CZU: 576.895.772(6)

IDENTIFICATION OF TRYPANOSOMES IN TSETSE FLIES IN SAMAYA AND DAMAKANYA, WESTERN GUINEA

SEYDOU SYLLA[a], YOUSSOUF SIDIMÉ[a], Valentina PETCU[b], LANCEÏ KABA[a], YACOUBA KONATÉ[a], Mouhamed KEYRA[a], Saran CAMARA[a]

[a]Institut supérieur des Sciences et de Médecine Vétérinaire (ISSMV) of Dalaba, 09 Dalaba, Guinea.

[b]The State Agrarian University of Moldova, Chisinau, R. Moldova

Abstract: Trypanosomes are parasitic protozoans that live in the blood of a great variety of species including human transmitted by the tsetse flies. Indentifying trypanosomes in tsetse flies provides a measure of disease risk and a basis for the design of control approaches. The objective of the present study was to identify the species of the trypanosomes in the tsetse flies. The trypanosomes had been indentified in this study using the key previously described by Enyaru 2010.

Key words: Trypanosome, tsetse fly, density.

INTRODUCTION

Trypanosomes are bloodstream parasites of the great variety of vertebrate species and generally are transmitted by an intermediated host (Habila, et al., 2012). The species of *Trypanosomavivax* (*T. vivax*), *Trypanosomacongolense* (*T. congolense*) and *Trypanosomabruceibrucei* (*T. b. brucei*) are mainly responsible for animal trypanomosis in western Africa (Ogbaje et al., 2015). This parasite can also affect the humans (Fernandez et al., 2009) and the trypanosomes are mainly disseminated by the Tabanid species (*Tabanus spp., Chrysops spp. and Hematopota spp.*) (Otto et al., 2010). In Sub-Saharan Africa, Tsetse flies inhabit 8.7 million km^2, known as the tsetse belt (PAAT, 2007). In the areas where the prevalence of the tsetse flies is higher, agricultural output is suboptimal because of the risk of animal trypanomosis (Eyford, 2011). The vertebrate blood is exclusively feed for adult tsetse flies. Thus, all species of tsetse flies are probably capable of transmitting pathogenic trypanosomoses during the process of feeding. The wild animals had been always implicated as important in the epidemiology of trypanosomoses but without direct evidence (Magona, 2008).

In Africa, the tsetse fly occurs in 37 countries and the entire tropical region is virtually affected by trypanosomiasis (Enyaru, 2010). Furthermore, the cases of trypasomiasis had been estimated at 300 000 with an impact of 1.59 million disability adjusted life years (WHO, 2006).

Identifying trypanosomes in livestock and/or in tsetse flies accurately provides a measure of disease risk, and a basis for the design of control approaches that resulted from knowledge that domestic animals are reservoirs (Magona, 2008). The monitoring of trypanosome in tsetse flies population provides information on the risk of infection, and is important in evaluating the success of control programs that attempt to eliminate parasites from circulation (Enyaru, 2010).

The aim of the present study is to investigate the rate of trypanosome in tsetse flies in the rural zones of Samaya and Damakanya in province of Kindia.

MATERIEL AND METHOD

Study area

Damakanya and Samaya were located in the western of Guinean between 9°30′ and 9°52′ north latitude, and 12°50′ and 13°4″ west longitude, respectively; with an altitude mean of 458,13 m. The climate of investigated areas is tropical type and the vegetation is mainly forest, characterized by a rainfall of 1,976 mm/year and the annual average temperature and humidity of around 20 and 62 % respectively. Both localities have several rivers: samou, kolente, kilissi, konkoure, garafiri, wantamba and myeriya for Damakanya, and fossekha, wantamba, dougna, kondekhoure, walanyi and kafossy for Samaya. These rivers are the area of tsetse flies.

Study design

The cross-sectional study was carried out in investigation using the structural questionnaire survey where the questions on the knowledge of breeders on the trypanosomose and tsetse flies, transmission cycle of trypanosome, enzootic and zoonosis were discussed. This investigation was undertaken from May to August 2012. The sites of development in the tsetse fly and morphology were used to identify the trypanosomes. The existence and density of tsetse fly were the factors which determinate the investigation site.

Capture, dissection and identification of trypanosome in the tsetse flies

The biconcave traps were used in the capture of the tsetse flies. The identification of trypanosome in the tsetse flies had been based on the site of development, differences in morphology and arrangements in the various tsetse fly organs that were parasitized as described in the early study (Enyaru, 2010). Briefly, after careful dissection using light microscopy to examine tsetse tissues, researchers designated trypanosomes that were found only in the mouthparts as belong to sub-genus *Duttonella* (with *T. vivax*as the type species), and those found in both mouthparts and midgut as members of *Nannomonas* (*T. congolense*), whereas those found also in the salivary glands were listed as Trypanozoon (*T. brucei*). The trypanosomes are found only in the midgut are classified as immature. When the trypanosomes were found in the midgut or proboscis, the difference is based on morphology analysis. The apparent density had been obtained using the following formula:

Where:*AD*is the apparent density, *FC* number of the fly captured and *NT* number of the trap.

Statistical analysis was performed using the SPSS statistical software (version 13.0, SPSS Inc., Chicago,Illinois, USA). Statistical significance was defined as a *P value* <0.05. The samples tested positive to CFT were defined as Seropositivity. All statistical analyses were based on the CFT-positive results.

RESULTS AND DISCUSSION

The interview of the farmers shows that 90% are breeders and cultivator, and 63.45% are men. In both localities 85% the farmers use animal traction, the N'dama and Zebus are the bred animals in those zones. Among the breeders of Damakanya and Samaya, 73.43% knew the trypanosomose and 87.34% of the farmers know well the tsetse flies but knew little about the implication of the tsetse fly in the transmission of the trypanosomose. Only 32.64% knew that trypanosomose is a zoonosis. Not any dispositions exist to fight against tsetse flies in these localities.

Flies Density and infected flies

As show in table 1, Sixty-five traps were used including 39 (60%) of the trap for Damakanyaand 26 (40%) for Samaya. Of the Forty-nine tsetse flies captured, 30 (60.67%) are of Damakanya and 19 (38.77%) are of Samaya. The density mean of both localities are 0.76 and 0.73 for Damakanya and samaya respectively. The *Glossinapalpalis*is only fly species found in this study. Of the 49 flies captured and dissected, 15 (30.61%) were infected, including 11 (22.44%) and 4 (8.16%) for Damakanya and Samaya respectively (table 2).

Table 1. Capture, dissection and identification of trypanosome in the tsetse flies

Localities		**n^t**	**n^f**	**Apparent density**	**Species of the flies**
Damakanya	Damakania village	10	4	0.4	*G. palpalis*
	Foulaya city	5	7	1.4	
	Samorya	6	5	0.83	
	Barenfory	5	6	1.2	
	Khariakory	4	2	0.5	
	Madinalaya	9	6	0.66	
Samaya	Samaya village	5	2	0.4	
	Komoya	4	9	2.25	
	Condeya	8	1	0.12	
	Kaporo	3	2	0.66	
	Walia	6	5	0.83	

n^t, number of the trap; n^f, number of the flies.

Statistically, the density of the tsetse flies in Damakanya (0.76 IC 95% 0.61-0.89) was higher than that found in Samaya (0.73 IC 95% 0.52-0.88), but the different is not statistically significant *P=0.98*. Howeiver, the prevalence of the infected flies in Damakanya (36.67% IC 95% 19.93-56.14) was largely higher compared to that found in Samaya (21.05% IC 95% 6.05-45.57) and the difference was statistically significant *P<0.04* (tableau 3).

Table 2. Prevalence of infection in tsetse flies

Localities		n^f	Flies infected	Prevalence (%)	95% IC	
					Low	Upper
Damakanya	Damakania village	4	2	50	6.76	93.24
	Foulaya city	7	3	42.85	9.90	81.59
	Samorya	5	1	20	0.50	71.64
	Barenfory	6	5	83.33	35.88	99.58
	Khariakory	2	0	0	0.00	84.10
	Madinalaya	6	0	0	0.00	45.93
Samaya	Samaya village	2	0	0	0.00	99.58
	Komoya	9	3	33.33	7.49	70.07
	Condeya	1	0	0	0.00	97.50
	Kaporo	2	1	50	1.26	98.74
	Walia	5	0	0	0.00	52.18

*T. congolense and T. vivax*are the trypanosomes species found with the prevalence of 53.33% and 33.33% for *T. congolense* and *T. vivax* respectively (table 3). Furthermore, 13.33% of the flies had been poly parasitized by *T. congolense and T. vivax.* Statistically, the prevalence of tsetse flies infected with *T. congolense*(53.33% 95%IC 26.59-78.73), *T. vivax* (33.33% 95%IC 11.82-61.62), and *T. congolense* and *T. vivax* (13.33% 95% IC 1.66-40.46); the differences between the species are statically signifiant*P<0.05* (table 4).

Table 3. Density of the flies and prevalence of infection in flies

Localities	n^t	n^f	AD	95% IC		*P-value*	infected Flies	Prevalence (%)	95% IC		*P-value*
				low	upper				low	upper	
Damakanya	39	30	0.76	0.61	0.89	0.98	11	36.67	19.93	56.14	0.04
Samaya	26	19	0.73	0.52	0.88		4	21.05	6.05	45.57	

AD, apparent density.

Table 4.Prevalence of the infection by trypanosomes species

Trypanosome species	n^i	Prevalence (%)	95% IC		*P-value*
			Low	Upper	
T. congolense	8	53.33	26.59	78.73	6.14×10^{-6}
T. vivax	5	33.33	11.82	61.62	
T. congolense and vivax	2	13.33	1.66	40.46	
Total	15	-	-	-	-

n^i, number of the infected flies

In this study, the density of the tsetse flies and the trypanosomal infections in tsetse flies had been determinate. The trypanosomes species in the tsetse flies have been identified using the key previously described by Enyaru (Enyaru, 2010). The trypanosomose is a devastating disease affecting livestock in large parts of Africa (Simukoko, 2011). African trypanosomose is a zoonosis caused by of the flagellated protozoan species *Trypanosoma*. This zoonosis is estimated at 300,000-500,000 cases of which 60 million people are at risk in 37 countries covering 40% of African continent approximately (Akso and Rio, 2005). In Guinea, an early investigation on human trypanosomose reported that 17 people were serologically tested positive of which four have developed human African trypanosomose (Ilboudo, 2011). The challenge of the trypanosomose is determined by the density of tsetse flies, the trypanosomal infections prevalence in tsetse flies and proportion of tsetse flies meal from host species (Kubi, 2007). In this study, we found a mean density of 0.75 for both localities. The both localities have several rives. The strong correlation between the serological status of the cattle and the proximity to the river or main tributaries (Sow, 2013),

The infection mean in the tsetse flies in this study was 30.61% which is largely higher than that reported in Zambia, with the monthly average proportion of infected tsetse flies of 9% (Simukoko, 2011). The density of the tsetse flies varied according to the seasons (Van den Bossche and De Deken, 2002). The present study was carried from May to August. The risk of infection with trypanosomes was constant during most of the year but the risk of infection increased significantly in the beginning of the rainy season (Sinyangwe, 2004).

The pathogenic of trypanosomes have a complex developmental cycle that involves both mammalian host and tsetse vector (Eyford, 2011). Control the trypanosomose in cattle is largely dependent to the vector control (Delespaux, 2010). In thisinvestigation, *G. palpalis* is the only specie founded. Furthermore, the *palpalis* group had been show as effective vectors in the transmission of the trypanosome species (Moloo and Kutuza, 1988). Thus, Damakanya and Samaya are the risk area for the trypanosomose in animal and human. In other hand, *T. congolense* and *T. vivax*have been the species found in this study, it had been reported that *T. vivax* and *T. congolense* are infect large variety of wild and domestic animals (de Araujo, 2011; Pimentel, 2012).

In Guinea despite the presence of trypanosomose control center, the data about the trypanosomose in cattle and human population are not complete on the national territory. However, the African human trypanosomose had been diagnosed in mangrove focus of Forecariah with 17 cases including 4 cases in sleeping sickness. The investigation should be extended to other tsetse flies endemic area.

RÉFÉRENCES

1. HABILA, Nathan, INUWA, Mairo H., AIMOLA, Idowu A., UDEH, Michael U., HARUNA, Emmanuel. Pathogenic mechanisms of Trypanosomaevansi infections. In: Research in Veterinary Science. 2012, vol. 93, pp. 13-17.
2. OGBAJE, Christopher Igoche, LAWAL, Idris Alao, AJANUSI, Ologunja Joseph. Sensitivity of Nigerian field isolates of Trypanosomavivax and Trypanosomacongolense to commonly available trypanocides. In: Asian Pac. J. Trop Dis. 2015, nr 5(3), pp. 214-218.
3. FERNANDEZ, D., GONZALEZ, E., Z-BARADAT, B., ELEIZALDE, M., GONZALEZ-MARCANO, E., PERRONE, T., MENDOZA, M., Trypanosomaevansi: a comparison of PCR and parasitological diagnostic tests in experimentally infected mice. In: Experimental Parasitology. 2009, vol. 121, pp. 1-7.
4. OTTO, M.A., DA SILVA, A.S., GRESSLER, L.T., FARRET, M.H., TAVARES, K.C.S., ZANETTE, R.A., MILETTI, L.C., MONTEIRO, S.G. Susceptibility of Trypanosomaevansi to human blood and plasma in infected mice. In: Veterinary Parasitology. 2010, vol. 168, pp. 1-4.
5. Programme Against African Trypanosomiasis (PAAT). Tsetse and Trypanosomiasis Information. PAAT, 2007, vol. 30, pt 2.
6. EYFORD, Brett A., SAKURAI, Tatsuya, SMITH, Derek, LOVELESS, Bianca, HERTZ-FOWLER, Christiane, DONELSON, John E., INOUE, Noboru, PEARSON, Terry W. Differential protein expression throughout the life cycle of *Trypanosomacongolense*, a major parasite of cattle in Africa. In: Molecular & Biochemical Parasitology. 2011, vol. 177, pp. 116-125.
7. MAGONA, J.W. et al. Acute haemorrhagic syndrome of bovine trypanosomosis in Uganda. In: Acta Trop. 2008, vol. 107, pp. 186-191.
8. ENYARU, J. C., OUMA, J. O., MALELE, I.I., MATOVU, E. and MASIGA, D.K. Landmarks in the evolution of technologies for identifying trypanosomes in tsetse flies. In: Trends in Parasitology. 2010, vol. 26, pp. 388-394.
9. WHO. Human African trypanosomiasis (sleeping sickness): epidemiological update. In: Wkly. Epidemiol. Rec. 2006, vol. 82, pp. 71-80.
10. SIMUKOKO, H., MARCOTTY, T., VERCRUYSSE, J., VAN DEN BOSSCHE, P. Bovine trypanosomiasis risk in an endemic area on the eastern plateau of Zambia. In: Research in Veterinary Science. 2011, vol. 90, pp. 51-54.
11. KUBI, C., BILLIOUW, M., VAN DEN BOSSCHE, P. Age prevalence of trypanosomal infections in female Glossinamorsitansmorsitans (Diptera: Glossinidae) on the plateau area of eastern Zambia. In: Onderstepoort Journal of Veteterinary Research. 2007, vol. 74, pp. 223-229.
12. AKSOY, S., RIO, V.M.R. Interactions among multiple genomes: tsetse, its symbionts and trypanosomes. In: Insect Biochemistry and Molecular Biology. 2005, vol. 35, pp. 691-698.
13. ILBOUDO, Hamidou, JAMONNEAU, Vincent, CAMARA, Mamadou et al. Diversity of response to Trypanosomabruceigambiense infections in the Forecariah mangrove focus (Guinea): perspectives for a better control of sleeping sickness. In: Microbes and Infection. 2011, vol. 13, pp. 943-952.
14. VAN DEN BOSSCHE, P., De DEKEN, R., Seasonal variations in the distribution and abundance of the tsetse fly, Glossinamorsitansmorsitans in eastern Zambia. In: Medical and Veterinary Entomology. 2002, vol. 16, pp. 170-176.
15. SINYANGWE, L., DELESPAUX, V., BRANDT, J., GEERTS, S., MUBANGA, J., MACHILA, N., HOLMES, P.H., EISLER, M.C. Trypanocidal drug resistance in Eastern Province of Zambia. In: Veterinary Parasitology. 2004, vol. 119, pp. 125-135.
16. Delespaux V, Vitouley HS, Marcotty T, Speybroeck N, Berkvens D, Roy K, et al. Chemosensitization of

Trypanosomacongolense strains resistant to isometamidium chloride by tetracycline and fluoroquinolone. PLoSNegl Trop Dis. (2010) 4, p. 828.

17. (Eyford A. Brett, Sakurai Tatsuya, Smith Derek, Loveless Bianca, Hertz-Fowler Christiane, Donelson E. John, Inoue Noboru, Pearson W. Terry, Differential protein expression throughout the life cycle of *Trypanosomacongolense*, a major parasite of cattle in Africa. Molecular & Biochemical Parasitology, (2011) 177, 116-125).

18. MOLOO, S.K., KUTUZA, S.B. Comparative study on the infection rates of different laboratory strains of Glossina species by Trypanosomacongolense. In: Medical and Veterinary Entomology. 1988, nr 2, pp. 253-257.
19. De ARAUJO, Melo S., BARROS, A.C., COSTA, F.B., de CARVALHO, Neta A.V., de CANDANEDO, Guerra R.M., ABREU-SILVA, A.L. Bovine trypanosomiasis an emerging disease in Maranh ã o State-Brazil. In: Vector Borne Zoonotic Dis. 2011, vol. 11, pp. 853-856.
20. PIMENTEL, D. de S., RAMOS, C.A., RAMOS, R.A., de ARAÚJO, F.R., BORBA, M.L., FAUSTINO, M.A., et al. First report and molecular characterization of Trypanosomavivax in cattle from state of Pernambuco, Brazil. In: Vet. Parasitol. 2012, vol. 185, pp. 286-289.