders or livers were collected to search for *Fasciola hepatica* infections. Lungs were not collected for *Dictyocaulus filaria*.

2.2 Intestinal content preservation

Five percent aliquots of the abomasum, small intestine and large intestine/cecum contents were removed during vigorous, constant stirring. The separate aliquots were formalized using a sufficient amount of 10% formalin and placed at room temperature until nematode isolation and identification. The abomasum (after the overnight soak) was thoroughly stripped by hand to ensure all the mucus had been dislodged and made part of the soak collection. One hundred percent of the soak collection was formalized and placed at room temperature until nematode isolation, identification, and quantification.

2.3 Intestinal helminth isolation,identification and quantification

The aliquots were washed over appropriate mesh sieves; abomasum content, No. 100 (aperture of 150 μm), abomasum soak, No. 400 (aperture of 38 μm), small intestine content, No. 120 (aperture of 125 μm), and large intestine content, No. 60 (aperture of 250 μm).

Subsamples of suspended (appropriately stirred) sieved residues were examined under a stereoscopic microscope at 10-60X for parasite isolation and counting (approximately 20 mL subsamples from a measured amount). Most of the adult and L4 parasites were identified using the stereoscopic microscope. Adults and L4s that could not be accurately identified were mounted in lacto-phenol for identification and counting using a compound microscope at 40-200X. All parasites were identified to genus, species (if possible), sex and stage of development. Adult and larval identifications were based on Van Wyk and Mayhew, 2013.

2.4 Fecal egg per gram count: Direct fecal flotation

One gram of feces was homogenized in 10 mL of saturated magnesium sulfate (MgSO4), and poured over a wet sieve (1mm aperture). The filtrate was poured into a 15 mL plastic centrifuge tube, and additional MgSO4 was added until a slight meniscus was visible over the rim of the tube. One glass coverslip was gently placed on the test tube. The tube was centrifuged for three minutes. The coverslip was then placed on a glass microscope slide and examined at 40-100X for the adhered egg counts. Using a compound microscope, eggs were identified and counted as strongyle, *Trichuris spp*., or *Nematodirus spp*. Eggs (Figure 14). The presence of Strongyloides and Moniezia eggs were noted.

2.5 Fecal coprocultures and harvesting of infective larvae

Samples of feces with an EPG greater than 20 were soaked in water, until softened, and thoroughly homogenized with vermiculite to yield a moist, standardized mixture. The fecal-vermiculite mixture (coproculture) was formed into a concave depression inside a 16 oz plastic cup. Multiple vertical ridges were pressed into the mixture to increase the available surface area. The culture was then covered with foil and allowed to stand at room temperature for 12-14 days before L3 harvest.

For larval harvest a one-inch section of the solo cup rim was scratched. Water was then added to the cup until a slight meniscus protruded over the rim of the cup. A Petri plate was inverted over the cup to form a seal. Using proper technique, the cup and Petri dish were inverted and left at an incline, with the scratched area facing the lowest point (for L3 to escape into the petri dish). Water was added to the Petri dish until the scratched area of the rim was fully covered. The culture was left undisturbed for over three hours. The water in the Petri dish was collected using a pipette and transferred to a glass centrifuge tube. Using a water squeeze bottle, the empty Petri dish was rinsed and the water was collected and placed into the same glass centrifuge tube. The L3 collection test tube was placed, uncovered, in a refrigerator for one day (to allow for L3 settling).

2.6 Larva preparation for identification

Once the L3s had settled overnight, the supernate was carefully discarded using a pipette, leaving about 4mls of water in the bottom of the tube. To kill the precipitated larvae, an equal amount of formalin was added to the larval precipitation and agitated by hand. To straighten out the L3, the suspension was heated over a flame until a transient boil. The killed and straightened L3 were centrifuged for 15 minutes. The top fluid was discarded via pipetting down to the L3 pellet. Using a pipette, the larvae were suspended, in the remaining liquid, and a drop of the larval suspension was placed on a glass microscope slide and covered with a coverslip.

2.7 Larva identification

Using a compound microscope, the genus-specific identification of the first 100 L3 per sample was accomplished using the length of the tail of the sheath (STE), the head shape, and overall L3 characteristics (Figure 15). Larvae were identified based on the published, detailed features. (VanWyk et al., 2013).

3.0 Results

The month from which a goat intestinal tract was harvested and categorized into a specific season is represented in Table 1. Spring season; March through June, includes 18 intestinal tracts, animal numbers 11-28. Summer season; July through October, includes seven intestinal tracts, animal number 1-5 and 29-30. Winter season; November through February, includes 16 intestinal tracts, animal number 6-10, 31-41.

Table 1
Goat intestinal tracts harvested during each season of the year over the duration of the study.

Animal #	Harvest Date	Season
1	10/18/2013	Summer
2	10/18/2013	Summer
3	10/18/2013	Summer
4	10/18/2013	Summer
5	10/18/2013	Summer
6	1/20/2014	Winter
7	1/20/2014	Winter
8	1/20/2014	Winter
9	1/20/2014	Winter
10	1/20/2014	Winter
11	3/25/2014	Spring
12	3/25/2014	Spring
13	3/25/2014	Spring
14	3/25/2014	Spring
15	3/25/2014	Spring
16	6/4/2014	Spring
17	6/4/2014	Spring
18	6/4/2014	Spring
19	6/4/2014	Spring
20	6/7/2014	Spring
21	6/7/2014	Spring
22	6/7/2014	Spring
23	6/30/2014	Spring
24	6/30/2014	Spring
25	6/30/2014	Spring
26	6/30/2014	Spring
27	6/30/2014	Spring
28	6/30/2014	Spring
29	10/29/2014	Summer
30	10/29/2014	Summer
31	12/10/2014	Winter
32	12/10/2014	Winter
33	12/17/2014	Winter
34	12/17/2014	Winter
35	12/17/2014	Winter
36	12/17/2014	Winter
37	2/5/2015	Winter
38	2/5/2015	Winter
39	2/11/2015	Winter
40	2/11/2015	Winter
41	2/11/2015	Winter

Note: Spring season includes the months of March - June
Summer season includes the months of July - October
Winter season includes the months of November - February

Of the 41 acquired intestinal tracts, fecal egg counts (FEC) were determined from 39 individual intestinal tracts. L3 larvae was harvested and quantified from 35 of those 39 fecal samples, with results reported in Table 2. All 39 fecal samples examined were positive for strongyle eggs. Strongyle egg per gram (EPG) counts ranged from 5 to 16,650 EPG. Strongyle egg counts in the summer season ranged from 9-271 EPG, spring season samples ranged from 20-16,650 and winter season samples ranged from 5-2,769 EPG. *Nematodirus spp.* and *Trichuris spp.* eggs were present in a small portion of the fecal samples; 10% and 23%, respectively. *H. contortus* was the highest percentage of harvested L3 followed closely by *T. colubriformis*. *Oesophagostomum spp.* L3 was present at a low number in 7 out of the 35 coproculture samples.

Table 2
Egg per gram (EPG) counts and genus specific percentages (%) of harvested coproculture infective larvae (L3).

	EPG as:			% of L3 as:		
Animal #	Strongyle	Nematodirus spp.	Trichuris spp.	H. contortus	T. colubriformis	Oesophagostomum spp
1	271	1	0	68	32	0
2	-	-	-	-	-	-
3	209	0	0	65	35	0
4	54	0	0	42	58	0
5	104	0	0	50	50	0
6	44	0	0	-	-	-
7	5	0	0	0	100	0
8	47	0	0	62	38	0
9	68	0	0	4	96	0
10	91	0	0	88	12	0
11	160	0	4	63	37	0
12	206	0	2	89	11	0
13	1248	0	0	50	50	0
14	138	0	0	96	4	0
15	20	0	2	F.L.	F.L.	F.L
16	176	0	0	99	1	0
17	195	0	0	98	2	0
18	800	0	0	97	3	0
19	170	0	0	93	7	0
20	113	6	3	88	22	0
21	48	3	60	3	15	0
22	445	18	51	71	19	0
23	2600	0	0	50	49	1
24	5850	0	0	61	35	4
25	16650	0	0	91	9	0
26	12650	0	0	86	14	0
27	8350	0	0	81	17	2
28	1550	0	0	8	88	4
29	27	0	3	-	-	-
30	9	0	0	-	-	-
31	2769	0	0	50	50	0
32	437	0	0	27	73	0
33	1185	0	0	10	90	0
34	117	0	0	3	97	0
35	1500	0	15	19	81	0
36	212	0	1	14	33	53
37	29	0	0	16	56	28
38	67	0	0	22	78	0
39	1504	0	0	0	100	0
40	272	0	0	56	36	8
41	-	-	-	-	-	-

Note: Fecal egg counts were quantified using direct flotation with $MgSO_4$ of fecal filtrate from one gram of feces collected directly from the rectum of each study sample.
FL represents free living larvae and - represents no sample available

One hundred percent of the goat intestinal tracts surveyed were positive for at least one species of adult nematode. The predominant adult nematodes found throughout the survey were *T. colubriformis* and *H. contortus*. *T. colubriformis* had large populations in samples obtained from the spring and winter seasons. Samples 22 through 28, obtained during the spring season, had the highest number of *H. contortus* adults and immatures. During the summer season is when *T. colubriformis* was at its lowest population. *C. curticei* was found in one small intestine sample during the spring season. In the large intestine, small populations of both *Trichuris spp.* and *Oesophagostomum spp.* were present throughout all seasons.

Table 3
Calculated, total count of adult nematodes in each section of the intestinal tract.

	Abomasum (content + soak)				S.I. Content			L.I. Content	
	H. contortus	*T. circumcincta*	*Teladorsagia spp.*	*Ostertagia spp.*	*T. colubriformis*	*N. spathiger*	*C. curticei*	*Trichuris spp.*	*Oesoph spp.*
Animal #	Adults	Adult Males	Adult Females	Males	Adults	Adults	Adults	Adults	Adults
1	100	40	0	0	320	0	0	0	0
2	80	20	140	20	240	20	0	60	20
3	120	160	240	0	540	0	0	0	0
4	220	0	60	0	380	0	0	0	0
5	180	40	20	0	340	20	0	0	0
6	19	13	9	4	0	0	0	0	0
7	0	0	0	0	0	20	0	0	0
8	80	0	0	0	0	0	0	0	0
9	40	20	0	0	60	0	0	0	0
10	228	22	20	20	60	0	0	0	0
11	494	23	0	0	1800	0	0	20	0
12	89	0	0	0	20	0	0	20	0
13	2704	62	402	22	11440	0	0	0	0
14	422	1	0	0	160	80	0	0	0
15	40	0	0	0	60	20	0	60	20
16	171	2	0	0	20	0	0	0	40
17	164	0	2	0	20	0	0	20	260
18	439	0	20	1	0	0	0	0	260
19	262	0	0	0	20	0	0	0	60
20	278	31	27	7	140	240	0	0	0
21	80	1	2	21	80	40	0	320	0
22	2335	441	914	144	820	440	60	220	0
23	14840	180	380	0	11720	0	0	20	360
24	2700	220	380	40	20480	0	0	40	200
25	19040	350	400	0	21080	0	0	20	320
26	19470	80	80	0	18560	0	0	40	240
27	9380	40	100	0	18800	0	0	0	320
28	9432	126	106	20	8020	0	0	0	60
29	43	0	0	0	160	0	0	20	0
30	204	0	0	0	80	0	0	20	0
31	1820	0	20	0	48800	0	0	80	0
32	490	10	30	0	20240	0	0	20	0
33	180	0	20	0	20800	0	0	0	0
34	0	0	0	0	3720	0	0	0	0
35	180	0	40	0	45000	0	0	160	0
36	80	50	30	0	860	0	0	220	100
37	100	1	0	20	8420	0	0	0	60
38	20	40	140	0	5500	0	0	0	20
39	241	0	0	0	24920	0	0	0	0
40	20	0	0	0	540	0	0	20	0
41	0	0	0	0	33220	0	0	0	0
Total	86785	1973	3582	319	327440	880	60	1380	2340

As shown in Table 4, goat intestinal tract samples obtained during the winter months of November through February showed the highest adult strongyle worm burdens. The summer months of July through October showed the lowest adult strongyle worm burdens. The total combined adult strongyle worm burden for the individual animals varied, ranging from 0 to 50,640 adults. All samples with high strongyle EPG counts had either a higher number of *H. contortus* and/or *T. colubriformis* adults in the intestinal tract. Immature strongyle worms were the most prevalent in the spring season followed by the winter season (Table 5). *H. contortus* immature worms were the most prevalent followed by *T. colubriformis.*

Table 4

Calculated total adult strongyle worm burden per study sample.

Season	Animal #	*H. contortus* Adult	*T. colubriformis* Adult	*Oesophagostomum spp.* Adult
Summer	1	100	320	0
Summer	2	80	240	20
Summer	3	120	540	0
Summer	4	220	380	0
Summer	5	180	340	0
Winter	6	19	0	0
Winter	7	0	0	0
Winter	8	80	0	0
Winter	9	40	60	0
Winter	10	228	60	0
Spring	11	494	1800	0
Spring	12	89	20	0
Spring	13	2704	11440	0
Spring	14	422	160	0
Spring	15	40	60	20
Spring	16	171	20	40
Spring	17	164	20	260
Spring	18	439	0	260
Spring	19	262	20	60
Spring	20	278	140	0
Spring	21	80	80	0
Spring	22	2335	820	0
Spring	23	14840	11720	360
Spring	24	2700	20480	200
Spring	25	19040	21080	320
Spring	26	19470	18560	240
Spring	27	9380	18800	320
Spring	28	9432	8020	60
Summer	29	43	160	0
Summer	30	204	80	0
Winter	31	1820	48800	0
Winter	32	490	20240	0
Winter	33	180	20800	0
Winter	34	0	3720	0
Winter	35	180	45000	0
Winter	36	80	860	100
Winter	37	100	8420	60
Winter	38	20	5500	20
Winter	39	241	24920	0
Winter	40	20	540	0
Winter	41	0	33220	0

Table 5
Immature nematode worm counts.

Animal #	*H. contortus*	*Teladorsagia spp.*	*T. colubriformis*	*Cooperia spp.*	*Oesophagostomum spp.*
1	0	0	0	0	0
2	0	0	0	0	0
3	0	0	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	7	0	0	0	0
7	0	0	0	0	0
8	0	0	0	0	0
9	2	0	0	0	0
10	45	0	0	0	0
11	6	1	20	0	0
12	2	0	0	0	340
13	1187	4	560	0	0
14	1363	22	0	0	0
15	1	0	0	0	0
16	0	0	0	0	880
17	0	0	0	0	500
18	2	0	0	0	540
19	0	0	0	0	120
20	476	31	60	0	0
21	206	2	20	0	80
22	1836	305	280	40	80
23	4180	230	460	0	60
24	2060	380	760	0	40
25	14560	880	540	0	40
26	19190	420	400	0	60
27	20980	280	340	0	0
28	3400	123	80	0	0
29	35	0	80	0	0
30	20	0	0	0	0
31	400	0	2600	0	0
32	2660	30	200	0	60
33	25	20	200	0	0
34	724	0	20	0	0
35	290	0	300	0	0
36	440	0	0	0	120
37	3230	209	120	0	100
38	980	50	40	0	40
39	87	0	640	0	0
40	0	0	0	0	0
41	0	0	980	0	0

Figures 1 through 3 depict the populations of adult *H. contortus* in the intestinal tracts of samples obtained in the winter, spring, and summer seasons, respectively, compared to the calculated *H. contortus* EPG of the fecal samples determined from those same samples.The greatest populations of *H. contortus* were found in the samples obtained during the spring season. All 18 samples were positive for adult H. contortus, ranging from 40-19,470 adults. Of the 16 fecal samples collected all were above 99 EPG. In the winter season samples 13 out of 16 samples were positive for *H. contortus* adults, ranging from 0 to 1820 adults. Out of the 14 winter fecal samples only 6 had a calculated *H. contortus* EPG of over 30. For the summer season samples an EPG was conducted from 4 out of the 7 samples. All summer samples had an *H. contortus* adult count ranging from 43-220.

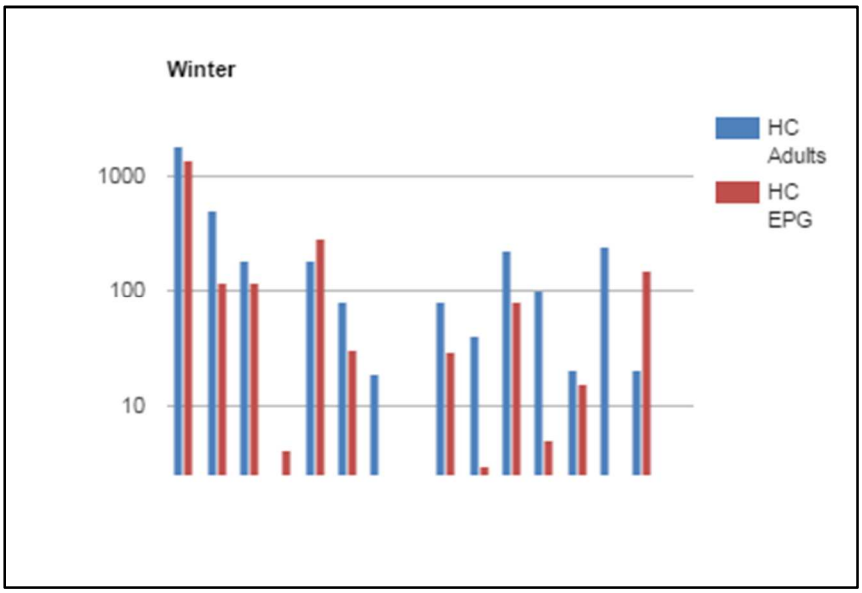

Figure 1
H. contortus adult count compared to calculated *H. contortus* EPG of winter season samples.

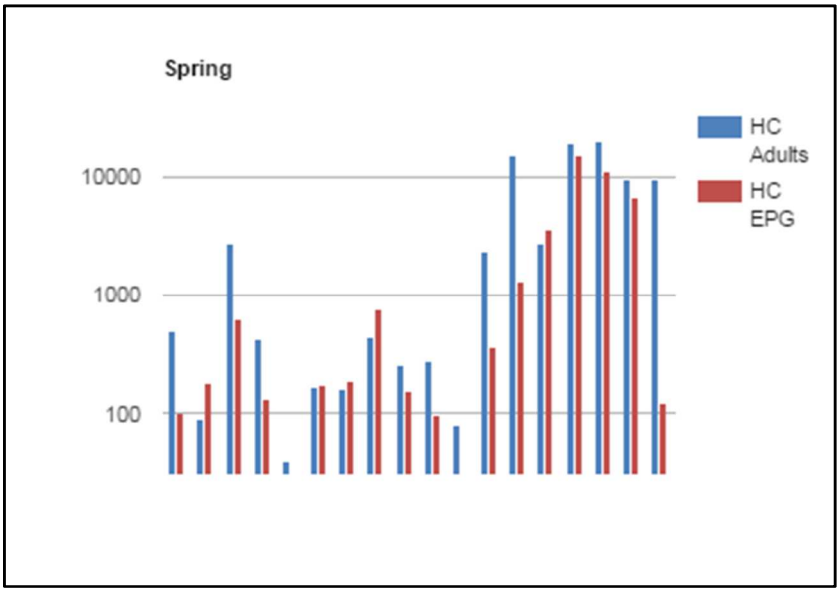

Figure 2
H. contortus adult count compared to calculated *H. contortus* EPG of spring season samples.

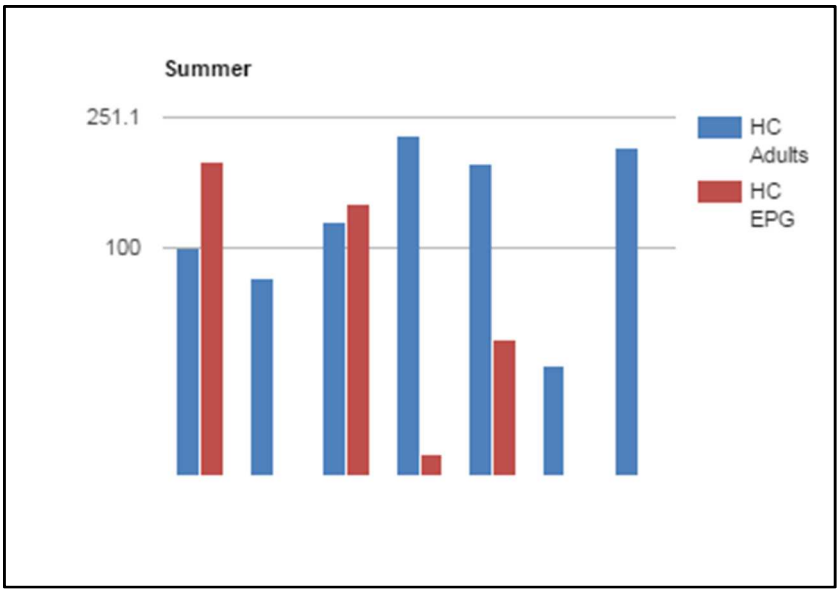

Figure 3
H. contortus adult count compared to calculated *H. contortus* EPG of summer season samples

Figure 4 and Figure 5 represent the regression analysis correlation that was calculated between the species specific adult nematode and the species specific calculated EPG for *H. contortus* and *T. colubriformis*, respectively. The coefficient of determination (R^2) of *H. contortus* for all samples obtained through the study is .71. The R^2 of *T. colubriformis* for all samples obtained throughout the study is .60. The trendline equation for *H. contortus* is represented by y= 0.5189x -47.825. The trendline equation for *T. colubriformis* is represented by y= 0.0401x +133.42.

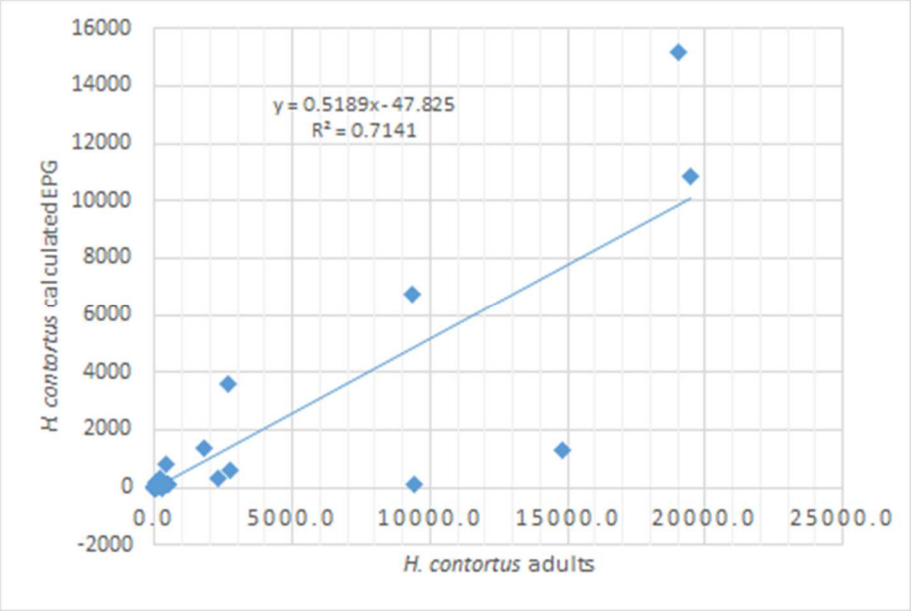

Figure 4

Regression analysis correlation of adult *H. contortus* and calculated *H. contortus* EPG.

Note: equation y=0.5189x-47.825 is the Linear Regression Equation

R^2=0.7141 is the coefficient of determination

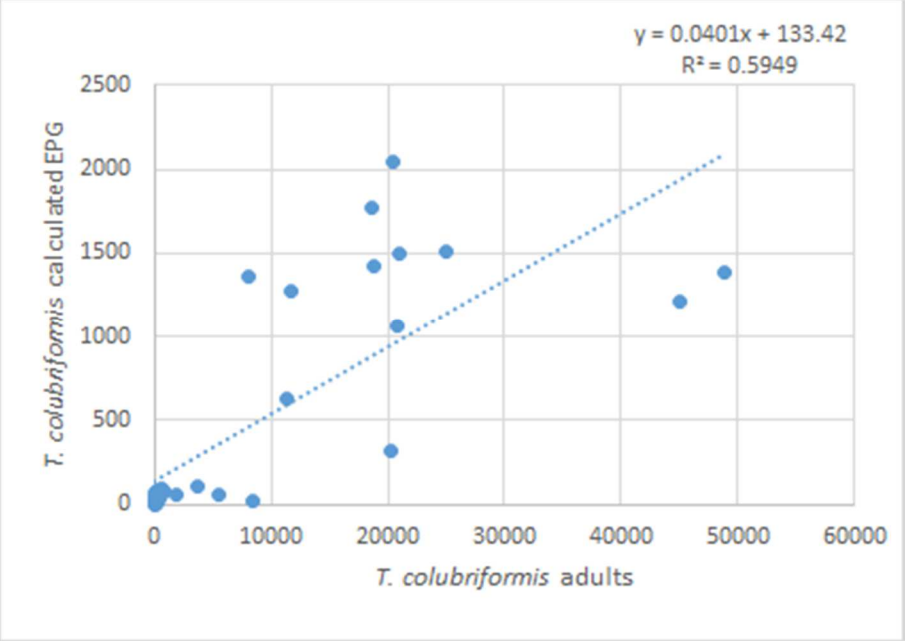

Figure 5

Regression analysis correlation of adult *T. colubriformis* and calculated *T. colubriformis* EPG.

Note: equation y=0.0401x + 133.42 is the Linear Regression Equation

R^2=0.5949 is the coefficient of determination

4.0 Discussion

Under natural environmental conditions, goats commonly harbor more than one species of nematode. The nematode burden of mature and immature worms varies greatly among animals in the same season, as shown in this study. The degree of nematode infection acquired by goats is determined by natural and management factors, seasonal and environmental conditions, grazing behavior, previous exposure to nematodes, physiological state of the goat, stocking rate, nutrition, age of the goat, and previous anthelmintic treatment.

As seen in this survey, worm burdens are not evenly distributed within the animal population. It is a rule of thumb that 20-30% of the animals in a population harbor about 80% of the parasites. These 20-30% of the animals with higher parasite burden are primarily responsible for contaminating the environment with infective larvae for all other animals (Kaplan, 2010, Fleming et al., 2006). Forty out of the 41 of the goats sampled in this survey were positive for at least one species of adult nematodes. Ninety-two percent and 90% of sampled goats were positive for *H. contortus* and *T. colubriformis*, respectively. The total adult nematode worm burden ranged from 0 to 50,640 adults.

Fecal egg counts are only relatively crude indicators of worm burdens. The number of eggs in the feces may not always correlate with the number of parasites present in the intestinal tract. Differences in fecundity may mask the number of nematodes and low EPG or negative counts occur due to a large number of immature or non-fecund worms (Merck Manual, McKenna and Watson, 1987, Hoste et al., 2001). The 39 fecal samples examined for this survey were all positive for strongyle eggs. The lowest strongyle count was 5 EPG, found in sample #7, obtained in the winter season. The highest strongyle count was 16,650 EPG, found in sample #25, obtained in the spring season. A large EPG range for each season of this survey was observed. Overall, the summer season observed the lowest EPG range while the spring season had the highest EPG values. All samples with high strongyle EPG were correlated either a

higher number of *H. contortus* or *T. colubriformis* adults in the intestinal tract. As demonstrated in figures 4 and 5, the calculated EPG of *H. contortus* and *T. colubriformis* both show positive linear correlation to the species specific adults obtained from the goat samples during the survey.

In the southern USA, the inhibited state of parasitic nematodes occurs during the heat of the summer and the cold of the winter, and is dependent upon the nematode species. Hypobiosis results in an extended time for an immature nematode to develop into an adult. During hypobiosis, few eggs are deposited into the environment. In this survey, the summer months of July through October showed the lowest adult worm burdens, and coincide with decreased egg counts and pasture infectivity.

More adult nematodes were found in the winter samples, as shown by the highest adult nematode worm burdens in samples from November through February. A more accurate picture of nematode species seasonal prevalence would have been possible if a consistent and representative number of intestinal tracts were inspected for each season. For example, the summer season for this study contained only a few samples and those samples were all collected during the month of October. In addition to more samples, information on each animal in the study would allow for a better understanding of the various factors that dictate parasitisms. Information such as age, exact farm location, herd size, grazing method, worming schedule and healthcare history.

5.0 Conclusion

Gastrointestinal nematode infections of grazing livestock are almost always a mixture of species and within each species there is a mixture of developmental stages. Each species of nematode confers deleterious effects and collectively lead to illness or decreased performance in the host animal (Waller, 2006). Effects of parasitisms are determined by the interactions between the type of parasites present in the geographical area, parasitic life cycles, the

environment (including weather patterns), type of farm management, and a number of host factors. According to Craig (1986), parasites cannot be eradicated but they can be limited in their ability to cause economic loss to the producer. In order to achieve this goal there must be a combination of proper treatment and strategic management. A major factor that contributes to the fact that goats are more susceptible to gastrointestinal nematodes is that the goat's immunity to the nematodes is slow to develop and incomplete, even in mature goats (Kaplan, 2010).

The main challenge associated with limiting the gastrointestinal nematodes is the fact that *H. contortus* and *T. colubriformis* have developed a high degree of anthelmintic resistance. To exacerbate the situation, goats metabolize anthelmintic drugs much more rapidly than do other livestock and require a higher dosage to receive "effective" chemical intervention (Kaplan, 2010). Depending on the anthelmintic used, goats need 1.5 -2 times the dose recommended for sheep (SARE, Zajac et al., 2000; Mckenna and Watson, 1987; Varady et al., 2011). As demonstrated in multiple studies, resistance to drugs can develop due to overuse and improper dosing (e.g. giving goats the doses specific to sheep). Anthelmintics should only be used in goats that actually need treatment. Untreated animals will "supply" unselected worms that will stay more vulnerable to anthelmintics, prolonging the anthelmintics effectiveness.

Managing a goat herd to minimize the loss associated with gastrointestinal nematode infections starts with selecting a good breed of goat that is acclimated or native to the farms' climate. It is highly important to know which parasites are in the goat herd through larval identification. By performing fecal egg count reduction tests (FECRT) it is possible to determine which anthelmintics, if any, are effective against those species of parasites in the goat herd. All farms should practice smart drenching, wherein treatment is confirmed only to those animals that are shown to need it. That can be shown through a combination of fecal samples, FAMACHA scores and body condition scores.

In order to maintain or work towards a resilient and resistant goat herd it is highly important to review and improve your herd. Cull those animal in poor condition or those that have to be treated with anthelmintics multiple times. Pasture management can also help with decreasing the infective larvae available for consumption by the goats. Implementing rotational grazing, having access to browse, resting the pastures, not allowing goats to graze forage shorter than 6 inches, multispecies grazing, etc. Internal parasites continue to be a major concern for small ruminant producers. Historically, producers were able to use anthelmintics to manage the intestinal nematodes in their herds and flocks. However, the constant use of anthelmintics is now known to be unsustainable and the cause of the high levels of anthelmintic resistance in the gastrointestinal nematodes.

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7.0 Appendices

Figure 6a: Adult male *Teladorsagia circumcinta* (100X magnification)

Figure 6b: Adult female *Teladorsagia circumcinta* (40X magnification)

Figure 7: Adult male *Ostertagia trifurcata.* (200X magnification)

Figure 8a: Adult male *Haemonchus contortus* (100X magnification)

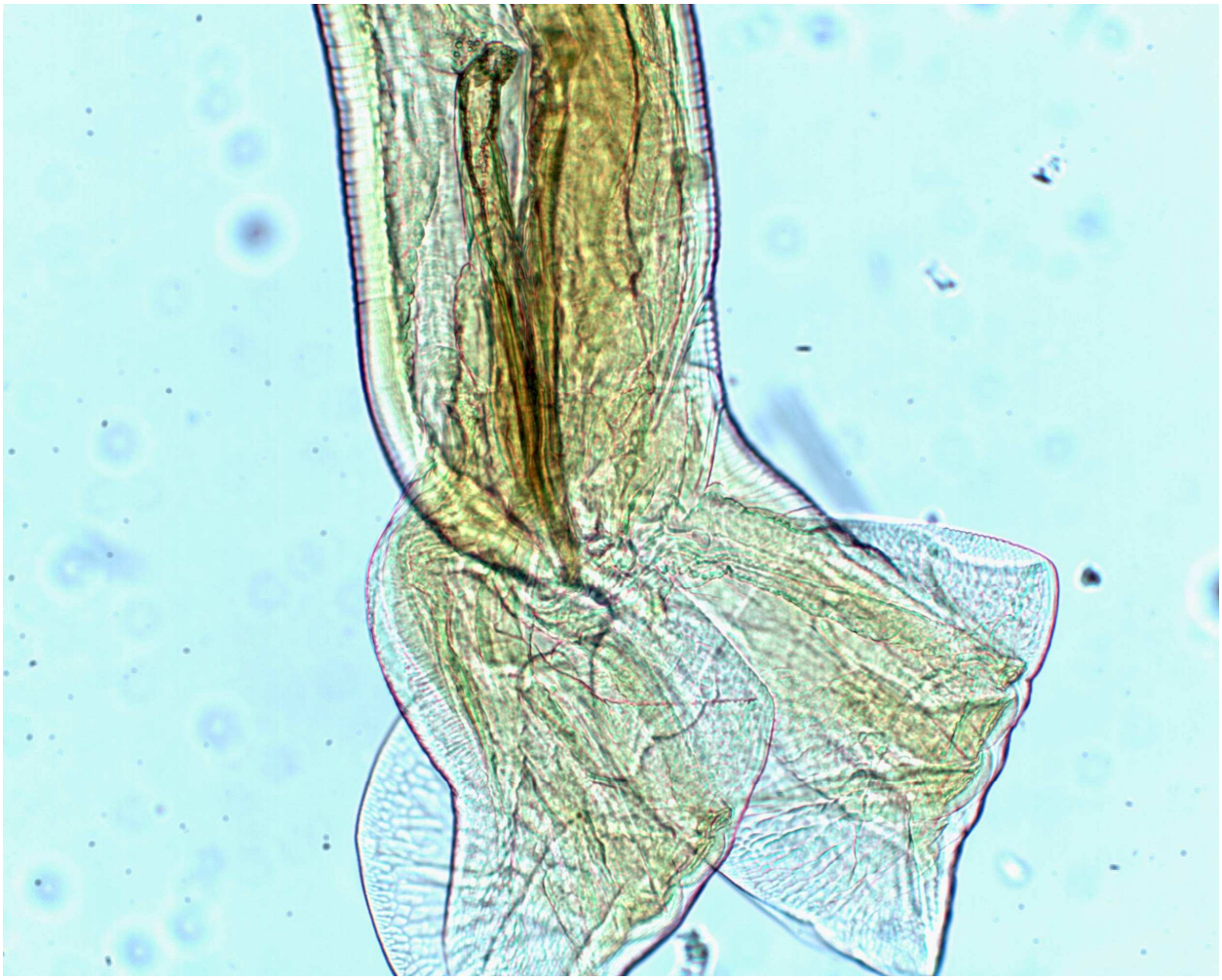

Figure 8b: Adult female *Haemonchus contortus* (40X magnification)

Figure 9a: Adult male *Trichostrongylus colubriformis* (100X magnification)

Figure 9b: Adult female *Trichostrongylus colubriformis* (100X magnification)

Figure 10: Adult male *Cooperia curticei*.(100X magnification)

Figure 11a: Adult male *Nematodirus spathiger* (100X magnification)

Figure 12: Adult female *Strongyloides papillosus* (40X magnification)

Figure 13: Adult *Oesophogostomum spp.* head (100X magnification)

Figure 14: Small ruminant eggs (200X magnification)

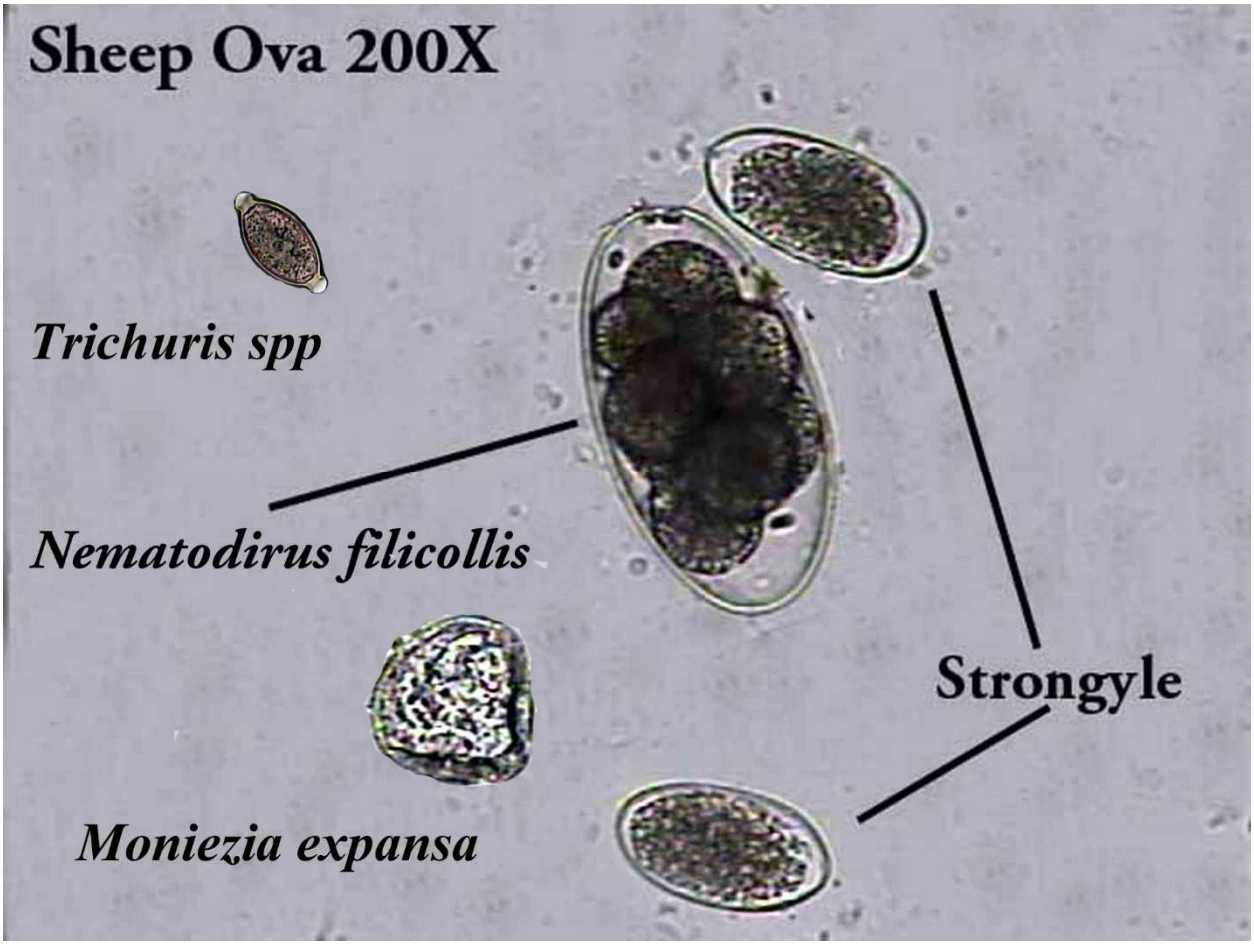

Figure 15: Small ruminant infective larvae (100X magnification)