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6 **COPROLOGICAL ASSESSMENT OF GASTROINTESTINAL PARASITE INFECTIONS IN CATTLE**
7 **AND SHEEP UNDER SMALLHOLDER FARMING SYSTEMS IN BOUKOMBÉ MUNICIPALITY,**
8 **NORTH-WESTERN BENIN**

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10
11 ***Abstract***

12 Gastrointestinal parasitism remains a major constraint to ruminant productivity in tropical smallholder farming
13 systems, largely due to inadequate diagnosis and control strategies. This study aimed to assess the prevalence and
14 intensity of gastrointestinal parasite infections in cattle and sheep raised under extensive and semi-extensive systems
15 in Boukombé municipality, north-western Benin, using the Mini-FLOTAC technique. Fresh fecal samples (20–50 g)
16 were collected rectally from randomly selected Djallonké sheep and Somba cattle across five localities and analyzed
17 using Mini-FLOTAC with a detection limit of 5 eggs per gram (EPG). Parasite prevalence was calculated, and
18 infection intensity was expressed as mean EPG \pm SEM. The effects of locality and sex on parasite burdens were
19 assessed using two-way ANOVA. Gastrointestinal strongyles and *Strongyloides* spp. were detected in all localities
20 and in both host species, indicating widespread endemicity. In sheep, the highest parasite burdens were recorded in
21 Boukombé Centre, with mean EPG values of 494.0 ± 111.65 for *Strongyloides* spp. and 168.0 ± 50.34 for
22 strongyles. In cattle, strongyle egg counts were highest in Manta (391.25 ± 59.85 EPG) and Boukombé Centre
23 (312.19 ± 33.83 EPG). *Moniezia* spp. and

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26 coccidia showed a restricted and locality-dependent distribution, while *Ne*
27 *matodirus* spp. and *Trichuris* spp. were absent. Parasite burdens differed significantly among localities ($p < 0.05$),
28 whereas sex had no significant effect. These findings highlight active and continuous transmission of gastrointestinal
29 parasites in the study area and demonstrate the high sensitivity and operational suitability of Mini-FLOTAC for
30 epidemiological surveillance under tropical field conditions. The study provides a robust scientific basis for the
31 implementation of locally adapted and evidence-based parasite control strategies to improve ruminant health and
32 productivity in northern Benin.

1. Introduction

Animal production systems in Benin are still largely dominated by traditional livestock management practices, particularly for cattle, sheep, goats, pigs, and poultry. In 2012, livestock populations were estimated at approximately 2,111,000 cattle, 842,000 sheep, 1,678,000 goats, 398,000 pigs, and 16,941,000 poultry (PAFILAV, 2014). Despite these numbers, domestic livestock production remains insufficient to fully meet national demand for animal-derived proteins, including meat, milk, and eggs (PAFILAV, 2014). Consequently, Benin remains highly dependent on imports of meat and animal products.

This reliance on imports results in substantial losses of foreign exchange and hampers the development of emerging and promising livestock value chains. Improving national livestock productivity is therefore a major priority to enhance food security and satisfy the growing protein needs of the population. Among the numerous constraints limiting optimal productivity and profitability of livestock farms, animal health issues, particularly those caused by gastrointestinal parasites, represent a major challenge (Koussihouèdè, 2016). Gastrointestinal parasitism negatively affects animal health and performance by reducing feed intake, growth rates, milk production, and reproductive efficiency, and may ultimately lead to increased mortality. Consequently, effective control of parasitic diseases is considered a key component of herd health management (Cabaret, 2004).

Successful control of gastrointestinal parasitoses relies heavily on early, accurate, and reliable diagnosis. Coproscopy is a rapid and practical diagnostic approach that encompasses a wide range of techniques used to detect and quantify parasite eggs and oocysts in fecal samples. Among these, the McMaster technique has long been the most widely used method for coprological diagnosis and fecal egg counting in veterinary parasitology (Roeber et al., 2013). This quantitative and qualitative technique is based on the flotation principle and allows estimation of parasite burden through egg counts. However, its analytical sensitivity depends on fecal sample size and the volume of flotation solution used, with common detection limits ranging from 5 to 100 eggs per gram of feces (EPG).

To overcome these limitations and achieve lower detection thresholds, several alternative techniques have been developed, including the modified Wisconsin technique, the Stoll egg-counting method, and the FECPAK system (Noel et al., 2016). Nevertheless, many of these methods require centrifugation to improve egg recovery, which significantly increases processing time and limits their routine application under field and laboratory conditions. Importantly, the reliability of fecal egg count results is strongly influenced by the analytical variability and precision of the diagnostic method used (Noel et al., 2016). Previous studies have demonstrated substantial differences in precision among coprological techniques, highlighting the need for more sensitive and reproducible methods.

In this context, the Mini-FLOTAC technique was introduced as an alternative to the conventional McMaster method. Mini-FLOTAC is a quantitative coprological technique based on a flotation system using two large counting chambers of 1 mL each, allowing improved sensitivity and accuracy without the need for centrifugation (Rinaldi et al., 2014). This technique has been extensively evaluated and compared with the McMaster method, yielding promising results in fecal samples from ruminants, pigs, and poultry in Europe (Knopp et al., 2009; Duthaler et al., 2010; Cringoli et al., 2010; Rinaldi et al., 2010), Asia (India), Africa (Alowanou et al., 2021), and South America (Cringoli et al., 2010; Rinaldi et al., 2010).

Notably, the comparative study conducted by Alowanou et al. (2021) demonstrated that quantitative fecal analysis using the Mini-FLOTAC technique provides higher sensitivity and greater precision than the McMaster technique for parasite egg counts in sheep, goats, and rabbits. These findings support Mini-FLOTAC as a reliable and efficient alternative to the conventional McMaster method for the diagnosis of parasitic infections in livestock.

Against this background, the present study aimed to assess the prevalence and intensity of gastrointestinal nematode infections in cattle and sheep in Boukombé municipality using the Mini-FLOTAC technique, with the ultimate goal of generating evidence-based data to support the development of locally adapted parasite control and treatment schedules for ruminants in the study area.

2. Materials and Methods

2.1. Study area

The present study was conducted in Boukombé municipality, located in the Atacora Department in north-western Benin. The area is predominantly rural and is characterized by a rugged topography with hills and plateaus, as well as a Sudano-Guinean climate. The climatic pattern includes a rainy season extending from May to October and a dry season from November to April. The local economy is largely based on agriculture and livestock production, particularly cattle and sheep farming, which are mainly practiced under extensive and semi-extensive management systems. Livestock husbandry represents a major source of livelihood and income for local communities, predominantly belonging to the Otammari, Oubièro, and Dendi sociocultural groups. The agroecological conditions and livestock management practices in this area are likely to influence the occurrence, persistence, and transmission of gastrointestinal parasites in ruminants.

2.2. Study animals

The sheep and cattle breeds encountered at the different sampling sites were the Djallonké sheep and the Somba cattle, respectively. These breeds are commonly raised under traditional extensive and semi-extensive production systems in northern Benin.

2.3. Sample collection, preservation, and transport

Approximately 20–50 g of fresh feces were collected directly from the rectum of randomly selected sheep and cattle using disposable latex gloves in the targeted farms. Each fecal sample was placed in a labeled plastic bag and immediately stored in a cooler at approximately 4 °C. Samples were subsequently transported to the Veterinary and Zootechnical Assistance Center laboratory located in Bantè Centre, where coprological analyses were performed upon arrival.

2.4. Coprological analyses

Mini-FLOTAC technique

The Mini-FLOTAC technique is a quantitative coprological method developed by Cringoli et al. (2010) for the detection and enumeration of gastrointestinal parasite eggs and oocysts. The device consists of a reading disc with two 1-mL flotation chambers and allows the observation of

a thin, uniform layer under the microscope, thereby improving reading accuracy and operator comfort.

For this technique, two (2) grams of feces were weighed and homogenized with 18 mL of flotation solution (saturated sodium chloride, NaCl), resulting in a dilution ratio of 1:10. The suspension was prepared using the Fill-FLOTAC device, which combines a collector and a built-in filter, allowing simultaneous homogenization and filtration of the fecal material.

The fecal suspension was then used to fill the two counting chambers of the Mini-FLOTAC. Each chamber was filled slowly until a slight meniscus formed. The Mini-FLOTAC was maintained in a horizontal position for at least 10 minutes to allow parasite eggs and oocysts to float to the upper layer. After this flotation period, uniform pressure was applied to the Mini-FLOTAC key, and the reading disc was rotated by 90°, separating the upper reading surfaces from the chamber bases.

Microscopic examination was performed using a light microscope at 100× and, when necessary, 400× magnification. Prior to microscopic observation, a dedicated adapter was fitted to the Mini-FLOTAC device to facilitate stable positioning on the microscope stage.

Analytical sensitivity and egg count calculation

The analytical sensitivity of the Mini-FLOTAC technique under the applied dilution conditions was 5 eggs per gram of feces (EPG). Each egg or oocyst observed corresponded to 5 EPG. Samples in which no eggs were detected were considered to have parasite burdens below the detection limit (< 10 EPG).

The egg count was calculated as follows:

$$\text{EPG (Mini-FLOTAC)} = \text{Number of eggs counted} \times 5$$

2.5. Statistical analysis

All data were entered into Microsoft Excel and analyzed using R software (R Foundation for Statistical Computing, Vienna, Austria). For each parasite group, fecal egg counts (EPG) were summarized as mean \pm standard error of the mean (SEM) by locality and sex.

Prior to inferential analyses, EPG data were explored for normality and homogeneity of variances using visual inspection (histograms and Q–Q plots) and standard tests (Shapiro–Wilk and Levene’s tests). Because fecal egg count data are typically right-skewed, EPG values were log-transformed as $\log_{10}(EPG + 1)$ when necessary to meet model assumptions.

To evaluate the effects of locality and sex on parasite burdens, a two-way analysis of variance (two-way ANOVA) was performed for each parasite group, with *locality*, *sex*, and their interaction (*locality* \times *sex*) included as fixed effects. When the interaction term was not significant, it was removed and the model was refitted using main effects only. When a significant locality effect was detected, post-hoc multiple comparisons were conducted using Tukey’s HSD test.

For parasite prevalence (presence/absence), results were expressed as percentages by locality and species. Differences in prevalence between localities were assessed using the Chi-square test, or Fisher’s exact test when expected cell counts were <5 .

All statistical tests were two-tailed, and statistical significance was declared at $p < 0.05$.

3. Results

Inventory of gastrointestinal parasites in ruminants in Boukombé municipality Gastrointestinal parasites identified in sheep Table 1 presents the distribution of the main gastrointestinal parasites identified in sheep across the different sampling localities in Boukombé municipality. Gastrointestinal strongyles and *Strongyloides* spp. were detected in all surveyed localities, indicating their wide distribution within the study area. In contrast, coccidia, *Nematodirus* spp., and *Trichuris* spp. were not detected in any of the localities. *Moniezia* spp. were identified only in Boukombé Centre and Dipoli. These results highlight spatial variability in parasite composition, which may be associated with differences in environmental conditions, pasture management, and animal husbandry practices.

Table 1.

Gastrointestinal parasites identified in sheep in Boukombé municipality Localities

Localities	Strongyles	Coccidia	Strongyloides	Moniezia	Nematodirus	Trichuris
Boukombé Centre	+	—	+	+	—	—
Dipoli	+	—	+	+	—	—
Korontière	+	—	+	—	—	—
Natta	+	—	+	—	—	—

Presence (+) or absence (—) of each parasite group in the surveyed localities.

Gastrointestinal parasites identified in cattle Table 2 shows the gastrointestinal parasites identified in cattle across the five surveyed localities in Boukombé municipality. Strongyles and *Strongyloides* spp. were systematically detected in all localities, reflecting a generalized infestation of cattle by these nematodes. With the exception of Korontière, other parasites—namely coccidia, *Moniezia*, *Nematodirus*, and *Trichuris*—were absent in all sites. Korontière stood out by the simultaneous presence of four parasite groups, suggesting particularly favorable environmental or management conditions for parasite survival and transmission.

Table 2.

Gastrointestinal parasites identified in cattle in Boukombé municipality Localities

Localities	Strongyles	Coccidia	Strongyloides	Moniezia	Nematodirus	Trichuris
Boukombé Centre	+	—	+	—	—	—
Dipoli	+	—	+	—	—	—
Korontière	+	+	+	+	—	—

Localities	Strongyles	Coccidia	Strongyloides	Moniezia	Nematodirus	Trichuris
Natta	+	—	+	—	—	—
Manta	+	—	+	—	—	—

Presence (+) or absence (—) of each parasite group in the surveyed localities.

Parasite burden in ruminants in Boukombé municipality Parasite egg counts in sheep Table 3 presents the mean parasite burdens expressed as eggs per gram of feces (EPG \pm SEM) for the main gastrointestinal parasites identified in sheep across the surveyed localities. *Strongyloides* spp. and strongyles showed the highest parasite burdens, particularly in Boukombé Centre, with mean values of 494.0 ± 111.65 EPG and 168.0 ± 50.34 EPG, respectively. Two-way ANOVA revealed a significant effect of locality on *Strongyloides* spp. ($p = 0.0164$) and strongyle egg counts ($p = 0.0201$), while no significant effect of sex was observed. These findings confirm a marked spatial heterogeneity in gastrointestinal parasite burdens among sheep in Boukombé municipality.

Table 3.

Mean EPG \pm SEM of gastrointestinal parasites in sheep in Boukombé municipality

Localities	Strongyloides	Moniezia	Nematodirus	Trichuris	Strongyles	Coccidia
Boukombé	494.0	± 146.0	$\pm 0.0 \pm 0.0$	0.0 ± 0.0	168.0	$\pm 60.0 \pm 33.47$
Centre	111.65	91.74			50.34	
Dipoli	94.0 ± 43.43	76.0 ± 76.0	0.0 ± 0.0		0.0 ± 0.0	0.0 ± 0.0
Korontière	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0		28.0	$\pm 0.0 \pm 0.0$
					28.0	166.0
						± 136.81

Localities	Strongyloides	Moniezia	Nematodirus	Trichuris	Strongyles	Coccidia
Natta	44.0 ± 22.49	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0

Parasite egg counts in cattle Table 4 shows the mean parasite burdens (EPG ± SEM) of gastrointestinal parasites identified in cattle. Strongyles exhibited the highest egg counts, particularly in Manta (391.25 ± 59.85 EPG) and Boukombé Centre (312.19 ± 33.83 EPG). Strongyloides spp. were present at moderate levels, whereas Moniezia, Nematodirus, and Trichuris were almost absent across the study area. Two-way ANOVA revealed a highly significant effect of locality on strongyle egg counts ($p < 0.001$), with no significant effect of sex.

Table 4.

Mean EPG ± SEM of gastrointestinal parasites in cattle in Boukombé municipality

Localities	Strongyles	Coccidia	Strongyloides	Moniezia	Nematodirus	Trichuris
Boukombé Centre	312.19 33.83	± 0.0 ± 0.0	19.06 ± 4.27	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
Dipoli	36.25 ± 17.32	0.0 ± 0.0	3.75 ± 2.21	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
Korontière	125.62 35.01	± 4.38 3.76	± 25.0 ± 10.49	0.62 0.62	± 0.0 ± 0.0	0.0 ± 0.0

Localities	Strongyles	Coccidia	Strongyloides	Moniezia	Nematodirus	Trichuris
Manta	391.25	± 0.0 ± 0.0	21.88 ± 5.86	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	59.85					
Natta	169.38	± 0.0 ± 0.0	12.5 ± 4.79	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	51.98					

Figure 1 illustrates the spatial variation in the prevalence of major gastrointestinal parasites in sheep across the surveyed localities. *Strongyles* and *Strongyloides* spp. were the most prevalent parasites, occurring at high levels in all localities and reaching 100% prevalence in Boukombé Centre. Their widespread distribution indicates intense and continuous transmission within the study area. In contrast, *Moniezia* spp. showed a limited and locality-specific distribution, being detected only in BoukombeCentre (40%) and Dipoli (20%). *Nematodirus* spp. and *Trichuris* spp. were either absent or detected at very low prevalence, confirming their marginal contribution to the parasitic fauna of sheep in the study area. Overall, the dominance of *strongyles* and *Strongyloides* spp., coupled with the heterogeneous distribution of other parasite groups, suggests that environmental conditions and local husbandry practices strongly influence parasite transmission dynamics. These findings highlight the need for targeted, locality-specific parasite control strategies rather than uniform treatment approaches.

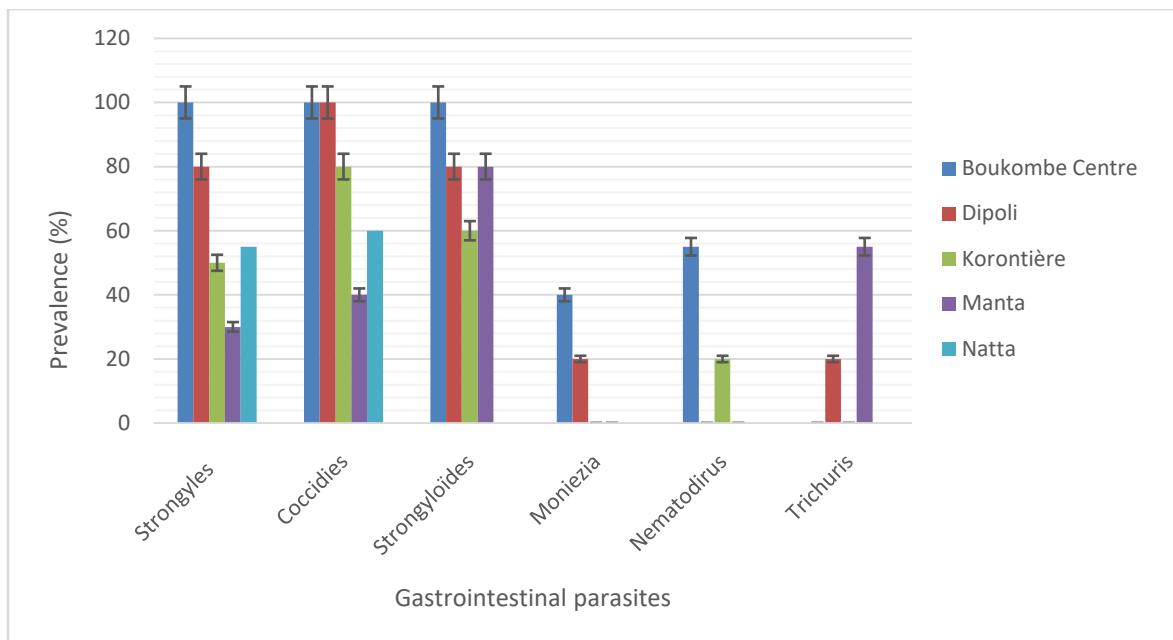


Figure 1. Prevalence (%) of gastrointestinal parasites in sheep in Boukombé municipality

Figure 2 highlights pronounced spatial differences in the prevalence of gastrointestinal parasites among cattle across the surveyed localities. *Strongyles* were highly prevalent in all areas, with particularly elevated prevalences in Manta and Natta ($\geq 90\%$), confirming their dominant role in bovine parasitism in the study area. *Strongyloides* spp. also showed a widespread distribution, although their prevalence varied markedly between localities, with lower values observed in Dipoli (approximately 20%) and Natta (around 45%). In contrast, coccidia and *Moniezia* spp. exhibited a restricted distribution, being detected only in Korontière, where they occurred at low prevalences. *Nematodirus* spp. and *Trichuris* spp. were not detected in cattle in any of the surveyed localities. Overall, these results indicate a heterogeneous spatial distribution of gastrointestinal parasites in cattle, likely driven by differences in herd management practices, grazing pressure, sanitary conditions, and local environmental factors. This heterogeneity underscores the importance of locality-specific parasite control strategies rather than uniform interventions across the study area.

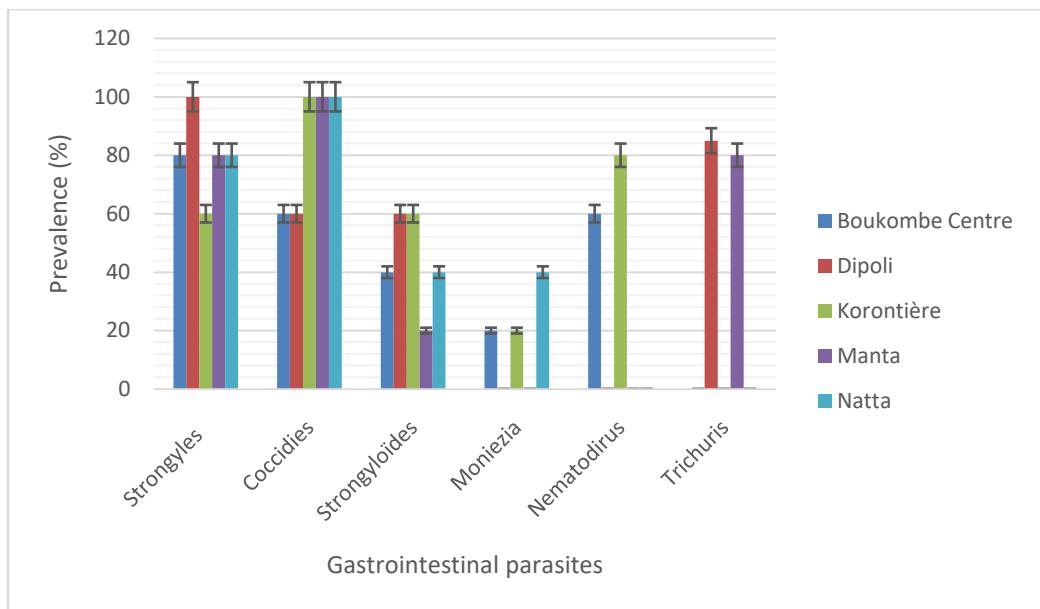


Figure 2. Prevalence (%) of gastrointestinal parasites in Cattle in Boukombé municipality

4. Discussion

The present study demonstrated that gastrointestinal strongyles and *Strongyloides* spp. were detected in all surveyed localities in both sheep and cattle, indicating a widespread and persistent circulation of these parasites in Boukombé municipality. Such a pattern is typical of grazing systems in tropical environments, where climatic conditions favor the continuous development and survival of infective stages on pasture. The high prevalence observed in several localities, reaching up to 100% for some parasite groups, reflects active and uninterrupted transmission, likely exacerbated by extensive grazing practices and limited parasite control measures.

The spatial heterogeneity observed in parasite distribution, particularly for *Moniezia* spp. and coccidia, suggests that local environmental and management factors play a major role in shaping parasite dynamics. The restricted occurrence of *Moniezia* spp. may be explained by the uneven distribution of intermediate hosts (oribatid mites) and differences in pasture ecology. Similar locality-dependent patterns have been reported in previous Mini-FLOTAC-based studies, which

highlighted the importance of adapting parasite control strategies to local conditions rather than applying uniform interventions (Cringoli et al., 2017; Rinaldi et al., 2014).

Quantitative analysis of parasite burdens expressed as eggs per gram of feces (EPG) revealed marked differences between localities. In sheep, higher egg counts for strongyles and *Strongyloides* spp. were recorded in Boukombé Centre, while in cattle, strongyle burdens were particularly high in Manta and Boukombé Centre. These findings are epidemiologically significant, as high EPG values are associated with increased pasture contamination and elevated risk of clinical disease and production losses. The ability to accurately detect such differences is strongly dependent on the diagnostic sensitivity and precision of the coprological method used.

In this regard, the present results further confirm the diagnostic robustness of the Mini-FLOTAC technique. The clear differentiation of parasite prevalence and infection intensity across localities obtained in this study is consistent with previous evaluations demonstrating that Mini-FLOTAC provides higher analytical sensitivity and lower variability than traditional techniques such as McMaster. Cringoli et al. (2017) established Mini-FLOTAC as a standardized and highly sensitive method for the diagnosis of helminth and protozoan infections in both humans and animals, supporting its application in epidemiological surveys.

Several comparative studies have confirmed these advantages across different host species and epidemiological contexts. Noel et al. (2017) showed that Mini-FLOTAC yields more accurate and precise fecal egg counts than McMaster in equine strongyle infections, while Godber et al. (2015) reported comparable or superior performance of Mini-FLOTAC relative to FECPAK. Similarly, Johnson et al. (2022) demonstrated that Mini-FLOTAC is particularly effective in capturing parasite overdispersion and prevalence in naturally infected North American bison, highlighting its suitability for both domestic and wild ruminants.

The present findings are also consistent with previous studies conducted in Africa using Mini-FLOTAC. Alowanou et al. (2021) reported that Mini-FLOTAC outperforms the McMaster technique in detecting gastrointestinal parasites in West African Dwarf sheep, goats, and crossbred rabbits, providing more reliable estimates of parasite burden. More recently, Alowanou et al. (2025) confirmed the superior performance of Mini-FLOTAC in West African long-legged

lambs in southern Benin, emphasizing its value for routine diagnosis and large-scale epidemiological monitoring under tropical conditions.

Strong methodological support also comes from studies conducted in Europe. Rinaldi et al. (2014) demonstrated that Mini-FLOTAC offers improved precision for assessing gastrointestinal strongyle infection intensity and anthelmintic drug efficacy in sheep, both at the individual and pooled-sample levels. Furthermore, Bosco et al. (2018) showed that Mini-FLOTAC achieves higher and more consistent recovery rates of nematode eggs from fecal samples than other commonly used coprological methods. More recently, Bosco et al. (2023) highlighted the versatility of Mini-FLOTAC by demonstrating its effectiveness in detecting and quantifying trematode eggs (*Fasciola hepatica* and *Calicophorondaubneyi*) in bovine feces, further reinforcing its applicability beyond nematode infections.

Taken together, the consistency between the present findings and those reported by Cringoli et al. (2017), Rinaldi et al. (2014), Bosco et al. (2018, 2023), Noel et al. (2017), Godber et al. (2015), Johnson et al. (2022), and Alowanou et al. (2021, 2025) strongly supports the use of Mini-FLOTAC as a reliable and comprehensive diagnostic tool for gastrointestinal parasite surveillance. In the context of Boukombé municipality, the application of Mini-FLOTAC enabled precise identification of high-risk localities and parasite groups, providing a solid evidence base for the design of locally adapted and sustainable parasite control strategies.

Overall, these findings underline the need to strengthen evidence-based parasite management, including rational use of anthelmintics, improved pasture management, and regular epidemiological monitoring using sensitive diagnostic tools such as Mini-FLOTAC. Such integrated approaches are essential to reduce gastrointestinal parasite burdens and improve the productivity and resilience of ruminant production systems in northern Benin.

5. Conclusion

The present study provides comprehensive evidence of the high prevalence and spatial heterogeneity of gastrointestinal parasite infections in cattle and sheep raised under smallholder farming systems in Boukombé municipality, north-western Benin. The widespread occurrence of

gastrointestinal strongyles and *Strongyloides* spp., combined with high egg counts in certain localities, indicates active and sustained parasite transmission, likely driven by extensive grazing practices and limited implementation of effective control measures. The application of the Mini-FLOTAC technique enabled accurate detection and quantification of parasite burdens, clearly discriminating differences in infection intensity between localities. These results confirm the robustness, sensitivity, and field applicability of Mini-FLOTAC as a reliable diagnostic tool for gastrointestinal parasite surveillance in tropical livestock systems. Overall, the findings emphasize the need to strengthen evidence-based parasite management, including regular parasitological monitoring, rational use of anthelmintics, and improved pasture management practices. Integrating sensitive diagnostic tools such as Mini-FLOTAC into routine veterinary surveillance programs will be essential to reduce the burden of gastrointestinal parasitism, enhance livestock productivity, and support the sustainability of ruminant production systems in northern Benin.

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7. Ethics approval and consent to participate

The present study was approved and conducted in accordance with the guidelines of the Ethical Committee of the National University of Sciences, Technologies, Engineering and Mathematics, Abomey, Benin (EC approval 2025/11454) and after receiving approval. Data collection was conducted using the guidelines of the World Association for the Advancement of Veterinary Parasitology (WAAVP). In addition, it is important to note that consent was obtained from all participants, especially from the owners of sheep and cow farms where the faecal samples used in this study were collected.

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