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Prevalence and Risk Factors of Bovine Trypanosomiasis in Khartoum State, Sudan, April-July 2012

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Prevalence and Risk Factors of Bovine Trypanosomiasis in Khartoum State, Sudan, April-July 2012

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Abstract- A cross-sectional study was conducted in the dry season from April to July 2012 in Khartoum State, Sudan to: estimate the prevalence of bovine trypanosomiasis (BT), identify the prevailing species of trypanosomes and investigate the associated potential risk factors of the disease. Blood samples were collected from 271 randomly selected cattle of the eight study areas and evaluated through standard parasitological methods (Haematocrit Centrifugation Technique (HCT) and thin blood smear). Also, the packed cell volume (PCV) of each animal was measured using Haematocrit Techniques for anemia estimation. The results indicated that the overall prevalence of BT was 4.8% (13/271). All the trypanosomes encountered in cattle belonged to a single species of *Trypanosoma vivax*. For investigation of associated risk factors of the disease, a univariate analysis using the Chi-square (χ^2) test identified 15 risk factors statistically significantly associated with BT ($p \leq 0.25$). These 15 risk factors were entered to the multivariate analysis using logistic regression for further analysis. The significant level of association in the multivariate analysis was set at ($p \leq 0.05$). The multivariate analysis revealed four risk factors that had significant associations with BT ($p \leq 0.05$). These were treatment of sick animals ($p = 0.012$), presence of other animal species in the farm ($p = 0.003$), veterinary care ($p = 0.023$) and location of livestock market ($p = 0.004$). The results of the study indicated that BT is prevalent in Khartoum State and the predominant trypanosome was *T. vivax*. Furthermore, the risk factors found statistically significantly associated ($p \leq 0.05$) with BT in this study should be considered as predictors for the disease and should be taken into account when planning for control programs of the disease.

Keywords: prevalence, risk factors, cattle, trypanosome-*vivax*, Khartoum state, sudan.

I. INTRODUCTION

African animal trypanosomiasis is indisputably a great problem on the African scene, but the perspective alters when it is examined against

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a global background, even when taking into account mechanically and venereally transmitted African trypanosomes which have spread to other continents (*Trypanosoma evansi* causing surra, mechanically transmitted *T. vivax* and *T. equiperdum* the causal agent of the venereal disease dourine) (Uilenberg 1998). Trypanosomiasis is a parasitic disease caused by species of flagellated protozoa belonging to the genus *Trypanosoma* which inhabit the blood plasma, various body tissues and fluids of vertebrate host. The disease is transmitted cyclically by tsetse flies (*Glossina* species), and non-cyclically by other biting flies (Tadesse et al. 2011). Trypanosomiasis induces loss of body condition in pregnant animals leading to birth of offspring with low birth weights, fetal and neonatal losses, besides production losses in lactating animals. The consequences of trypanosomiasis are less severe in better-nourished animals but good nutrition does not by itself provide protection. Adequate energy, protein and vitamin nutrition enhances the ability of trypanosome-infected animals to withstand the adverse effects of infection (Pathak 2009). *T. vivax* is enzootic in wide areas of Sudan more than 2,000 km away from the known tsetse belt of the country. Previous outbreaks of bovine trypanosomiasis (BT) reported to be due to *T. congolense* were reported as far north as Kosti area along the White Nile, more than 1,000 km north of the Tsetse belts. The transmission of the disease is due to the enormous populations of *Tabanidae* and biting flies (Rahman 2005). *Trypanosoma* infection restricts the animal production and causes economic losses by the clinical signs of the infection such as restricted growth, abortion, anemia, treatment cost, and death of the affected animals. Hence, the objectives of this study were to estimate the prevalence of BT in Khartoum State, to identify the prevailing species of trypanosomes and to investigate the potential risk factors associated with the disease.

II. MATERIALS AND METHODS

a) Study area

Khartoum is the political capital of the Sudan. The state lies between longitudes 31.5°-34° east and latitude 15°-16° north in an area about 28.165 km². Geographically, Khartoum is divided into three

governorates (first governorate Khartoum (Jabal Awlia and Khartoum localities), second governorate Khartoum North (East Nile and Bahari localities) and third governorate Omdurman (Omdurman, Ombeda and Karari localities) (Khartoum state website (2012). www.krt.gov.sd). Diurnal and annual temperature variations are very wide. The highest mean maxima are between March and May, ranging from 35° to 43 °C and the lowest mean minima is between December and February ranging from 15° to 28 °C.

The population of animals in Khartoum state is 6300000 for birds, 513000 for sheep, 19000 of goats, 6585 camels and 240003 cattle. The number of cattle distributed in Khartoum state is 138067 in East Nile, 28016 in Bahri, 13578 in Ombeda, 13901 in Karari, 20455 in Omdurman, 20360 in Jabal Awlia and 5626 in Khartoum localities (Khartoum state census, Khartoum, Ministry of agriculture and animals' resources and irrigation, Khartoum state, Sudan, 2008).

b) Study design

A cross sectional survey was conducted from April to July 2012 to estimate the prevalence of BT, to identify the prevailing species of trypanosomes and to investigate potential risk factors associated with the disease.

c) Sampling Method

A multistage random sampling method was followed to select the study animals based on state, governorates (Khartoum, Khartoum North, and Omdurman), localities (Jabal Awlia, East Nile, Omdurman and Ombeda), and areas (Ombeda, Felstine, Almoilih, Aldekhinat, Edbabekir, Alozozab, Alkeriab and Suba). Finally, farms and animals were conveniently selected.

i. Sample size determination

The sample size was determined using the following formula (Thrusfield 2005):

$$n = (1.96)^2 \times P^{\wedge} \times Q^{\wedge} / L^2$$

Where:

n = the sample size.

(1.96)² = constant.

P[^] = expected prevalence of BT, which was 4.4% (Abdalla et al. 2008).

Q[^] = 1 - P[^]

L² = allowable error (5%).

n = (1.96)² × 0.044 × 0.956 / 0.0025 = 64 samples.

To increase the precision of the study; the sample size was multiplied by 4 (Thrusfield 2005), so the number of samples became 256 samples. Finally, the investigators were able to collect samples from 271 animals.

ii. Samples collection and laboratory tests

Blood samples were collected from each animal by puncture of the jugular vein using a sterile needle

(Abenga et al. 2004, Adam et al. 2011, Mogona et al. 2011, Batista et al. 2012). Blood was obtained in dry clean sterile heparinized tube 5 milliliters (containing EDTA (ethylene diamine tetra acetic acid, as anticoagulant) and was put in cold box with ice and transported to the laboratory (Central Veterinary Research Institute Laboratories, Soba, Khartoum) as soon as possible for diagnosis.

iii. Diagnostic techniques

For diagnosis, parasitological methods (Haematocrit Centrifugation Technique (HCT) and thin blood smear stained with Giemsa stain) was used to identify the parasite, also packed cell volume (PCV) of each animal was measured for anemia estimation.

iv. Investigation of potential risk factors

A pre-tested questionnaire was completed in an interview with the farms owners and/or workers to collect information data about selected potential risk factors on the occurrence of BT.

d) Statistical analysis

The overall prevalence was calculated based on positive results divided by the total number of animals tested. Potential risk factors (independent variables) and the laboratory test outcome (dependent variable) were analyzed using SPSS (version 16.0). The Chi-square test (χ^2) was used for univariate analysis; the significance level was set at $p \leq 0.25$. In order to control for confounding, risk factors found significant at $p \leq 0.25$ were entered to the multivariate analysis using logistic regression, the significance level of association was set at $p \leq 0.05$. The strength of association was measured by the odds ratio (OR) accompanied by the 95% confidence interval (CI) for OR.

III. RESULTS

a) Prevalence and trypanosome species

Out of the total 271 animals tested 13 were positive. The overall prevalence of BT in Khartoum state, Sudan was 4.8% (13/271). The prevalent trypanosome species in the study was *Trypanosoma vivax*.

b) Univariate analysis

The Chi-square test (χ^2) showed that there were 15 out of 33 potential risk factors statistically significantly associated with BT (P-value ≤ 0.25). These were: locality (p = 0.084), area (p = 0.000), sex (p = 0.219), body condition (p = 0.009), herd size (p = 0.094), treatment of sick animal (p = 0.003), presence of other species of animals in the farm (p = 0.000), farm hygiene (p = 0.148), veterinary care (p = 0.000), surgical operation or wound in the animal's body (p = 0.108), location of livestock market (p = 0.002), presence of insects in the farm (p = 0.108), species of insect found in the farm (p = 0.106), presence of ticks in the farm (p = 0.099) and farmer awareness about the disease (p = 0.065) (Table1).

c) *Multivariate analysis*

The multivariate analysis using the logistic regression showed that there were four potential risk factors statistically significantly associated with BT ($p \leq 0.05$). These were: treatment of sick animals ($p = 0.012$), presence of other animal species in the farm ($p = 0.003$), veterinary care ($p = 0.023$) and location of livestock market ($p = 0.004$).

IV. DISCUSSION

Trypanosomiasis is a major constraint that affects the health and productivity of livestock. The major consequences of infection include mortality, loss of body condition and abortion (Delafosse et al. 2006). Among all species of animals, the disease has been regarded as one of the most serious animal disease problem in Sudan. *Trypanosoma vivax* occurs in most parts of the country (Rahman 2005).

In our study, the prevalence and risk factors of BT in Khartoum state, Sudan was investigated. *T. vivax* was the only trypanosome species found during the study period. This is in agreement with previous studies (Tadesse et al. 2011, Rahman 2005, Abdalla et al. 2008, Batista et al. 2012, Delafosse et al. 2006) which reported that *T. vivax* constituted the majority of infection in the tsetse free zone.

The prevalence in our study was 4.8%. This result is not different from another study carried out in the Blue Nile area between Addamazin and Khartoum where the prevalence was 1% in the dry season and 6% in the rainy season (Rahman 2005), and in Sinnar, Sudan the prevalence was 4.4% (Abdalla et al. 2008). Another study carried out in Lake Chad showed a prevalence of 1.6% (Delafosse et al. 2006). Also, in another study in South Western Ethiopia the prevalence was 4.4% (Tadesse and Tsegaye 2010). However, the prevalence in our study was very low compared with other studies in Sudan and different countries which was 50.3% in Sinnar, Sudan where there is no tsetse fly in the area (Abdalla et al. 2005), 43% in Blue Nile State, Sudan (Salim et al. 2011), and 70% in Western Kenya (Thumbi et al. 2010). The difference in BT prevalence between our study and different studies may be due to different factors associated with the disease like: Khartoum state is out of the tsetse belt and the disease is transmitted mechanically, environmental factors which affect the vector, control strategy applied in the areas, variation on climatic conditions and dry season which affects the vectors breeding and low parasitaemia.

Distribution of the prevalence of BT in the localities showed significant association ($p = 0.084$). The higher prevalence in Ombeda might be attributed to the proximity of the area to livestock market.

A significant association between the disease and areas was observed ($p = 0.000$). This association could be due to the presence of irrigation canals which

extended in the area as well as good vegetation making a suitable environment for vector breeding.

This study showed a prevalence of 5.3% in females. There was a significant association in the univariate analysis between BT and sex ($p = 0.219$). This result is in agreement with a previous study (Mogona et al. 2011) which reported that a female is at risk two times higher than a male. Females have a higher prevalence than males, because females remain longer for production purposes, and also due to physiological stress like estrous cycle, pregnancy and lactation, so the disease has more chance to develop.

The significant association of body condition with the disease in this study ($p = 0.009$) agrees with other studies (Tadesse and Tsegaye 2010, Begna et al. 2011, Bitew et al. 2011, Mulaw et al. 2011) which found that the prevalence in poor body condition animals was significantly higher than good body condition animals ($p \leq 0.05$). This could be due to the chronic nature of the disease that resulted in anemia, poor body condition and emaciation.

There was a significant association between the disease and small herd size ($p = 0.094$). This could be attributed to the fact that most of < 30 groups, in this study, were located in areas with high density of insects, thus more fly attacks to the herd of small size than large size one.

The current study investigated the use of drugs for treatment of animals. There was significant association between the disease and treatment of sick animals ($p = 0.003$). The association of treatment of animals with the disease was also significant in the multivariate analysis ($p = 0.012$). These results agree with a previous study (Kidanemariam et al. 2002) which reported that chemotherapy and chemoprophylaxis is important against trypanosomiasis. Logically, treatment of sick animals restricts spread of the disease.

There was significant association between BT and the presence of other species of animals in the farm ($p = 0.000$). This risk factor was also significant in the multivariate analysis ($p = 0.003$). This result agrees with Mustafa (2004) who reported that sheep can act as a potential reservoir in mixed herd. Thus, presence of other species of animals in the farm may act as a source of the disease.

Also, the study investigated the farm hygiene as a risk factor possibly associated with the disease and there was a significant association between the disease and farm hygiene ($p = 0.148$). This could be attributed to the fact that poor farm hygiene provides a suitable environment for breeding of insects.

For veterinary care the study showed a significant association between BT and the availability of veterinary care in the univariate analysis ($p = 0.000$). Also, veterinary care had a significant association with BT ($p = 0.000$) in the multivariate analysis with a

protective effect. Logically, veterinary care plays a role in restricting the disease by intervention if any clinical signs appear.

Surgical operation and wounds in the animal's body were investigated and a significant association between BT and surgical operation or wounds in the animal's body was observed ($p = 0.108$). That is logical because surgical operation or wounds in the animal's body invite insects to have their blood meal.

There was a significant association between the disease and location of livestock market ($p = 0.002$). Also, location of livestock market had a significant association with the disease ($p = 0.004$) in the multivariate analysis. These findings are consistent with a previous study (Abdelkarim 1991) which reported that *Trypanosoma* infected animals in the livestock market can act as a source of infection to the neighboring farms. Logically, if there is one animal with the disease in the market all the area around may be at risk when the vector is present.

Also, the study investigated presence of insects in the farms. There was a significant association between the disease and presence of insects in the farm ($p = 0.108$). Furthermore, another related risk factor investigated was insects' species found in the farm. There was a significant association between the disease and insects' species found in the farm ($p = 0.106$). This result agrees with a previous study (Rahman 2005). These insect species may act as a mechanical vector for the disease.

Furthermore, presence of ticks in the farm was investigated in our study. There was a significant association between the disease and presence of ticks in the farm ($p = 0.099$). Ticks may predispose animals to infection with BT by infection with other tick-borne diseases which reduce the animal immunity.

Farmer awareness about the disease was investigated as a risk factor possibly associated with the disease. There was a significant association between the disease and farmer awareness ($p = 0.065$). Logically, aware farmer could apply control methods to control the disease and vectors.

In this study, the univariate analysis showed that 15 risk factors were statistically significantly associated with BT ($p \leq 0.25$). However, in the multivariate analysis, there were only four risk factors that were found statistically significantly associated with BT ($p \leq 0.05$), a result which indicates a presence of confounding between these risk factors.

V. CONCLUSION

The current study confirmed that BT is widely distributed in Khartoum state farms with an overall prevalence of 4.8%. Furthermore, the study indicated that *T. vivax* is the predominant species implicated in BT in Khartoum state.

The presence of various biting flies and the absence of tsetse flies in this investigation indicated that BT in Khartoum state is caused by mechanical transmission mediated by these biting flies.

The results of the multivariate analysis showed that four risk factors had statistically significant associations with BT ($p \leq 0.05$). These risk factors were: treatment of sick animals ($p = 0.012$), presence of other species of animals in the farm ($p = 0.003$), veterinary care ($p = 0.023$) and location of livestock market ($p = 0.004$). These risk factors should be considered as predictors for the disease and should be taken into account when planning for control programs of the disease.

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Table 1: Univariate analysis of association between bovine trypanosomiasis and potential risk factors in 271 cattle in Khartoum state, Sudan, using the Chi-square (χ^2) test

Risk factors	No. tested	No. +ve (%)	d. f.	χ^2	p-value
1. Locality					
Ombeda	22	3 (13.6)	3	6.648	0.084
Omdurman	22	0			
JabalAwlia	34	0			
East Nile	193	10 (5.2)			
2. Area					
Felstine	22	3 (13.6)	7	27.293	0.000
Almoilh	22	0			
Aldkhinate	34	0			
Alozozab	42	1 (2.4)			
Eidbabeker	67	1 (1.5)			
Sudan university farm	8	0			
Alkeriab	45	8 (17.8)			
Soba	31	0			
3. Sex					
Male	27	0	1	1.511	0.219
Female	244	13 (5.3)			
4. Body condition					
Poor	119	11 (9.2)	2	9.327	0.009
Good	122	2 (1.6)			
Very good	30	0			
5. Herd size (# of animals)					
<30	195	12(6.2)	1	2.803	0.094
≥30	76	1(1.3)			
6. Treatment of sick animals					
Not treated	72	8 (11.1)	1	8.559	0.003
Treated	199	5 (2.5)			

7. Presence of other animal species in the farm					
Present	67	11 (16.4)	1	26.319	0.000
Not present	204	2 (1.0)			
8. Farm hygiene					
Good	115	3 (2.6)	1	2.095	0.148
Poor	156	10 (6.4)			
9. Veterinary care					
Not available	121	12 (9.9)	1	12.550	0.000
Available	150	1 (0.7)			
10. Surgical operation or wound in animal's body					
Yes	5	1 (20.0)	1	2.578	0.108
No	266	12 (4.5)			
11. Location of livestock market					
Close to the farm	67	8 (11.9)	1	9.944	0.002
Away from the farm	204	5 (2.5)			
12. Presence of insects in the farm					
Present	5	1 (20.0)	1	2.578	0.108
Not present	266	12 (4.5)			
13. Insects species found in the farm					
<i>Tabanus</i>	136	9 (6.6)			
<i>Tabanus</i> + <i>Stomoxys</i>	16	1 (6.2)	3	6.122	0.106
<i>Stomoxys</i>	17	2 (11.8)			
Other species of insects	102	1 (1.0)			
14. Presence of ticks in the farm					
Not present	45	0	1	2.79	0.099
Present	226	13 (5.8)			
15. Farmer awareness about the disease					
Not aware	217	13 (6.0)	1	3.398	0.065
Aware	54	0			

Table 2: Multivariate analysis of association between bovine trypanosomiasis and potential risk factors in 271 cattle in Khartoum state, Sudan, using logistic regression

Risk factors	No. tested	No. +ve (%)	OR	95% CI for OR	p-value
1. Treatment of sick animals					
Treated	199	5 (2.5)	ref		
Not treated	72	8 (11.1)	5.536	1.4-21.06	0.012
2. Presence of other animal species in the farm					
Not present	204	2 (1.0)	ref		
Present	67	11(16.4)	.076	0.014-0.418	0.003
3. Veterinary care					
Available	150	1 (0.7)	ref		
Not available	121	12 (9.9)	.092	0.012 -0.724	0.023
4. Location of livestock market					
Away from the farm	204	5 (2.5)	ref		
Close to the farm	67	8 (11.9)	.118	0.028-0.497	0.004

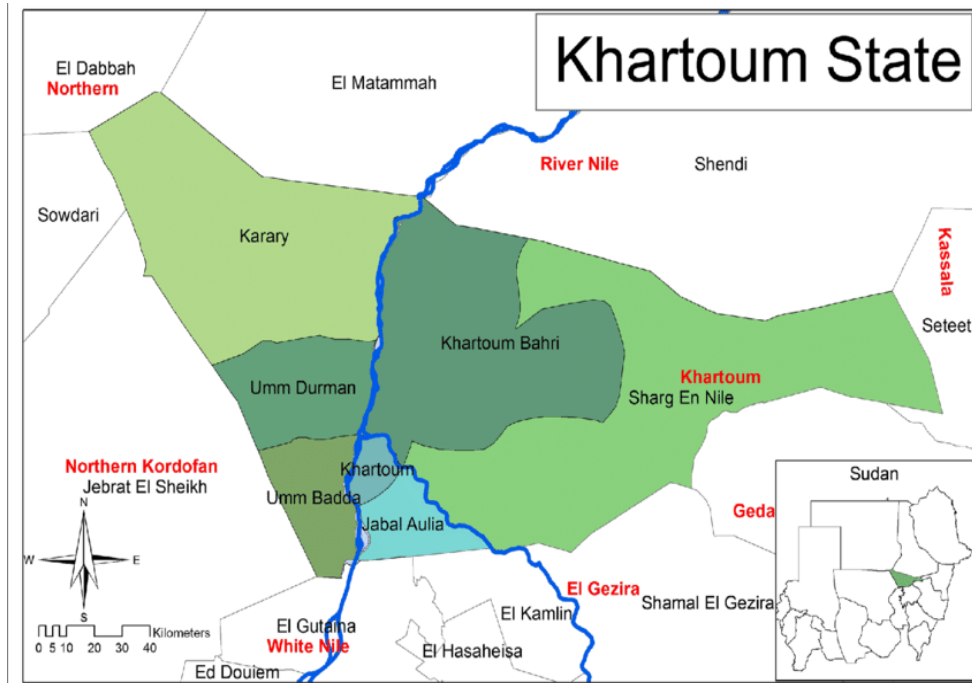


Figure 1: Map of Sudan and Khartoum state showing the study areas for bovine trypanosomiasis