Assessment of the occurrence and cause of bloody urine in cattle and management practices in Cheliya, Jibat, and Dire Inchini districts of the West Shewa Zone, Oromia, Ethiopia

Bizunesh Mideksa Borena
Ambo University
Edilu Jorga Sarba
jedilu8@gmail.com

Ambo University
Waktole Terfa Eteya
Ambo University
Negassa Feyissa
Ambo University
Morka Dandecha Bayu
Ambo University
Birhanu Kebede
Ambo University
Getachew Kebebew Tola
Selale University

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Abstract

Background

A study was conducted to assess the causes of bloody urine and management practices in cattle from the Cheliya, Jibat, and Dire Inchini districts of the West Shoa Zone, Oromia, Ethiopia.

Materials and methods

Two kebeles from each of the three districts were selected based on bloody urine case reports, and a total of 215 cattle-owning households were selected for the questionnaire survey. Field observations were performed to screen for possible causes of bloody urine in the study areas. Six animals with symptoms of bloody urine were clinically examined, followed by postmortem and histopathological examinations.

Results

The questionnaire survey results showed that 72.1% (n = 155) of the households had at least one bloody urine case in their cattle herd. Among the three districts, more bloody urine cases were reported in the Cheliya district (95.0%), compared to 67.4% in Jibat and 53.5% in Dire Inchini. About 33.0% of the households reported the death of at least one cattle due to bloody urine; 58.8% of which were from the households in Cheliya, while 40.8% in Jibat, and 4.7% in Dire Inchini district. The occurrence of bloody urine was related to the dry season, feed shortage, access to bracken fern-infested pasture, and a known history of eating bracken fern. There was a high infestation rate of bracken fern (Pteridium aquilinum) in the grazing areas. All six suspected cattle exhibited chronic wasting and hematuria. Centrifugation of the urine samples revealed hematuria. Postmortem examination revealed petechial hemorrhage, papillomatous formations, thickened areas of mucosa, and white/red nodules on the urinary bladder.

Conclusions

Severe infestation of the bracken fern in the study areas, chronic hematuria, and wasting and postmortem findings in the bladder were all suggestive of bracken fern poisoning. Therefore, an immediate and integrated approach should be implemented to prevent cattle from accessing infested areas, remove this plant from grazing areas, and replace it with appropriate forage.

Introduction

Livestock health and productivity can be impacted by infectious and noninfectious diseases, including plant poisoning, throughout the world. Among other diseases, diseases causing red urine are common in some parts of the world, including Ethiopia. In most cases, red urine can be either haematuria, which is the presence of intact red blood cells in the urine and is caused by chronic ingestion of bracken fern,

bovine papillomaviruses, infection, or damage to the urinary tract and kidney. Red urine can also be hemoglobinuria (the presence of lysed blood/hemoglobin in the urine), which is caused by hemoparasites such as *Babesia* spp. and *Anaplasma* spp. and bacterial infections such as bacillary hemoglobinuria (1).

Among the other causes of hematuria, the poisonous plant known as bracken fern is a known concern for grazing livestock worldwide. Bracken fern (*Pteridium esculentum*), a member of the Dennstaedtiaceae family that consists of several subspecies found in multiple parts of the world, is characterized by aggressive growth and extensive underground rhizomes, allowing it to dominate vast areas of grazed land and is a troublesome weed that is difficult to remove (2). It usually grows on burned areas, in woodlands and other shaded places, on hillsides, in open pastures, and in sandy or gravelly soils. All the parts of the fern, including stems, fronds, and spores, are known to contain carcinogens (3, 4).

The health effect of bracken fern poisoning varies depending on the species of animal, the ingested dose, and the duration of consumption (5). Among domestic animals, bracken fern poisoning primarily affects cattle and, less frequently, sheep. Some of the toxic effects of bracken ferns observed in animals include cyanide poisoning, thiamine deficiency, progressive retinal degeneration, acute hemorrhagic syndrome or hemorrhagic diathesis, bovine enzootic hematuria, and upper gastrointestinal tract neoplasia. These clinical conditions arise because of the toxic compounds present in bracken fern, the most poisonous toxin being ptaquiloside, which has various biological effects, including acute bracken fern poisoning, bright blindness, and mutagenic, mitogenic, and carcinogenic effects (4).

Bracken fern poisoning in cattle could result in either acute poisoning or enzootic hematuria. Acute poisoning occurs because of the ingestion of large quantities of bracken fern for a few weeks. Under such conditions, bracken fern poisoning results in depression of the bone marrow with a subsequent reduction in the production of white blood cells and platelets, and cattle may develop an acute syndrome resulting from aplastic anemia and thrombocytopenia. Clinical signs such as depression, loss of appetite, bloody diarrhea, fever, weakness, and death are evident, and secondary infections are common(1, 6). Chronic poisoning (enzootic hematuria) occurs because of prolonged consumption of small quantities of bracken fern, which could result in the development of urinary bladder tumors, leading to persistent hematuria and blood loss. Moreover, tumors can develop in the gastrointestinal tract. Chronic enzootic hematuria is the most common type of bracken fern poisoning in cattle and is characterized by intermittent hematuria and anemia and other signs, such as acute hemorrhagic syndrome, hemorrhagic diathesis, and upper gastrointestinal tract neoplasia (7, 8).

Bracken fern poisoning commonly occurs in the late summer when other feed is scarce, and animals feed excess amounts of bracken fern. In Ethiopia, seasonal fluctuations in grazing pastures are common problems, and especially during the dry season, green pastures are scarce. There are few reports documenting bracken poisoning in domestic animals in addition to the wide distribution of the plant in many parts of the country. The clinical disease enzootic hematuria is reported to cause the death of cattle and loss of production in Ethiopia. The incidence of urinary bladder tumors varies between geographic regions related to the geographic distribution of bracken fern in Ethiopia (9, 10).

This study was initiated in response to repeated clinical reports of mass occurrence of bloody urine cases in cattle from the Cheliya and Jibat districts of the West Shoa zone of the Oromia region, Ethiopia. According to the reports from the respective district's livestock agencies, cattle with clinical signs of bloody urine and emaciation were frequent in these areas, and a significant number of animals died because it did not respond to any treatment. A preliminary field assessment was performed before the actual study, which showed high infestation of bracken fern in the areas and bloody urine cases in the district clinics. Hence, the present study was conducted to assess the occurrence, causes, and management practices of bloody urine cases in cattle of Cheliya, Jibat, and Dire Inchini Districts of West Shewa Zone, Oromia, Ethiopia

Materials and methods

Description of the study area

The West Shoa Zone is one of the zones in the Oromia National Regional State and was found west of Addis Ababa, the capital city of Ethiopia. The Zone has 23 districts, with the administrative center being Ambo Town, which is located 114 km west of Addis Ababa. The zone is located in the central part of Ethiopia at 9°08'22.1"N (9.1394800°) latitude and 37°51'00.6"E (37.8501600°) longitude. It has highland (Baddaa), midland (Badda-Dare), and lowland (Gammoojjii) agroecological zones that cover 27%, 56%, and 17%, respectively, of the zone. The altitude of the zone ranges from 1166–3238 meters above sea level (m.a.s.l.), and most areas lie between 2300 and 2630 m.a.s.l. The mean annual temperature and rainfall range from 11–21°C and 880–1200 mm, respectively. The zone has a bimodal rainfall characterized by a small rainy season from February to May and a large rainy season from June to September. A crop-livestock mixed farming system is a common agricultural practice. The zone has a large livestock population, which is estimated to include 2,294,593 cattle; 1,074,939 sheep; 264,931 goats; 263,558 horses; 11,210 mules; and 265,736 donkeys. More than 97% of the cattle are indigenous breeds that are managed under an extensive system where animals are kept on grazing pasture, but there are also few intensive and semi-intensive cattle production systems where crossbred cattle are kept on improved management in urban and peri-urban areas (11, 12). The human population is estimated to be 2,973,697, of which 1,479,243 are male and 1,494,454 are female (13).

The study was conducted in three districts of the West Shoa Zone, Oromia regional state of Ethiopia, namely, Cheliya, Dire Inchini, and Jibat, which are dominated by highland agro-ecological climatic conditions and dense human and livestock populations. The human population is 229,292,101,371 and 100,724 in the Cheliya, Jibat, and Dire Inchini districts, respectively. The cattle population of each district is estimated to be more than 100,000 (14). The administrative center of Cheliya, Gedo, is located 78 km west of the zonal center Ambo, whereas Dire Inchini (Inchini) and Jibat (Shenen) are located 50 km and 78 km, respectively, southwest of Ambo (Fig. 1).

Study population

Cattle of all ages and sex groups and livestock owner farmers living in the selected study districts were considered the study population.

Study design and data collection methods

A cross-sectional study was carried out from June 2016 to June 2017. Three study districts that have reported frequent bloody urine cases were selected deliberately from the West Shoa Zone of the Oromia region, and within the selected districts, kebeles (the lowest administrative unit within the districts) were also selected purposively based on bloody urine reports from the respective livestock and fisheries agency and field assessment records. Accordingly, two kebeles from each district were selected based on the availability of the bracken fern in the area and complaints from the farms about the bloody urine cases. The data were collected via a questionnaire survey, field observation, clinical examination, postmortem examination, and laboratory investigation of blood and urine samples.

Questionnaire survey

A total of 215 volunteer households (HHs) were selected from the study districts for the questionnaire survey. The head of the HH (male or female) or any elder member of the family above 18 years of age who was familiar with the history of the animals were interviewed. A semistructured questionnaire survey was used to collect the demographic characteristics of the households, livestock ownership, number of sick cattle, number of dead cattle, and cattle management systems. Animal management was categorized as semi-intensive when cattle were kept indoors or tethered around the backyard with some supplemental feed or extensive if cattle were allowed to graze on communal grazing land. Furthermore, farmers were interviewed for their knowledge and practice regarding the bracken fern plant, its poisoning and treatment, and the effect of bloody urine cases on their livelihood. The questionnaire was prepared in English and later translated into the local language '*Afan Oromo*' and administered. Animal health practitioners working in the study districts were trained and served as enumerators to fill out the questionnaires.

Field observations

Grazing areas and cultivated land in the selected kebeles and adjacent areas identified by the district veterinary practitioners were scanned for the presence of the bracken fern, and the plant samples were collected for identification in the Biology Laboratory of Ambo University. Veterinary clinics were also visited to see the availability of bloody urine cases in cattle, and case records were also checked whenever available.

Clinical, postmortem, and laboratory examinations

Six cattle showing typical clinical signs of bloody urine were selected randomly and clinically examined. The body conditions of the animals were scored as good, medium, or poor (15). Approximately 10 ml of blood sample was collected in a new vacutainer tube containing EDTA from the jugular vein of the affected cattle. The samples were subsequently taken to the Veterinary Laboratory of Ambo University

and processed, after which the hematocrit values were recorded. Bloody urine samples were also collected while the affected cattle were micturating naturally, and urine samples were centrifuged at 3000 rpm for 5 minutes to differentiate hematuria from hemoglobinuria based on the appearance of urine after centrifugation. Bloody urine samples were categorized as hemoglobinuria if they remained homogenous after centrifugation or as hematuria if the centrifuged urine formed two layers where the blood settled to the bottom of the tube while the clear yellowish supernatant appeared on top. The sediment was examined microscopically after a thin smear was obtained for the presence of intact red blood cells (RBCs).

The six cattle were slaughtered, and a postmortem examination was performed following the standard necropsy procedure (16). Accordingly, gross pathological changes in each organ of the slaughtered animal were checked, and the changes were recorded. Tissue samples from organs with gross lesions were collected for histopathological examination. The collected tissues were fixed in 10% neutral buffered formalin, processed through graded concentrations of ethanol and xylene, and embedded in paraffin wax. Embedded tissues were sectioned at 3–5 µm and stained with hematoxylin and eosin (H&E) before being examined for histopathological lesions (17).

Data analysis

The data collected through the questionnaire survey were entered into Microsoft Excel, coded, and analyzed by descriptive statistics using STATA version 14. Descriptive analyses, such as frequency and percentage, were used to describe the results, while chi-square test was used to see the associations between bloody urine cases and potential risk factors. Clinical case data from cattle with a history of bloody urine and survey data from the environment were described as observational findings. The gross and histopathology results are also described and supported by the images.

Results

Livestock ownership in the study areas

Among the livestock species, the highest average number of cattle was 8.8 ± 4.1 in Cheliya district, followed by Jibat (8.1 ± 5.2) and Dire Inchini district (7.0 ± 4.3). The household ownership of the different livestock species in the three districts is detailed in Table 1. About 46%, 55%, and 67% of the respondents in Cheliya, Jibat, and Dire Inchini, respectively, own all the mentioned livestock mixed (data not shown).

Table 1
Livestock ownership in the study areas

District	Livestock species	Total No. HH	Total No. of livestock	Average owning	STDV
Cheliya	Cattle	80	700	8.8	4.1
	Shoat	77	646	8.4	5.5
	Equine	66	207	3.1	1.8
	Poultry	45	241	5.4	4.8
Jibat	Cattle	49	395	8.1	5.2
	Shoat	33	284	8.6	6.5
	Equine	32	99	3.1	1.8
	Poultry	28	187	6.7	4.9
Dire Inchini	Cattle	86	598	7.0	4.3
	Shoat	76	552	7.2	6.0
	Equine	76	225	2.9	1.9
	Poultry	65	411	6.3	4.1

HH: household, STDV: standard deviation

Occurrence of bloody urine cases and associated factors in the study area

Among the 215 HHs in the three districts who kept cattle, 155 (72.1%) reported that at least one of the animals in their cattle herd had clinical signs of bloody urine. All the respondents reported that they had bloody urine only in cattle. Among the three districts, the maximum number of HHs with bloody urine cases were 76 (95.0%), in Cheliya, and the minimum of 46 (53.5%) was in Dire Inchini. Similarly, among those HHs who reported bloody urine cases in their cattle, 71 (33.0%) reported that they lost or culled at least one of their animals following the bloody urine symptom. In the Cheliya district, the highest number of HHs (47; 58.8%) and the lowest number of HHs (4; 4.7%) in the Dire Inchini district reported the death of at least one animal from their cattle herd (Table 2).

Table 2
Prevalence of bloody urine in cattle at the herd level in three districts

District	HH with cattle with bloody urine			HHs with death reports from herds with bloody urine cases		
	No of HH	No. HH with sick cattle in a herd (%)	Chi-square (p-value)	No of HH	HH with death report in their herd (%)	Chi-square (p-value)
Cheliya	80	76 (95.0)	36.2 (< 0.001)	80	47 (58.8)	56.6 (< 0.001)
Jibat	49	33 (67.4)		49	20 (40.8)	
Dire Inchini	86	46 (53.5)		86	4 (4.7)	
Total	**215**	**155 (72.1)**		**215**	**71 (33.0)**	

Analysis of the potential risk factors revealed that the type of grazing, season of feed shortage, presence of bracken fern in the locality, and consumption of bracken fern were significantly associated ($p < 0.001$) with the occurrence of bloody urine (Table 3).

Table 3
Chi-square analysis of risk factors for bloody urine cases at the household/herd level

Variables	Category	No. HH	No. HH with bloody urine case in a herd (%)	Chi-square	p-value
Management	Extensive	188	133 (70.7)	1.3528	0.245
	Semi-intensive	27	22 (81.5)		
Grazing type	Communal	121	77 (63.6)	16.56	0.000
	Stall feeding/Tethering	33	22(66.7)		
	Mixed	61	56(91.8)		
Feed shortage	Yes	179	128 (71.5)	0.11	0.738
	No	36	26 (72.2)		
Frequency of feed shortage	Every year	163	116(71.2)	2.56	0.278
	Every 2 year	13	9(69.2)		
	Two wise a year	7	3(42.9)		
	No response	32			
Season of feed shortage	Dry season	168	126 (78.0)	13.0	0.001
	Wet season	34	16 (50.0)		
	Both season	13	7 (53.9)		
The presence of bracken fern in the locality	Yes	173	141(82.6)	49.78	0.000
	No	42	12 (28.6)		
Do animals eat bracken fern	Yes	169	143 (84.6)	57.4	0.000
	No	46	12 (26.1)		
Have you seen symptoms of red urine?	Yes	183	154 (84.1)	85.4	0.000
	No	32	1 (3.1)		
When have you seen symptoms?	This year	125	110 (88.0)	6.5	0.159
	Last year	35	26 (74.3)		
	Last 2–3 years	23	18 (78.3)		
	No response	32			
Age affected	Young age	4	3(75)	1.8839	0.597

HH: Household

Variables	Category	No. HH	No. HH with bloody urine case in a herd (%)	Chi-square	p-value
	Middle age	44	38 (86.4)		
	Adult age	121	105(86.8)		
	All age	9	9(100)		
	No response	37			
Sex affected	Male	29	25(86.2)	0.5470	0.761
	Female	33	30(90.9)		
	Both	115	99(86.1)		
	No response	38			

HH: Household

Findings of clinical and laboratory examinations

All six animals (cattle) with a history of bloody urine cases were stimulated to urinate, and their urine was found to be red. Most animals (66.7%, 4/6) had poor body condition, and the remaining 33.3% (2/6) had good or medium body condition (Table 4). Four out of the six animals had a history of bloody urine for more than a month, whereas the remaining two had a history of bloody urine for less than 15 days. A pale mucus membrane was detected upon examination of the visible conjunctival membrane in four out of the six animals. The collected urine samples had varying degrees of redness. After centrifugation, all the collected bloody urine samples formed two layers, with red sediment at the bottom and clear supernatant at the top, indicating that intact red blood cells settled at the bottom, which is indicative of hematuria. Moreover, analysis of blood samples revealed lower hematocrit levels in four of the six cattle, while these levels were near the normal range in the remaining two cattle.

Table 4
Observational and gross postmortem examination findings

Animal code	Body condition	Change in urinary bladder	Change in heart and kidney	Changes in the rumen, intestine, and liver	Changes in lung	Changes in the carcass
01	Poor	Varying size of tumor and hemorrhage	Petechiae and ecchymosis on the pericardium and endocardium of the heart. swollen and very pale kidneys	Tumor on rumen mucosa and hemorrhagic lesion on intestine	White/red hepatization	white
02	Good	Full of bloody urine, varying sizes of tumor	petechial hemorrhage of the pericardium, red mark on kidneys	Reddish tumor on rumen, lesion on intestine	White/red hepatization	white
03	Poor	Bloody urine and whitish tumor	Extremely whitish kidneys	petechial hemorrhage on the intestinal wall	White and hepatization	White
04	Poor	Small sized tumor	No change in kidneys and heart	No change in rumen and intestine	White	white
05	Medium	Full of bloody urine and nonbleeding tumors on the wall	Petechial hemorrhage on the surface of the heart and bleeding inside the chambers	Lesion on the rumen and intestine, but less severe	White but no hepatization	Less white
06	Poor	Full of bloody urine and varying sizes of bleeding tumors	Heart affected	A small lesion on the small intestine wall	White	white

Gross and histopathological findings

Postmortem examinations revealed multiple hemorrhages throughout the carcass in three of the animals with bloody urine. The urinary bladder of all the necroptized cattle exhibited multiple tumors in the wall and hemorrhages in the urinary bladder mucosa. The gross appearance of neoplastic lesions in the bladder and rumen varied greatly. The neoplastic growths exhibited pedunculated nodules of different sizes that were scattered across the mucosal surface in the urinary bladders. The walls of the urinary bladder of most of the affected cattle were thickened, hyperemic, and edematous, and some of the

tumors had active bleeding. Close observation of the bladder mucosa with bleeding points revealed small hemorrhages; dilated vessels; or vascular, fibrous, or epithelial neoplasms. The other lesions observed during postmortem examination were hemorrhages found on the pericardium and endocardium of the heart. The intestines were filled with blood clots and hemorrhages varying from petechiae to ecchymoses. Additionally, the lung, liver, and kidney were swollen and contained multiple white foci and hemorrhages (Fig. 2).

Histopathological examination of organs from animals with bloody urine symptoms revealed massive hemorrhage, edema, giant cells, and thrombosis in the different affected organs (Fig. 3). Histological examination of liver samples revealed edema, hemorrhage, and inflammation resulting from hepatocyte infiltration with mononuclear cells. Hemangiosarcoma was diagnosed in the urinary bladder in which the tumor mass was composed of blood vessels that were either small or large (Fig. 3D). The urinary bladder samples were characterized by diffuse hemorrhage and infiltration of the lamina propria with mononuclear cells (cystitis). Hemorrhage and mononuclear cell infiltration were also observed in the lung tissue.

Observational findings

Screening of the study areas for possible causes of bloody urine showed the presence of a poisonous plant known for its local name ‘Karo’ and for the scientific name of the species *Pteridium aquilinum* (L.) Kuhnsubsp. *capense* DSC (Thunb.) C. Chr., Antota (in Afan Oromo); Bitibiti (in Bench) and Bracken fern (in English). During the field visit, it was observed that there was a lack of adequate grazing land, the districts were highly populated, and most of the land was used for cultivation. During the wet season, cattle were allowed to graze on the land left between the cultivated plots of land, which were mostly infested with bracken fern, and during the dry season, cattle graze on the cultivated land after harvesting where these plants covered, especially those lands that were recently cleaned from forests or bushes. The grazed leaves of the bracken fern were also observed, and only the stem was left standing in the surveyed localities (Fig. 4).

Management practices for cattle bloody urine cases and the local use of bracken ferns

Farmers in the study areas used to treat their animals suffering from bloody urine using a series of combinations of traditional remedies. These remedies include the use of chopped, ground, and diluted leaves from several medicinal plants; powders from cereals; raw chicken eggs; and wild honey, which is combined by traditional healers. The farmers reported that the treatment responses to these traditional remedies were poor or almost nonexistent. Most farmers knew that none of the modern or traditional treatments applied were effective at curing animals suffering from bloody urine.

It was also observed that farmers cut and use fresh bracken fern stems and leaves for household services, such as traditional coffee ceremonies, instead of grasses, which are commonly used as bedding

under coffee trays locally known as ‘Bidiruu’ and as bedding material to sleep over at home, particularly during the dry season. After the use of the bracken fern for such services, people just discard it somewhere near their homes where cattle have access.

The effect of bloody urine cases on the livelihood of farmers

Among the total surveyed households (n = 215) owning a total of 1693 cattle, 268 (28.9%) cattle had clinical signs of bloody urine, and the farmers lost 221 (13.1%) cattle following bloody urine symptoms and failing to respond to any traditional or modern treatments (Table 5). In addition to the direct losses due to the death of cattle, farmers were also victims of indirect losses associated with reduced productivity of the sick animal, low sale price of live animals, and lack of ox power for crop cultivation.

Table 5
Cattle ownership and loss associated with bracken fern poisoning.

	No. owning	No. sick	No. died
No of cattle	1693	489	221
Mean	7.8	2.3	1.0
STDV	4.5	3.3	2.7
STDV: standard deviation, No.: number			

Discussion

This study was conducted primarily to determine the cause of bloody urine in cattle in the districts of the West Shoa zone of Ethiopia, where bloody urine cases are commonly reported. Studies from different parts of the country, such as Afar (northeastern), Wellega (western), and central Ethiopia, showed that bracken fern, which is a poisonous plant, is present (10, 18). According to these studies, bracken fern was also the predominant cause of bloody urine in cattle in different parts of the country. The present study investigated the causes of bloody urine in cattle suspected to be caused by bracken fern toxicity, which is in line with previous reports in the country.

Chronic intoxication with bracken fern in grazing cattle causes enzootic hematuria in cattle due to the carcinomatous bladder tumors that develop in the urinary bladder (6, 19). The carcinogenic effect of bracken fern poisoning is reportedly associated with its high content of Ptaquilocide, which is the major carcinogen in bracken ferns. Hematuria in bracken fern-poisoned cattle results from vascular changes associated with urinary bladder tumors and/or hemorrhagic cystitis (6).

In the present study, bracken ferns, which are evergreen during the feed-scarce season of the year, are highly prevalent in the study areas, and it was also confirmed that cattle eat the leaves of the plant

whenever green grass is scarce. As poisonous plants, bracken fern is not palatable, and cattle usually avoid eating it; thus, poisoning with these plants is related to poor feeding, such as a shortage of forage, overstocking, poor pasture management, or heavy contamination of hay with bracken fern (20). Accordingly, there was a shortage of grazing land during both the dry and wet seasons in the present study area, which was characterized by highland altitudes and dense human and livestock populations. During the dry season, animals were allowed to graze freely on the cultivated land after harvesting the crops, but since there was no adequate green grass, cattle might have been forced to eat the bracken fern. During the wet season, since most of the land is cultivated, animals are allowed to graze on roadsides and the leftover land between cultivated plots and around the backyard; infertile land is left uncultivated, or some farmers purposely leave some plots of land for private or communal grazing purposes. This is a common situation in most Ethiopian highland areas, which are characterized by high human and livestock populations, and shortages of land for grazing and overgrazing are common problems. This scenario was realized in the present study, in which farmers held a significant number of various species of animals in the studied districts.

It is not simple to diagnose the causes of hematuria in cattle because there is no single confirmatory diagnosis. However, in this study, a combination of approaches was used to investigate the cause. First, the availability of the bracken fern in the areas where bloody urine cases were reported and the access of cattle to the bracken fern were considered. Primary evidence, including the signal that cattle in the study areas eat this plant whenever there is a shortage of feed, was recorded. Clinical examination of the patients revealed clinical signs such as chronic hematuria and severe loss of body condition, followed by signs of anemia, suggestive of bracken fern toxicity. The postmortem lesions and histopathological changes observed, especially in the urinary bladder and several other organs, indicated the presence of varying sizes of tumors associated with bracken fern poisoning. Studies show that tumors of the urinary bladder in cattle are of epithelial and mesenchymal origin and are commonly associated with chronic enzootic hematuria, a syndrome caused by prolonged ingestion of bracken fern (21, 22). In line with this, the postmortem examination of all the affected animals in the present study showed multiple tumors in the urinary bladder wall, which were either pedunculated or broad with marked areas of bleeding; moreover, histologically, hemangiosarcoma and hemorrhages were observed in the mesenchymal part of the bladder, revealing the presence of tumors (7).

There were also tumors in the rumen, hemorrhages in the endocardium of the heart, petechiae hemorrhages in the intestine, swollen and pale kidneys, pneumonic and hepatized lungs, and whitish carcasses, which agrees with the findings elsewhere (23). Hence, the clinical and postmortem findings are consistent with chronic bovine enzootic hematuria induced by chronic ingestion of bracken fern in cattle.

The extent of bracken fern toxicity was very extensive in the area, and almost all the respondent farmers disclosed bloody cases in cattle. There was also clear variation between the districts; Cheliya was the most severely affected district, where 95.0% of the respondents reported at least one hematuria case in their herd and 58.8% of the respondents lost at least one animal after showing symptoms of hematuria compared to other districts. This variation could be related to the level of bracken fern infestation on the

grazing land and the difference in the availability of animal feed among the districts. Moreover, risk factors such as free access to open grazing areas, a season of feed shortage, especially during the dry season, the presence of bracken fern in the area, and a known history of eating bracken fern were found to be significantly associated with the occurrence of bloody urine. Therefore, cattle that have prolonged access to bracken fern probably due to the longer dry season and shortage of other feed, are the most likely candidates for intoxication and may die due to toxicity. In addition, bracken fern is commonly used for different purposes at home, such as cultural bedding under the coffee cup tray during the Ethiopian coffee ceremony and as bedding material to sleep over during the night at some farmer houses. After serving this purpose, households discard the bracken fern somewhere closer to the barn. Cattle in the study areas are thus reported to eat such disposed bracken fern and this practice might have aggravated the bloody urine problems.

Hematuria caused by bracken fern poisoning has no specific treatment (1); however, in the current study area, there were attempts to treat hematuria with antibiotics and Sulpha drugs probably to prevent secondary bacterial infection, or to treat using traditional medications, which are made from local alcohols and medicinal plants. The responses to these medications were very low or absent; as a result, almost all the cattle showing hematuria did not recover. Hence, the socioeconomic effect of this condition is expected to be high. In this study, economic data were not collected; however, it is possible to presume a large loss of valuable animals, such as milking cows and plowing oxen, due to death. When sick cattle are undesirably culled or sold at low prices or are slaughtered at a salvage value or if kept, death is the most likely outcome, implying that the loss is greater than expected. Treatment of sick animals is also attempted, which is costly and has no return, as their recovery is always less possible. During the field survey, the investigation team came across a household that lost 48 cattle due to hematuria in the affected areas, and many households currently do not own a single ox or cow because of death due to this intoxication. Farmers in the study area also reported that the selling price of animals from known bloody urine-affected areas is very low, and most individuals even avoid buying animals from such places. As is already known in most Ethiopian communities, cattle owning has much social value beyond wealth to farmers; hence, losses associated with cattle would have a significant socioeconomic effect on farmers and the country in general.

As a limitation, it was not possible to detect the presence or level of the toxin in the serum, milk, other body fluids, or meat of the affected cattle due to the lack of an appropriate kit in our laboratory or Ethiopia.

Conclusion

The studied areas in the three districts were highly infested with the poisonous plant bracken fern, but the degree of infestation varied among the districts. Bracken fern poisoning was a prevalent cattle health problem in the study areas, with a significant number of cattle affected at the household level resulting in a substantial number of deaths. Factors such as the availability of bracken fern in the area, seasonal shortage of feed, type of grazing, and known history of eating bracken fern were detected as risk factors

for the occurrence of bloody urine in the study areas. The prominent clinical features were loss of body condition, chronic hematuria, petechial hemorrhage in organs such as the heart and intestine, and bleeding tumors in the urinary bladder. The socioeconomic impact of bracken fern poisoning was not studied in the present study; nevertheless, it is presumed that the farmers in the affected areas are greatly suffering from the consequences. Therefore, prevention could include limiting the access of cattle to bracken fern-infested pastures, removing bracken fern by burning and plowing and applying appropriate herbicides to successfully reduce the impact of these plants in affected areas. Cultivation of improved forage could be the best option for managing the shortage of feed during the dry season. Moreover, communities need to be aware of the appropriate management and control of bracken fern in severely infested areas.

Declarations

Acknowledgments

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Ethics approval and consent to participate

Participation in the study was fully voluntary, and all participants provided Informed consent. All the protocols followed in the study were reviewed and approved by the Ambo University Ethics Review Committee.

Consent for publication

Not applicable.

Availability of data and materials

All datasets supporting the conclusions of this article will be available upon reasonable request from the corresponding authors.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

WT, EJ, and BM designed the project. WT, GK, NF, and EJ participated in the field data collection, postmortem inspection, and laboratory work. GK and MD participated in the histopathology analysis and interpretation, BK participated in the botanical work, and BM and EJ performed the data analysis and drafted the manuscript. All the authors critically reviewed the draft manuscript. All the authors read and approved the final manuscript.

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Figures

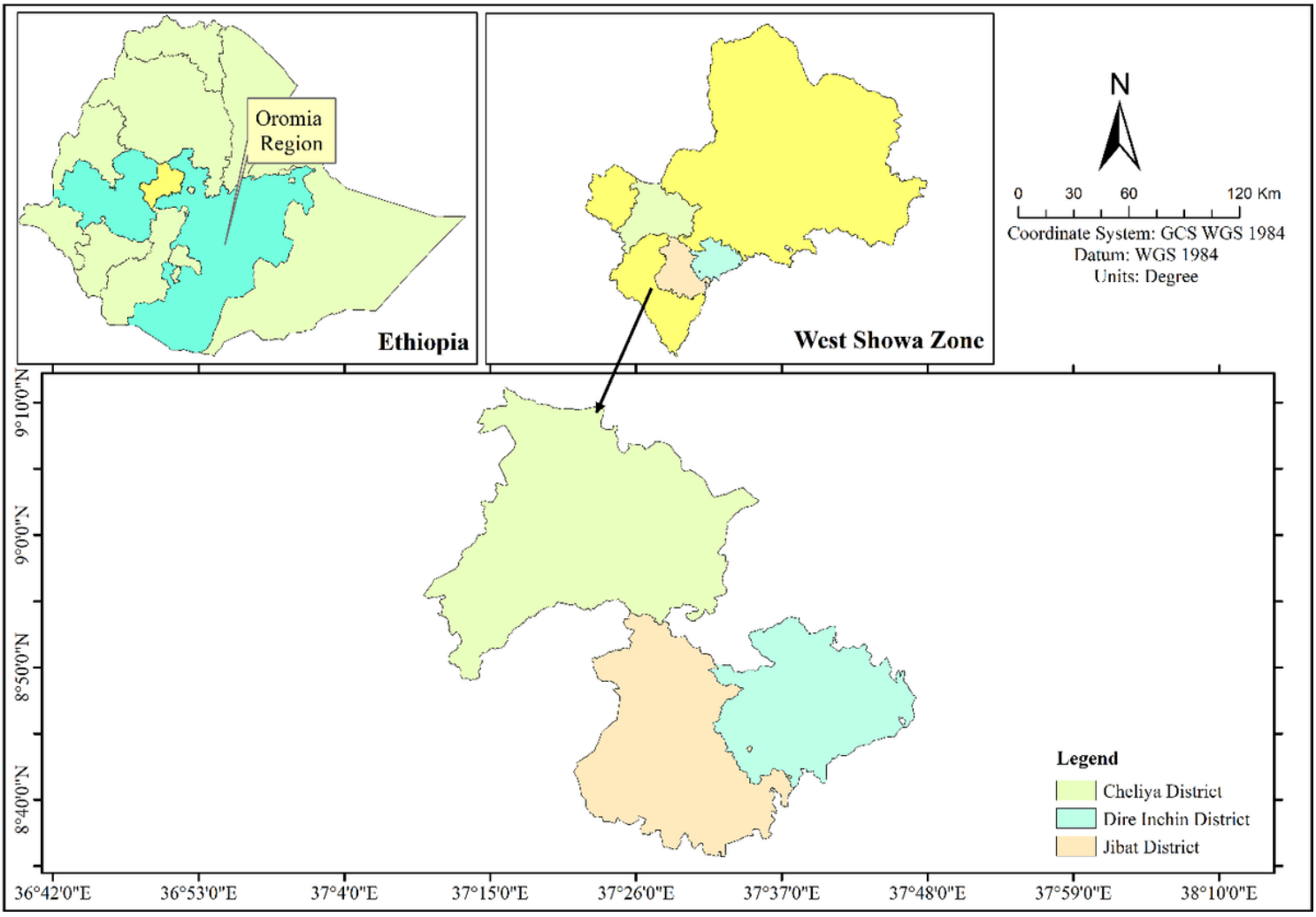

Figure 1

Map of the study area (prepared using ArcGIS)

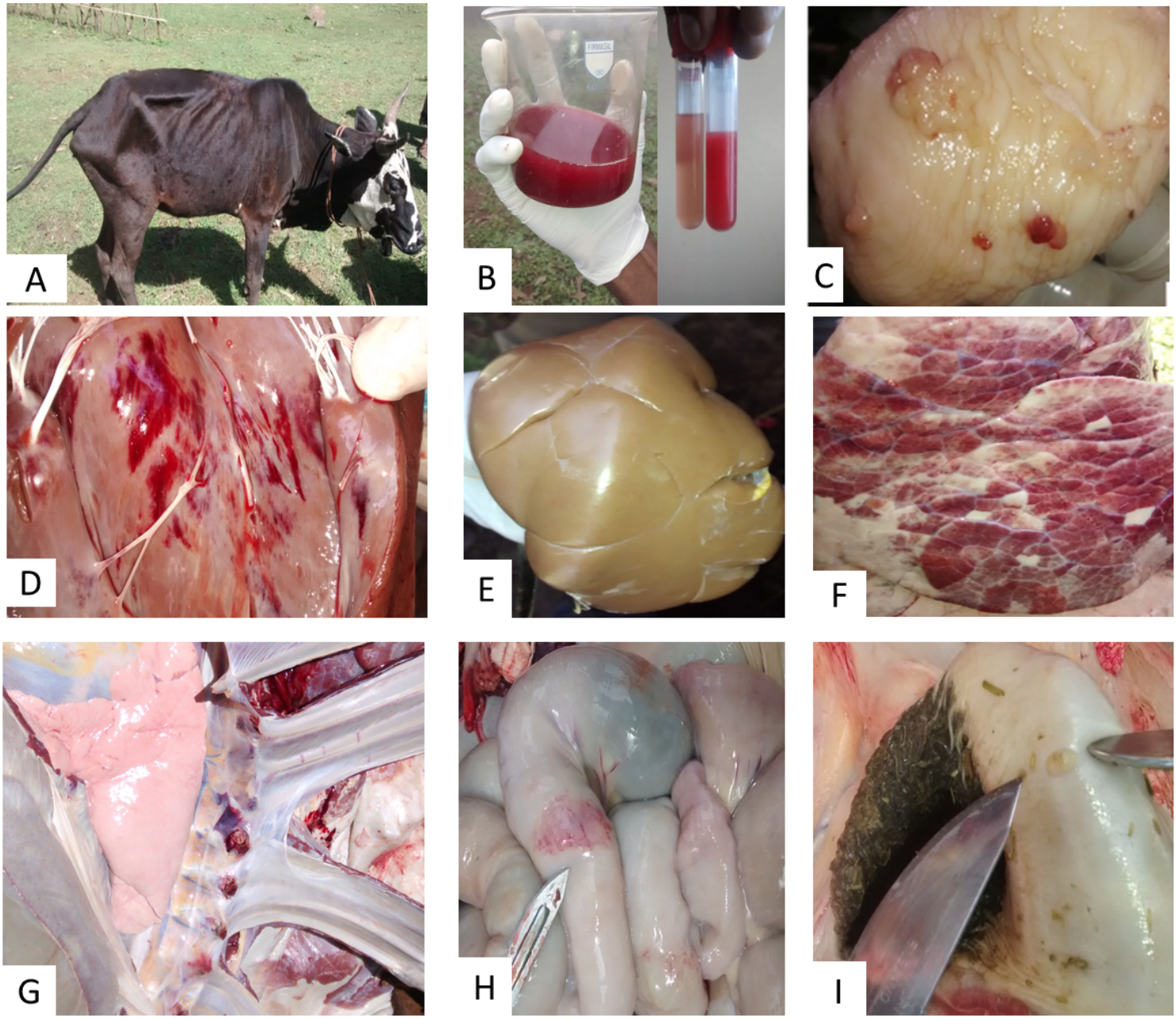

Figure 2

A-I. Clinical signs and macroscopic lesions on organs of cattle with bloody urine. Animal with poor body condition (A), red urine (hematuria) (B), several hemorrhagic masses on the urinary bladder mucosa (C), hemorrhage in the endocardium (D), swollen and pale kidney (E), pneumonic lung with whitish/red hepatization (F), white carcass (G), subserosal hemorrhages in the intestine (H) and papillary growth on the mucosal surface of the rumen (I).

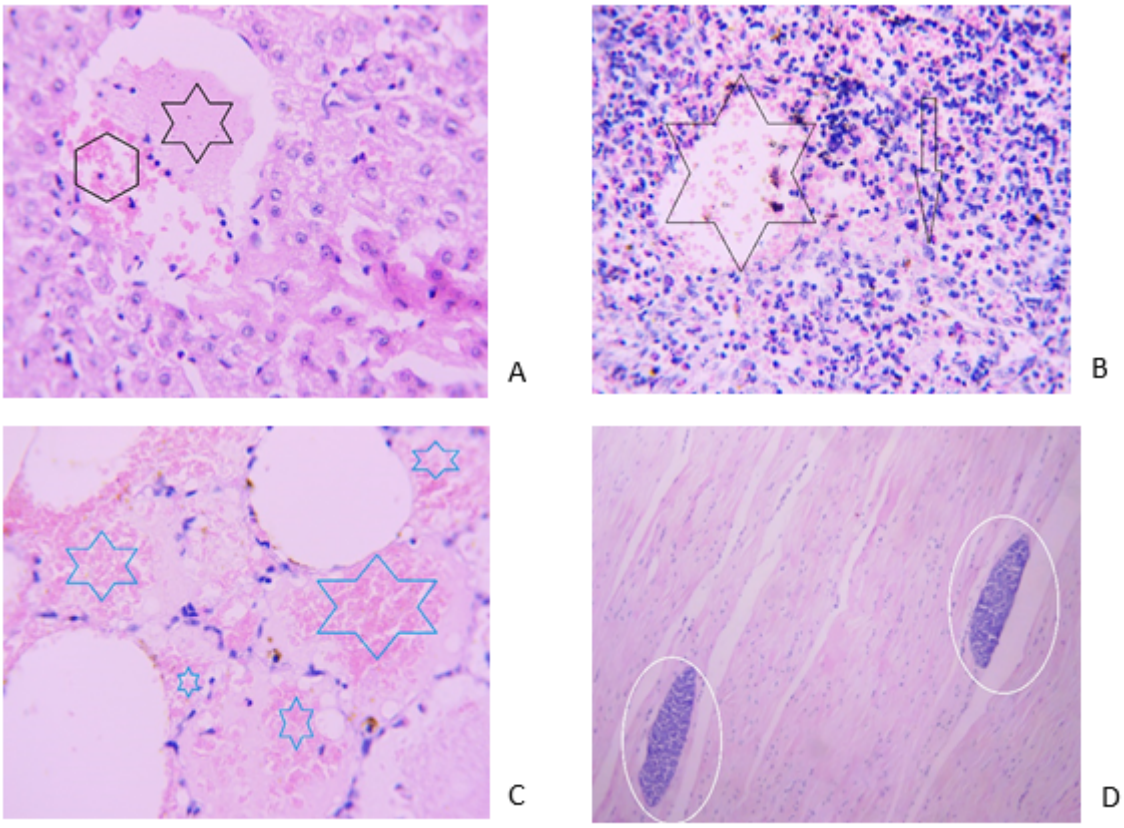

Figure 3

Microscopic lesions on organs from animals with bloody urine symptoms. Hemorrhagic, edematous, and inflammatory hepatocyte infiltration with inflammatory cells (star) in liver tissue (A); diffuse hemorrhage and infiltration in the urinary bladder with mononuclear cells (B); hemorrhage and infiltration of inflammatory cells in the lung tissue (C); and hemangiosarcoma in the urinary bladder (D) in a circular area.

Figure 4

Bracken fern on farmed land after harvesting the crop (A) and Bracken fern that was grazed during the dry season (B).