Prevalence of Gastrointestinal Parasitic Infections of Sheep and Goats in Bui and Donga-Mantung Divisions of the North West Region of Cameroon

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Abstract

Background: This study was carried out to determine the prevalence, intensity of infection and management systems associated with gastrointestinal parasite infections in sheep and goats from 12 villages in the Bui and Donga-Mantung Divisions.

Methods: A total of 704 animals consisting of 342 males and 362 females were examined for gastrointestinal parasites, out of which 383 were goats and 321 sheep aged 5 months to 7 years. Also, 463 of these animals were adult ruminants and 241 young (kids/rams). Qualitative and quantitative analysis of stool samples were carried out using the McMaster technique.

Results: The 704 stool samples examined, 630 samples were found positive with one or more gastrointestinal parasites giving an overall prevalence of 89.5%. Sheep recorded the highest prevalence (90.0%). Haemonchus species recorded the highest prevalence (18.7%) in sheep, followed by Trichostrongylus species with a prevalence of (13.7%) still higher in sheep; Strongyloides species showed a prevalence of 10.4% in goats, and Trichuris species showed the least prevalence (2.4%) in sheep. The mean intensity for Haemonchus species was higher in all the animal groups, 694.4 ±1904.2 in goats but 189.5±137.3 in sheep. Mixed infections of Haemonchus species and Eimeria species were most prevalent in sheep (19.9%). The prevalence of Trichostrongylus species, Strongyloides species and Eimeria species were significantly low in all the two animal groups in the study area. Adults were more infected compared to young stock animals (lambs and kids). Concerning the various management techniques, prevalence of gastrointestinal tract parasites was higher in free range grazing animals (95.5%), followed by tethered animals (84.5%). Animals confined in paddocks had a lower prevalence (76.8%).

Conclusion: This study provides an important step in minimizing economic losses recorded in sheep and goats by providing information that will help farmers in these areas to practice the right traditional management techniques and strategic deworming methods, providing information on some medicinal plants that can be used to reduce the infection rate of these parasites on the ruminants.

Keywords: Gastrointestinal Parasites • Ruminants • Prevalence • Bui • Donga-Mantung • Cameroon

Introduction

Sheep and goat farming contribute an important role in the enhancement of the Cameroonian economy through production of meat, milk, skin, hair and manure. The production performance of these animals depends on good health and better management. The production of these animals can be affected by poor managerial practices, and diseases caused by various pathogens such as; bacteria, viruses, fungi, parasites etc. In Africa and Cameroon in particular, parasitic infection is one of the most common problems of sheep and goats. In animals, parasitic diseases comprise of helminths, arthropods and protozoa but gastrointestinal parasites are most common and widely distributed helminth parasites. Gastrointestinal nematodrosis is the most common infection in these animals caused by a variety of nematodes such as Haemonchus, Ostertagia, Trichostrongylus, Strongyloides, Cooperia, Nematodirus species result into gastroenteritis in Sub Sahara Africa and worldwide [1]. Among these parasites, Haemonchus contortus is a predominant and highly pathogenic gastrointestinal nematode responsible for decreased productivity in small ruminants throughout the world [2].

Despite the relative importance of nematode parasites in ruminants worldwide, other gastrointestinal parasites like the trematodes, cestodes and coccidians have also shown higher prevalence rates in most countries of the world. The trematodes of traditional veterinary and medical significance are almost all digenetic flukes that require a mollusc or snail as the first intermediate host. Prevalence studies reveal that Fasciola species are by far the most economically important trematodes of ruminants in the tropics [3]. The occurrence of flooding, water pans and swamps are important habitats for propagation of snail intermediate hosts of these flukes. Some of the regions in Cameroon have their borders to the west of the Atlantic

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Received date: 04 January, 2021; Accepted date: 24 January, 2021; Published date: 24 March, 2021
Ocean which makes them swampy, thus suitable for survival of snail intermediate hosts. Prevalence of parasitic infections is most commonly found in tropical and sub-tropical regions where environmental factors like temperature, humidity and rainfall are available necessary for propagation of pre-parasitic stages. The prevalence of gastrointestinal infections greatly varies from region to region due to diversity in agro-climatic conditions. Several reports have been documented on prevalence of gastrointestinal nematodosis and other parasites from different parts of the world [4].

Ruminants are the most widespread livestock in Cameroon and are reared in traditional systems. Cattle, goat and sheep rearing systems are either nomadic or pastoral, mixed farming or the peri-urban systems. Production and management systems vary from free range in less populated areas, to year-round confinement and cut-and-carry feeding in densely populated areas. Ruminants under extensive systems rely on natural grazing. As a result of shortage of water and forage, malnutrition is often the major limiting factor for profitable production of ruminants particularly during the dry season. Grazers of the Fulani tribe in the study area seek refuge during dry periods down the valleys and back to the mountains during the rainy season. Animals suffer from stress and disease effects especially during the periods of transhumance...

In most semi-arid and arid regions of sub-Saharan Africa, ruminants play a vital role in rural economies through provision of meat, milk, household income, manure and skin [5]. In most cases, the animals are run in large flocks or herds, concentrated in confined areas or tethered on pegs where they are likely to pick up infective larvae or oocysts from contaminated pastures [1] these poor management systems have contributed immensely to economic losses in ruminant production in sub-Saharan countries of the world [5] and Cameroon in particular. As a result, most livestock farmers pay keen attention to parasites that may likely cause death of their animals.

Control of gastrointestinal diseases in sheep and goats is a big challenge for veterinary parasitologists due to emergence of drug resistance and lack of other effective control methods. The rapid development of resistance to commonly used drugs against these infections, associated with high cost, environmental pollution and food residues have given new interest in medicinal plants or use as an alternative source of anthelmintic drugs. Therefore, there is urgent need for alternate methods of control to reduce worm burden, which should be less toxic, cheaper, with a wide margin of safety, availability and eco-friendly. Variations in the distribution of gastrointestinal parasites depend on temperature, humidity, rainfall, texture of soil, types of vegetation, age, sex and species of animals. Therefore, in order to formulate an effective control strategy against gastrointestinal diseases, it is necessary to obtain epidemiological data of the parasite in a particular area.

Materials and Methods

Area of study

The study was conducted at Holeta Municipal abattoir which is found in Holeta town. The study was carried out from November 2011 to March 2012. Holeta is located at Finfine Surrounding Oromia special Zone, Welmera districts of Oromia. The town is located at about 44 KM west of Addis Ababa, the capital of Ethiopia, geographically located at latitude of 09° 03’ 00”N and longitude of 38° 30’ 00”E. The altitude of the area is 2391 m.a.s.l and the annual mean temperature ranges between 11°C –22°C. Walmara Ocean which makes them swampy, thus suitable for survival of snail intermediate hosts. Prevalence of parasitic infections is most commonly found in tropical and sub-tropical regions where environmental factors like temperature, humidity and rainfall are available necessary for propagation of pre-parasitic stages. The prevalence of gastrointestinal infections greatly varies from region to region due to diversity in agro-climatic conditions. Several reports have been documented on prevalence of gastrointestinal nematodosis and other parasites from different parts of the world [4].

The study was conducted at Holeta Municipal abattoir from different areas surrounding the town [7]. The study was conducted in Bui and Donga-Mantung Divisions with Kumbo and Nkambe as headquarters respectively in the North West Region of Cameroon. The climate of this region is characterized by a long rainy season ranging from Mid-March to October, while annual average rainfall ranges from 1,500 mm to 2,000 mm and an altitude of about 1,100 m above sea level [6]. The dry season stretches from November to Mid-March, with monthly average temperatures in June reaching a maximum of about 21°C. These two Divisions are typical mountainous areas covered with grassy hills and valleys which constitute the major natural resource that the ruminant population (of livestock) depends on. The main form of agriculture practiced here is mixed crop/livestock production system. Most families are also involved in livestock farming, especially rearing sheep and goats. Flock sizes under the tethering system in these Divisions are in the order of 1 to 10 goats or sheep per household. Also, livestock farmers with large animal sizes like cattle are mostly owned by the Fulani tribes and they form separate communities in the upland grazing zones of Kumbo and Nkambe.

Selection of Study sites and farms

The study sites (Figure 1) below were selected on the basis of having a higher concentration of livestock. The sites included the following twelve villages: Tobin, Kikaikom, Mbah, Nkar, Shukai, Oku, Jakiri, Shisong, Ndu, Nkambe centre, Tabiken and Born.

![Figure 1: Age related prevalence of gastrointestinal parasites in sheep and goats](image)

Criteria for choosing study subjects

A total of 704 animals consisting of 342 males and 362 females were examined for gastrointestinal parasites. Of this, 338 were goats and 321 sheep. Also, 463 of these animals were adult ruminants while 241 were kids/lambs. For animals to be qualified as subjects, the sheep and goats must have been living in the study area and or its environs for the past three months. Samples were collected from ruminants of both sexes. Ages of the animals were determined from interviews with the farmers. Animals with ages ranging from 0-11 months were classified as young stock (lambs for sheep, kids for goats) while those from 12 months and above were categorized as adults. A larger number of goats were sampled because of their predominance in the area. Animals were sampled from 8 villages in the Bui Division and 4 villages in the Donga Mantung Division. Examination of animals was done by close inspection after proper restraining and by taking faecal samples to the laboratory for further processing.

Study design

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A preliminary survey was carried out prior to sample collection to sensitize interested farmers on the objectives of the study. Questionnaires were administered to farmers whose animals were selected for examination. It included information on the age/sex/breed of the animal, farm management practices and health conditions of the animals.

Collection of faecal samples and microscopic examination

During sample collection, bio data of the sheep and goats as to their age, sex, village and type of husbandry management system were recorded and labels put on plastic containers. A total of seven hundred and four (704) faecal samples from 321 sheep and 383 goats in twelve villages of the two Divisions were collected and taken to the Laboratory for analysis and confirmation of eggs/oocysts of gastrointestinal parasitic infections.

About 15 warm, moist, soft faecal pellets were taken directly from the rectum of the animal with the finger using gloves and placed in sealable plastic containers. Few minutes after collection, the samples were placed in a cooler and then transported to the Laboratory of the National Centre for Animal Husbandry and Veterinary Training School Jakiri, Kumbo, Bui Division, and stored in a refrigerator for analysis. All along, samples were kept cool until examination, in order to prevent trichostrongylid larvae from hatching.

The sedimentation technique as described by [7] was used to detect presence of helminth eggs and coccidian oocysts. The eggs per gram (EPG) of faeces were quantitatively analyzed to determine intensity of infection using the McMaster technique.

Statistical analysis

Data were stored in a Microsoft Excel spread sheet cleaned by checking for errors or missing variables and then exported to SPSS (Statistical Package for Social Science, Version 20) Software for analysis. Summary statistics were generated using the same software. For the purpose of modeling these data, explanatory variables were first explored for associations between parasites using Chi-square (χ2) test. The prevalence of helminth parasites was compared between demographic parameters using the chi square test. The Chi square test was equally used to examine the effects of the various risk factors. The non-parametric test of Kruskal Wallis was used to compare mean intensity between age group and locality, intensity with animal gender, breeding system and state of health. Egg per gram was notably high for Haemonchus spp., Trichostrongylus spp., Oesophagostomum spp and Eimeria spp. were the most prevalent parasites recorded in all the two groups of animals, with a significant difference. From the results obtained, of the eight nematodes identified, goats recorded the highest prevalence with Haemonchus spp. being the most prevalent (18.0%) while sheep showed the least prevalence of 2.4% in Trichuris spp. with no significant difference (P>0.05). Of the single trematode (Fasciola spp) identified, the highest prevalence was in goats (7.0%) while in sheep, the highest prevalence was 6.2% with no significant difference. The Eimeria spp identified was more prevalent in sheep (13.4%) than in goats (7.8%) with a significant difference (P<0.05). Concerning the intensity of infection of the nematode species, the study revealed that mean egg per gram was notably high for Haemonchus spp., Trichostrongylus spp., Strongyloides spp. and Eimeria spp. in sheep and goats. However, faecal egg counts revealed overall low mean egg per gram of faeces for the other parasites identified as seen in the table below.

Prevalence and Intensity of gastrointestinal parasites in sheep and goats

The Gastrointestinal parasites identified from faecal samples were nematodes, trematodes and protozoans. These parasites, their prevalence and intensity are shown on Table 1. Haemonchus spp., Trichostrongylus spp., Strongyloides spp., Oesophagostomum spp and Eimeria spp. were the most prevalent parasites recorded in all the two groups of animals, with a significant difference. The Eimeria spp identified was more prevalent in sheep (13.4%) than in goats (7.8%) with a significant difference (P<0.05). Concerning the intensity of infection of the nematode species, the study revealed that mean egg per gram was notably high for Haemonchus spp., Trichostrongylus spp., Strongyloides spp. and Eimeria spp. in sheep and goats. However, faecal egg counts revealed overall low mean egg per gram of faeces for the other parasites identified as seen in the table below.

Prevalence of mixed infections in sheep and goats

Most of the animals had mix infections, with most of the combinations being Haemonchus spp/Eimeria spp. and a highest prevalence was observed in sheep as seen on Table 2 below.

Results

Overall prevalence of gastrointestinal parasites in sheep and goats in the study area

<table>
<thead>
<tr>
<th>Mixed Infections</th>
<th>Goats (N=383)</th>
<th>Sheep (N=321)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No Infected (%)</td>
<td>No Infected (%)</td>
</tr>
<tr>
<td>Trichostrongylus spp/Haemonchus spp</td>
<td>18 (4.7%)</td>
<td>47 (14.6%)</td>
</tr>
<tr>
<td>Trichostrongylus spp/Strongyloides spp</td>
<td>13 (3.4%)</td>
<td>28 (8.7%)</td>
</tr>
<tr>
<td>Trichostrongylus spp/ Haemonchus spp/Eimeria spp</td>
<td>21 (5.5%)</td>
<td>14 (4.4%)</td>
</tr>
<tr>
<td>Trichostrongylus spp/Strongyloides spp/Eimeria spp</td>
<td>8(2.1%)</td>
<td>5 (1.5%)</td>
</tr>
<tr>
<td>Haemonchus spp/Eimeria spp</td>
<td>26 (6.8%)</td>
<td>64 (19.9%)</td>
</tr>
<tr>
<td>Trichostrongylus spp/Eimeria spp</td>
<td>12 (3.1%)</td>
<td>27 (8.4%)</td>
</tr>
<tr>
<td>Strongyloides spp/Eimeria spp</td>
<td>19(5.0%)</td>
<td>32 (9.96%)</td>
</tr>
</tbody>
</table>
Age related prevalence of gastrointestinal parasites in sheep and goats in the study area

A total of 463 adult ruminants and 241 young kids/lambs were examined during the study. Out of these, prevalence of 94.3% infection was recorded in adult followed by 87.7% in adult goats and the least prevalence was observed in young sheep (lambs) (82.0%). However, the overall prevalence of gastrointestinal parasites among the different age groups showed that generally, adult sheep and goats were the most infected (91.8%) while in young goats (kids) and young sheep (lambs), an overall prevalence of 85.5% was recorded (Figure 3). However, the Chi square value revealed no significant difference among the different age groups (P>0.05).

Table 2. Sex related prevalence of gastrointestinal parasites in sheep and goats

<table>
<thead>
<tr>
<th>sex</th>
<th>Ruminants</th>
<th>No Examined</th>
<th>No Infected</th>
<th>Prevalence (%)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Females</td>
<td>Sheep</td>
<td>168</td>
<td>146</td>
<td>86.90%</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>194</td>
<td>169</td>
<td>87.10%</td>
<td>--</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>362</td>
<td>315</td>
<td>87.00%</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Males</td>
<td>Sheep</td>
<td>153</td>
<td>143</td>
<td>93.50%</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td>Goats</td>
<td>189</td>
<td>172</td>
<td>91.07%</td>
<td>--</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>342</td>
<td>315</td>
<td>92.10%</td>
<td>&gt;0.05</td>
</tr>
</tbody>
</table>

Prevalence of gastrointestinal parasites in sheep and goats raised under traditional management systems

The present study also revealed details on the prevalence of gastro intestinal tract (GIT) parasites in animals kept under different traditional management systems. We noted that, animals confined in paddocks recorded low infection rates of 76.8% compared to free range grazers with highest overall prevalence (95.5%) and tethered animals (84.5%). Sheep and goats that were grazed in confined paddocks had prevalence rates of 97.7% and 51.8%, respectively, with sheep recording the highest prevalence.

For both sheep and goats, tethered animals had highest infection rates of 100% and 90.8%, respectively with all tethered sheep infected. Free range grazers had prevalence rates of 97.2% and 75.0% for sheep and goats, respectively with sheep still recording the highest prevalence. A significant difference in prevalence was observed in both sheep and goats practicing the different grazing systems and also in confined animals (Tables 4-6).

Table 4. Prevalence of gastrointestinal parasites in free range grazing ruminants

<table>
<thead>
<tr>
<th>Ruminants</th>
<th>No Examined</th>
<th>No Infected</th>
<th>Prevalence (%)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sheep</td>
<td>36</td>
<td>35</td>
<td>97.20%</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Goats</td>
<td>48</td>
<td>36</td>
<td>75.00%</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Total</td>
<td>84</td>
<td>71</td>
<td>84.50%</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

The present study revealed an overall prevalence of gastrointestinal tract parasites in ruminants to be 89.5% with 90.0% and 89.0% in sheep and goats, respectively. These results are in consonance with the findings of in Ethiopia and in Nigeria. The high prevalence of gastrointestinal parasites in small ruminants as a whole agrees with earlier reports [8-11]. The higher prevalence in sheep and goats in the study area might be due to poor management systems. In the North West Region of Cameroon, most especially in the Bui and Donga Mantung Divisions where this study was conducted, mixed crop livestock production predominates where few numbers of small ruminants are kept together. Majority of the sheep and goats are tethered on farm lands. As a result of this, most of the animals are re-infected due to pasture contamination as they graze within a confined region for several months.

The higher prevalence of gastrointestinal tract parasites in sheep compared to goats is in agreement with the report of [1] whose assertions explained that the grazing habits of sheep (grazing closer to the earth soil) warrants these animal species to be more infected than goats. However, in the present survey, the difference in philosophy with the previous findings may be because the majority of the goats were kept under poor veterinary infrastructure and medication. More importantly, this may be due to slow development of immunity in goats to GIT parasites as compared to sheep [12].

The most prevalent gastrointestinal tract parasites were the Strongyles, Strongyloides and Eimeria oocysts. These results corroborate several findings in Africa including those of [13]. Similarly, high levels of these infections have also been reported in India, Rwanda, and Pakistan [14]. Strongyle nematodes were of the genera Trichostrongylus, Haemonchus, Oesophagostomum, and Nematodirus. The climatic conditions of the study area (warm moist) are highly suitable for survival of strongyles and transmission of the parasites. Another contributing factor towards the high prevalence of strongyle nematodes may be due to poor farm management techniques including construction of housing, feeding, watering systems and generally poor hygienic conditions of the farms.

However, it is important to remember that in many regions of Africa, endemic haemonchosis overlaps with other important parasitic causes of anaemia in small ruminants, notably fascioliasis and trypanosomiasis. The prevalence of Trichuris spp and Fasciola spp. in the study area for
the two animal groups was extremely low. A low prevalence for Fasciola spp. may be due to the vegetation cover characteristic of the two Divisions. The typical mountainous area covered with grass on the hills does not favour propagation of snail intermediate hosts. It is probable that, the few ruminants infected with Fasciola spp might have gotten their infection during transhumance in the neighboring villages having valleys, a period during which there is scarcity of pasture and water in most mountainous villages in the study area [15].

In the present study, sheep and goats were raised under an extensive management system, in mostly low-lying pastures where stocking densities were high, nutrition was limited, and veterinary care was almost nonexistent. One study demonstrated a positive correlation between nematode infection and land elevation [16]. These factors promote the exposure of livestock to infective larvae from pastures and the establishment of infections in animals [17]. Upland grazing areas recorded the highest prevalence of gastrointestinal parasites in sheep as compared to lowland grazing areas which had higher prevalence of gastrointestinal tract parasites in goat ruminants. Upland grazing areas are occupied mostly by the Fulani tribes while the vast lowland grazing areas are occupied mainly by the indigenes of Bui and Donga Mantung Divisions for crop farming. The Fulani tribes form the minority group and often are faced with a problem of limited grazing land. They often pitch their tents and small huts closer to their cattle herds on mountainous grazing areas for proper supervision of their animals. Most of them rear cattle and few sheep inherited from their parents. They do not keep goats since they attach more religious significance to sheep during Ramadan festivities. This therefore implied that the low prevalence of GIT parasites in small ruminants in the upland grazing community was not due to absence of parasites on contaminated pastures but rather might have been due to a relatively small sample size of small ruminants kept by the exclusive Fulani community.

Though infection rates were higher in traditionally managed animals (tethered ruminants), a study carried out in Mankon in the North West Region of Cameroon [18] showed that mortality rates were relatively low for all animals reared under the traditional management systems. The reason behind this could be that, local breeds of small ruminants in Mankon Bamenda (Cameroon Dwarf goats and Dwarf Forest sheep) have acquired strong immunity to infection with GIT parasites due to recurrent infections.

Generally, adult animals had a slightly higher prevalence rate of gastrointestinal tract parasites compared to the young animals. These results contradict the findings of [19,20] whose results revealed that, lambs and kids were the most infected because they are more susceptible to infection than adults due to low levels of immunity. A slightly higher prevalence in young stock may be due to failure in separating young stock from the adults at pre weaning age due to lack of knowledge to do so, overgrazing of infested pastures coupled with inappropriate and inadequate use of anthelmintics [21]. However, the higher prevalence in adult animals compared to younger ones might have been due to the use of anthelmintic treatment of individual young animals by some herdsmen, this may well have reduced the magnitude of group mean egg counts for the young male and female groups before the study.

Sex related prevalence and intensity revealed that male animals were the most infected (92.1%) compared to the female animals (87.0%) though there was no significant difference. However, from this study, higher prevalence (100%) of parasitic infection was not also associated with sex (p>0.05). Though not statistically significant, single parasites actually recorded a higher prevalence of gastrointestinal parasitic infection in females than males. This high rate of infection in females could be due to the fact that female ruminants appear to be more susceptible to infections than males. This finding is consistent with other reports, and it was not surprising because naive females frequently graze the same areas, hence resulting to high infection rate and intensity. The intensity of infection however is reportedly related to the level of hygiene [22]. This finding is contrary to the work of other authors who reported a low prevalence in females compared to males [23].

Locations in the study area where farmers practiced traditional management systems showed higher prevalence rates and intensities of infection compared to areas managed by the government parastatal called “Societe de Developpement des Petit Ruminants” (SODEPA) under the semi intensive management system [18]. The low prevalence rate in SODEPA in Bui Division could be explained by the fact that the parastatal has a curved out vast grazing land reserved only for ruminants of the parastatal. Animals kept by SODEPA are well catered for, frequently drenched (dewormed), well fed with supplemental feed and constantly monitored for any irregularities that might lead to death of the animal [15].

Conclusion

This study was undertaken to determine the prevalence, intensity of infection and management systems associated with gastrointestinal parasitic infections in small ruminants (sheep and goats) from 12 villages in the Bui and Donga Mantung Divisions. It is worth noting that the prevalence of gastrointestinal parasitic infections evaluated for the concerned sample was relatively high (89.5%) with a high intensity of infection too. The study clearly indicates that control measures should make use of the variations in helminth prevalence and intensity among management systems, sex and age groups, to achieve rational use of anthelmintics. Also, tethered animals should not be allowed to graze on a particular spot continuously for several weeks. Grazing spots should be rotated to reduce the chances of ruminants being re-infected from contaminated pastures. Farmers should be educated on the importance of using dry season feed reserves as means of ensuring safe feed for zero-grazed ruminants. Finally, field veterinarians should assist livestock farmers in strategic deworming campaigns with effective anthelmintics and medicinal plants used at the beginning and after the end of the rainy season. This should also be followed by periodic inspections unannounced to these farmers and other farmers if possible. Periodic seminars or workshops should be organized as a means of creating awareness in these farmers.

Availability of Data and Materials

Data and material are available to other researchers upon request.

Competing Interest

The authors declared that they have no competing interest.

Funding

No funding.

Acknowledgements

The author sincerely grateful to research unit of applied biology and ecology.

Author’s Contributions

MEM, VKP, YC, NACN, ML and MR contributed to the design of the study, data collection, led the analysis and drafting of the manuscript. All authors read and approved the final manuscript.

Ethical Approval and Consents to Participate
All authors hereby declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised 1985) were followed, as well as specific national laws where applicable. All experiments have been examined and approved by the appropriate ethics committee.

Consent for Publication

Not applicable

References


How to cite this article: Mbong Erica Malla, Vincent Khan Payne , Yamssi Cedric, Noumedem Anangmo Christelle Nadia, Megwi Leonelle and Matsinkou Rosine/Prevalence of Gastrointestinal Parasitic Infectious of Sheep and Goats in Bui and Donga-Mantung Divisions of the North West Region of Cameroon J Vet Sci Technol 12(2021)