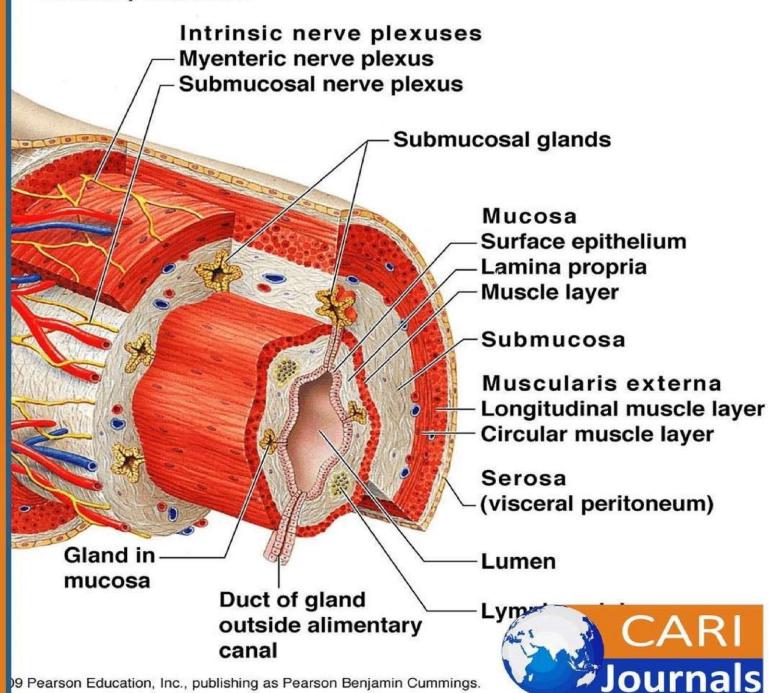
Efficacy of on Farm Bovine Stool Examination as Compared to Postmortem for Fascioliasis

Visceral peritoneum



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Efficacy of on Farm Bovine Stool Examination as Compared to Postmortem for Fascioliasis

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Abstract

Purpose: The aim of this research was to determine prevalence of fascioliasis by examination of liver for adult flukes and bovine stool at the farm in Uasin Gishu County.

Methodology: Stool examination and routine meat examination procedures were used.

Findings: The results at postmortem showed that female bovine were significantly (P<0.05) more infected (47.4%) compared to male cattle (32.8%). Avishire breed had higher infection rate (38.1%) compared to the other breeds, with the local breed having a significantly (p < 0.05) lowest infection rate (11.5%) Older cattle (over 48 months) had a significantly (p < 0.05) higher infection compared to the younger age groups. More slaughtered cattle were sourced from Uasin Gishu compared to other regions and they were also significantly (P<0.05) more infected (at 47.1%) than cattle from the other regions. Results for stool examination also had a similar trend. There was a significant difference in the rates of Fasciola infections in cattle in the six Sub counties. Fasciola infection between the male and female, between Ayrshire, Friesian and local breed, between age 8-24, age 25-48 and age 48 were also significant. More females were infected followed by animals aged between 25-48 months. Friesian breed was the most infected and sampled on farms in the field among the breeds with the least infected being the local breeds which showed more resistance to fascioliasis than the other groups. F. hepatica was the most common species infecting the cattle, although some animals were found to have mixed infections. The results of stool analysis also showed that more female cattle and the animals aged between 25-48 months were more infected. Friesian breed were more infected during stool examination among the bovine breeds with the least infected being local breeds which suggests that they are more resistance to fascioliasis than the exotic breeds. Stool analysis prevalence was 28% while liver examination was 40%. Stool analysis was found to be a suitable method for routine management of livestock in order to reduce losses that are associated with liver flukes.

Unique contribution to theory, policy and practice: Uasin Gishu contributed the highest number of slaughtered animals in the Eldoret main abattoir and therefore farmers in the region should be advised on the best cattle management practices such as regular stool analysis deworming and avoiding grazing their cattle in potentially snail infested areas.

Keywords: Fascioliasis, Bovines, Prevalence, Species, Stool.





INTRODUCTION

One of the most significant helminthic parasites that cause fascioliasis, often known as liver fluke illness in ruminants is the Fasciola species (Bozorgomid et al., 2018). According to Aryaeipour et al. (2017), fascioliasis is a Neglected Tropical Disease (NTD) that is widespread throughout the tropics. In addition to affecting cattle, sheep, and goats, the disease also affects humans, with over 2 million cases of infection reported annually (CDC, 2015). The two primary species of Fasciola, Fasciola hepatica and Fasciola gigantica, are the etiological agents of fascioliasis (Mekky et al., 2015). The bile duct and gall bladder of the sick animal are home to both species, and they severely injure the affected organs (Mas-Coma et al., 2019). Freshwater snails of the genus Lymnae, which live in marshy environments, are the carriers of Fasciola hepatica and Fasciola gigantica (Jaja et al., 2017). In the six sub-counties of Kenya's Uasin Gishu County, suitable habitat for the snails is widespread in the marshy regions (Murgor et al., 2018). The favourable climatic conditions in Uasin Gishu County encourage the production of cattle (Mabonga and Ogalo, 2020), however the prevalence of fascioliasis prevents ideal animal productivity. By causing mortality, decreased milk output, stunted growth, miscarriages, decreased meat production, and increased production costs, fascioliasis causes significant economic losses in livestock (Jaja et al., 2017). In order to determine the prevalence of fascioliasis, the majority of African nations heavily rely on data from slaughterhouse surveys (Assefa et al., 2015). Tests that are both preliminary and confirmatory are used to make the fascioliasis diagnosis (Daksa et al., 2016). According to Daksa et al., (2016) the basis for a tentative diagnosis is the observation of clinical symptoms, prior knowledge of fascioliasis in the area, and the presence of the snail vector in the grazing pasture. Contrarily, confirmation diagnosis entails observing fecal eggs during routine laboratory fecal examination, examining infected animals after death, and showing immature and adult flukes in the liver or by parasite-specific antibody detection (Daksa et al., 2016). The use of molluscicides in marshy areas, feeding cattle with pasture harvested from parasite-free areas, draining and fencing marshy areas, and other strategies were described by Ashrafi et al. (2014) as ways to decrease the exposure of sensitive animals to snail-infested areas. This current study aimed at determining the Efficacy of on farm bovine stool examination of fascioliasis as compared to liver examination in Uasin Gishu County.

MATERIALS AND METHODS

Study area

Postmortem study was based in Eldoret town. Nearly 300,000 people have settled in Eldoret town due to employment opportunities, while others are there due to immigration. There are a variety of livelihoods available in the region, mostly in the agricultural sector. The study area is 315 kilometers northwest of Nairobi, the capital of Kenya. It is located at an average elevation of 2071 meters above sea level. Its coordinates are 0.514277 latitude and 35.269779 longitude. 0° 30' 51.3972" N and 35° 16' 11.2044" E are the GPS coordinates.

Sampling technique and study procedures



Systematic sampling of slaughtered bovines was used until the desired sample size was achieved.

Routine inspection of the livers at the main slaughterhouse in Eldoret.

The primary/main abattoir for Eldoret Municipality was chosen on purpose since it accepts livestock from practically every region of the Republic of Kenya. Using the routine liver examination, systematic sampling of slaughtered bovines was performed until the desired sample size was obtained. The Fasciola species infection in the bovines was determined and compared. Each owner of cattle presented for slaughter was contacted and requested to consent for participation and complete a structured questionnaire. The sex, breed, and origin of the animals that were slaughtered were all recorded during the interview. To assess the relative abundance of the identified Fasciola species, the liver was thoroughly examined and adult liver flukes counted for each infected animal liver. Each infected liver had its bile ducts punctured, and at least six exposed adult liver flukes from infected cattle livers were collected, kept in universal bottles containing 70%, alcohol and transferred to the laboratory at the Regional Veterinary Laboratory-Eldoret for morphological taxonomic analysis. The eruption of the teeth was used to estimate the age of the bovine. Using the findings from the examination of every liver during the study period, the prevalence of fascioliasis in the abattoir was calculated. By dividing the total number of positive (infected) animals by the total number of examined animals, multiplied by 100, the distribution prevalence by bovine attributes (sex age, breed and source) was determined. The main abattoir was visited on Monday, Wednesday, and Friday of each week to examine and count the number of liver flukes from animal carcasses. The source, breed, age and sex of the cattle was noted and analyzed to ascertain the fascioliasis distribution by these attributes. The preserved flukes were transferred to petri plates and spread out on the table while being subjected to morphological study. They were placed on a sheet of fresh white paper then their size and morphology were used for their identity. A pencil and a ruler were used to measure their sizes. By tracing and measuring the liver flukes' length and width on the white paper and recording the results, the exact size of the liver fluke was ascertained. F. hepatica liver flukes are defined as being less than 40mm long, shaped like a leaf with very broad shoulders, pointy at the back, and grey in color when preserved in 70% alcohol. While F. gigantica is 24-27mm long ,5-13 mm wide with an average length/ width ratio of 4.39-5.20 mm

Using stool examination to diagnose bovine fascioliasis

In order to complete the geographical scope of the study in Uasin Gishu County, Ainabkoi, Kapseret, Kesses, Moiben, Soy, and Turbo sub-counties/constituencies were sampled in stool diagnosis for fascioliasis and snail survey. Large marshy areas and papyrus reeds in the aforementioned sub-counties/constituencies act as snail habitats. The targeted livestock farms were chosen using a straightforward random selection method (Kusmayadi & Hadist, 2023)

A prospective cross-sectional study was done for a period of six months. Cattle of all ages and both sex were included in the investigation. The desired number of fecal samples (420) was distributed equally across the six sub counties. Stool was collected directly from rectum of live animals in the



farm into vials and kept in an ice-filled cooler box. The samples were then transported to the laboratory for analysis using the Foreyt (2005) outlined sedimentation procedure to look for *Fasciola* eggs.

By identifying the existence or lack of *Fasciola* eggs in the feces, fecal analysis was used to validate the results of the routine liver inspection by comparison analysis. About five grams of the fecal matter was collected from the rectum of each sampled animal and placed in a labeled 40g universal bottle, and transported to the laboratory in a cool box, Before the stool specimen is passed through a tea strainer into a 250ml beaker, the samples were thoroughly mixed using a wooden spatula.

The sieve's interior was flushed with a forceful stream of tap water to push through the clinging tiny fecal particles. The 250ml beaker was then filled with water until it is 3/4 full, and the sediments were allowed to settle for five minutes before being decanted. Tap water was used to refill the 250 ml beaker, and this process continued until the overlap is gone. Two drops of methylene blue stain was then applied to the resulting sediment in a clean petri plate. The dyed sediment was examined with an X400 compound microscope. According to Berhe (2009), the eggs of *Fasciola* species are seen as yellow structures on a blue background.

Statistical Analysis:

Postmortem diagnoses was determined using Chi-square with significance set at P=0.05. while stool analysis was done using one way ANOVA.

RESULTS AND DISCUSSION

RESULTS

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Table 1: Factors associated with the prevalence of fascioliasis in cattle slaughtered at the main abattoir in Eldoret municipality, Uasin Gishu County, Kenya

Variable	Category	Ν	Positive (%)	P value (χ^2)
Sex	Male	228	74 (32.5)	18.85; $P < 0.05$; $\chi^2 = 3.841$
	Female	192	91 (47.4)	
Breed	Ayrshire	181	69 (38.1)	36.45P < 0.05; $\chi^2 = 5.991$
	Friesian	187	42 (22.4)	
	Local	52	6 (11.5)	
Age (Months)	8 - 24	270	35 (13.0)	59.17P < 0.05; $\chi^2 = 5.991$
	25-48	122	41 (33.6)	
Source	48 and above West Pokot	28 59	12 (42.9) 6 (10.2)	69.09 P < 0.05;
	Nandi	123	35 (28.5)	$\chi^2 = 7.815$
	Uasin Gishu	225	106 (47.1)	
	Marsabit	13	2 (15.4)	
Fasciola species	F. hepatica	139	35 (25.2)	17.00.P < 0.05;
in cattle				$\chi^2 = 5.991$
	F. gigantica	139	21 (15.1)	
Total	Mixed infection	142 2,100	17 (12.0) 592 (28.2)	

Key: N = sample size

Table 1 shows that females (47.5%) were significantly (P<0.05) more infected with fascioliasis compared to male cattle (32.5%). In terms of breed, Ayrshire were found to have higher fascioliasis infection rate (38.3%) compared to the other breeds, with the local breed having the lowest

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infection rate (12.1%), the differences in infection rates between breeds were statistically significant (P < 0.05). Older cattle (over 48 months) had a higher rate of fascioliasis infection than the younger groups, this was also statistically significant (P < 0.05). More cattle were sourced from Uasin Gishu compared to other sources and they were also significantly, (P < 0.05), more infected (at 47.0%) than cattle from the other regions. In terms of species, *F. hepatica* was found to be the most common species infecting the cattle, although some animals were found to have mixed infections.

Burden of fascioliasis infection in slaughtered cattle at Eldoret main abattoir

Factor	Category	Fascioliasis infection burden (%)				
		Light (1 -30)	Moderate(31-	Heavy (>100)		
			100)			
Sex of Cattle	Male	33(52.5)	28(44.7)	02(2.80)		
	Female	56(52.3)	29(26.4)	23(21.3)		
Breed of Cattle	Ayrshire	28(40.9)	32(46.2)	09(12.9)		
	Friesian	15(34.7)	19(44.3)	08(22.7)		
	Local	01(16.7)	03(58.3)	04(25.0)		
Age of Cattle (Months)	8 - 24	09(26.9)	16(44.8)	10(28.3)		
	25-48	14(34.7)	21(50.6)	06(14.7)		
	48 and above	06(48.1)	05(44.4)	01(7.5)		
Source of Animal	West Pokot	01(9.1)	02(36.4)	03(54.5)		
	Nandi	15(42.4)	13(36.4)	7(21.2)		
	Uasin Gishu	33(31.2)	52(49.0)	21(19.8)		
	Marsabit	02(100)	00(00)	00(00)		

Table 2: Burden of Fascioliasis infection in cattle liver in Uasin Gishu County.

Infection with fascioliasis was observed to differ significantly depending on source of animal and sex (P < 0.05).

In order to identify potential risk factors for the disease, fascioliasis infection load in cattle slaughtered at the main abattoir in Eldoret was determined and divided into light, moderate, and heavy infections. As shown in Table 2, the parasite was present in more female cattle than male cattle, with the majority of these infections being mild. The parasite burden was higher in Ayrshire and Friesian breeds than in local breeds.

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In comparison to the other regions, animals from West Pokot had a larger percentage of elderly animals over 48 months which were infected.

Cow Vari	able	Ainabkoi	Kapseret	Moiben	Kesses	Soy	Turbo
	Male	7.01±0.01 ^a	6.11±0.13 ^b	5.42±0.08 ^c	4.52±0.37°	9.25 ± 0.48^d	7.25±0.47 ^a
Sex	Female	13.21±0.05 ^b	14.06±0.36 ^b	15.86±0.04 ^b	13.21±0.25 ^b	12.25±.21 ^a	$12.02{\pm}0.08^{b}$
	Friesian	10.26 ± 0.07^{d}	11.26±.15 ^b	14.09±0.21 ^b	10.80±0.56 ^b	6.58 ± 0.03^{b}	4.06±0.12 ^{ab}
u	Ayshire	7.22±0.23 ^a	$8.32{\pm}0.04^d$	5.25±0.02 ^c	$8.28{\pm}0.14^d$	13.06±0.58 ^b	10.26±0.29 ^b
Breed	Local	5.02±0.02 ^c	1.01±0.02 ^{ab}	0.21±0.11 ^{ab}	1.09±0.62 ^{ab}	4.81±0.16 ^{ab}	5.02±0.16 ^c
	8-24	5.56±0.07°	2.61±0.30 ^{ab}	3.31±0.12 ^{ab}	3.28±0.03 ^{ab}	2.28±0.30 ^{ab}	4.28±0.15 ^{ab}
onths)	25-48	$9.90{\pm}0.04^{d}$	12.07±0.03 ^b	$8.07{\pm}0.28^d$	11.90±0.46 ^c	13.53±0.13 ^b	8.05 ± 0.40^d
Age (Months)	48 and above	5.64±0.16 ^c	$7.25{\pm}0.05^{a}$	8.53 ± 0.02^d	4.24±0.35 ^{ab}	5.91±0.30°	7.52±0.36 ^a

Table 3: Mean Fasciola egg counts in stool of cattle in sub-counties of Uasin Gishu

Means with similar superscript letter do not differ significantly from each other at $P \le 0.05$

There was a significant difference in the rates of *Fasciola* infections in cattle in the six Sub Counties (P < 0.05). There were also significant differences (P < 0.05) in *Fasciola* infection between the male and female, between Ayrshire, Friesian and local breed, between age 8-24, age 25-48 and age 48.

More females were infected followed by animals aged between 25-48 months. Friesian breed was the most infected and sampled on farms in the field among the breeds with the least infected being the local breeds which showed more resistance to fascioliasis than the other groups.

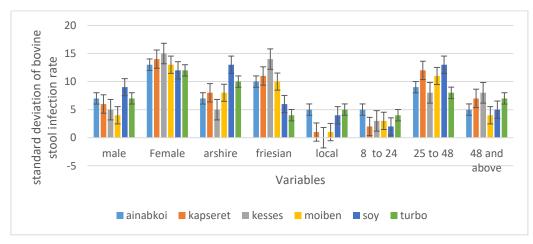




Figure 1: Distribution of bovine stool infections with *Fasciola* in the six sub-counties of Uasin Gishu County.

DISCUSSION

Fasciola parasite infection rates of bovines slaughtered at Eldoret main abattoir.

In the main abattoir in Eldoret, more female animals are slaughtered than male cattle. Farmers in Kenya's Uasin Gishu County and other Counties favor raising more female milk-producing animals over male ones (Njenga et al., 2011; Riang'a et al., 2017). This explains why there are more female cows available for slaughter at calling than male cattle. According to the current study's findings, more female cattle were found to be infected than male cattle, which is consistent with the results of other earlier research (Ardo et al., 2013). The infection difference by sex is statistically significant. Other studies by Phiri et al., (2005); Adedokun et al., (2008) and Ardo et al., (2013), indicated that female cattle had higher infection rate than male cattle by more than 5%. This shows that the higher infection rates in female cows may be attributable to the variations in the numbers of sampled female and male animals, increasing the likelihood that female are more likely to be infected than males in the study area. Less than 20% of local Zebu and Boran breeds are raised by farmers in the study area, with the majority being Friesian and Ayrshire breeds (Mabonga, and Ogallo, 2018). Numerous cattle in the area are hybrids of exotic and indigenous breeds. The productivity of the cattle, particularly in milk production, influences the choice of breed (Mabonga, and Ogallo, 2018). All age groups of cattle experience an equal attack by fascioliasis causing parasites. However, cattle older than 48 months in the current study exhibited a greater infection rate than the other age groups (Takeuchi-Storm et al., 2018; da Costa et al., 2019). This is probably caused by the weakened immune systems of older animals and the emergence of parasite resistance to commonly used anti-helminth medications (Welstenholme et al., 2004). Compared to other nearby Counties, Uasin Gishu County contributed the highest number of livestock to the main abattoir in Eldoret. This may be because catchment regions are closer to the main abattoir in Eldoret than other Counties, where travel by expensive motor vehicle may be necessary because they are far away. Furthermore, because Uasin Gishu is primarily a highland plateau with abundant rainfall, it has wetlands (slow-moving streams, swamps, and dams) that are ideal habitats for the snails that transmit fascioliasis (Njenga et al., 2011). This could explain why there were more infections in the cattle from this County than from other source Counties. The cattle livers that were examined for fascioliasis, F. hepatica was the predominant species (Ichikawa et. al., 2016; Baran et. al., 2017; Getahun et. al., 2017; Pinilla et. al 2020). This is due to its smaller size, which allows for effective development in a variety of hosts including small ruminants, F. hepatica may be the dominant species. The larger species F. gigantica probably experience narrow host range and thus its smaller population distribution.

Burden of Fascioliasis in infected cattle

Infection levels were primarily light to moderate. Only the animals that came from West Pokot were heavily infected. This may be attributed to the high rate of illiteracy among the many cattle

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farmers in the West Pokot region (Njenga et al., 2020). It is also probable that the veterinary officers' efforts may be less than in other regions where the animals were sourced. West Pokot is among the country's arid and semi-arid regions, which means it has fewer predisposing factors (lack of suitable habitats for the snail vectors). According to Kanyari *et al.*, (2017) and Kipyegen *et al.*, (2022), areas where there are more snails and other wetlands—suitable habitats for the snails that transmit fascioliasis typically have higher parasite infection rates.

Bovine stool in the six sub counties of Uasin Gishu County and fascioliasis infection

Uasin Gishu County is found in the Kenya highlands characterized by many wetlands which rarely dry and papyrus reeds (Mulei *et al.*, 2016), this offers a good environment for thriving of snails. There were more infected female than male cattle. In most homesteads, there were more female than male with some of the homesteads having only female cattle. This is attributed to the financial returns that come with keeping female cows by selling of milk and expanding the herd when they give birth (Mabonga and Ogallo, 2018). Animals aged between 25-48 months came second in infection. This is attributed to their physiological status and activity (Agyemang *et al.*, 1991). This is the age that is more productive, most cows are served from the eighteenth month and therefore start milk production and parturition on a yearly basis (Esslemont *et al.*, 2020). When they are incalf and lactating, their immune defense is lower and hence more predisposed to parasites than the other age groups (Agyemang *et al.*, 1991).

During stool examination, Friesian were the ones that were most infected and the most sampled during on farm visits. The least infected were the local breeds. In most parts of the country under normal or natural conditions, they rarely survive and are less adaptable unless under zero grazing with close monitoring (Voinot *et al.*, 2020). Local breed showed more resistance to fascioliasis than the other groups. Local breed being native, there might be co evolution with the parasites as compared to Friesians which are not native in Kenya (VanderWaal *et al.*, 2014).

There was a significant difference in the rates of infections in the six sub counties P < 0.05. Soy recorded the highest infection percentage on average with the least infection recorded in Moiben. This may be due to the frequency of deworming, some farmers normally do a regular deworming compared to others (Thapa *et al.*, 2020)

Comparison of stool analysis and normal meat inspection procedures

Results from stool analysis indicated that there were more females infected than males. More homesteads had more females domesticated than males due to the financial returns associated with dairy animals (Mabonga and Ogallo, 2018). Stool analysis also showed that animals aged between 25 to 48 months were more infected in all the age groups due to their physiological status as some are lactating and at the same time in-calf (Agyemang *et al.*, 1991). Additionally Friesian breed was more at risk of infection than the other breeds. This being an exotic breed, it is less adapted and also lacks coevolution with the local parasite species (VanderWaal *et al.*, 2014). The Local breed was more resistant to the parasites attack than the other breeds based on stool analysis.

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When using normal meat inspection procedures on the other hand, it was found that more female animals were slaughtered than male animals in the Eldoret main abattoir. Farmers in Kenya's Uasin Gishu County favor dairy cows for milk production over male (Njenga *et al.*, 2011; Riang'a *et al.*, 2017). After liver inspection, there were more females infected with fascioliasis than males. In addition, animals greater than fourty –eight (48) months were more infected than the other age groups. This could be due to deworming of the more productive animals in the lower age groups by farmers Thapa *et al.*, (2020) and also this older animals have a compromised immune system (Welstenholme *et al.*, 2004). Uasin Gishu County contributed a large number of slaughtered animals to the Eldoret main abattoir and out of this more infections were realized from animals sourced from Uasin Gishu County. Most farmers in the County practice dairy animal farming and crop farming (Njenga *et al.*, 2011). There was a higher infection rate in exotic breeds Friesian and Ayrshire with locals being the least infected.

The two procedures show similarity as shown in the results. Therefore farmers should be encouraged to use stool sample analysis to detect infection and hence make informed decisions during deworming. The percentage prevalence while using stool was 27.9%. While liver inspection was 40.7%. Therefore stool analysis as a method could make farmers make informed decisions for deworming. It will also make them avoid the losses by not deworming their cattle.

CONCLUSIONS AND RECOMMENDATIONS

CONCLUSIONS

This research determined prevalence of fascioliasis by liver examination for adult flukes and bovine stool at the farm in Uasin Gishu County. Stool analysis prevalence was 28% while liver examination was 40%. Stool analysis was found to be a suitable method for routine management of livestock in order to reduce losses that are associated with liver flukes hence recommended.

RECOMMENDATIONS

Liver fluke causes a lot of losses to livestock farmers. Stool examination though had a lower percentage of prevalence is a suitable method for determining fascioliasis in livestock. This will be suitable to reduce losses that arise from liver fluke. Policies should be enacted so that farmers in the county start a regular routine analysis of stool in livestock in order to reduce the losses.

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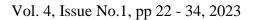
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