Current Research Journal of Biological Sciences 6(6): 222-228, 2014

DOI:10.19026/cribs.6.5197

ISSN: 2041-076X, e-ISSN: 2041-0778 © 2014 Maxwell Scientific Publication Corp.

Submitted: April 08, 2014 Accepted: June 18, 2014 Published: November 20, 2014

Research Article

Factors Influencing Seasonal Abundance of *Glossina fuscipes fuscipes* (*Glossina: Glossinidae*) in Kajo-Keji County, South Sudan

¹Yatta S. Lukaw, ²Mubarak M. Abdelrahman, ³Yassir O. Mohammed, ⁴Erneo B. Ochi and ²Intisar E. Elrayah

¹Department of Basic Sciences, College of Natural Resources and Environmental Studies, University of Juba, P.O. Box 82, Juba, South Sudan

²Tropical Medicine Research Institute, P.O. Box 1304,

³Veterinary Research Institute, P.O. Box 8067, Khartoum, Sudan

⁴National Ministry of Animal Resources and Fisheries, Directorate of Research and Development, P.O. Box 126, Juba, South Sudan

Abstract: A 2-year tsetse field survey was conducted in Kajo-keji County South Sudan to determine factors influencing the seasonal abundance of Glossina fuscipes fuscipes. A total of 13,323 tsetse flies of both sexes were collected in the dry and wet seasons. Dry season catches were significantly higher than those of the wet season (DF = 10, t (2011) = 3.477, P (2011) = 0.006; DF = 10, t (2012) = 7.943, P (2012) = 0.0001). The number of male flies was significantly higher (DF = 15, t (2011) = 4.260, P (2011) = 0.0001; DF = 15, t (2012) = 5.492, P (2012 = 0.000) than the female ones during the study period. The mean apparent density of the flies/trap/day in the dry season was significantly higher (DF = 92, t (2011) = 2.147, P (2011) = 0.003; DF = 92, t (2012) = 3.764 and P (2012) = 0.0003. Average infection rate revealed 4.9% and 9.0 in the dry and wet seasons, respectively. An overall infection rate was 4.9% which varied significantly with the season ($X^2 = 602$, p<0.000). Positive associations were observed for atmospheric temperature, rain fall and relative humidity with monthly fly apparent density at R = 0.72, 0.71 and 0.82, respectively in 2011. However, in 2012 rainfall and relative humidity showed significantly negative association with the monthly fly apparent density at R = -0.85, P = 0.000 and R = -0.55, P = 0.031, respectively, whereas atmospheric temperature showed no statistical significant association at p<0.05 with the monthly fly density (R = 0.311, P = 0.163). Atmospheric temperature, rainfall and relative humidity seem to be influencing the seasonal abundance of G. f. fuscipes in the study area. These ecological factors could be used in developing tsetse control strategies.

Keywords: Ecological factors, *Glossina fuscipes fuscipes*, infection rate, seasonal abundance, South Sudan, tsetse fly apparent density

INTRODUCTION

Glossina fuscipes fuscipes is one of the seven species and subspecies of tsetse flies infesting both animals and humans in South Sudan. Moreover, evidence has shown that *G. f. fuscipes* is the sole vector of *Trypanosoma brucei gambiense*, the causative agent of Human African Trypanosomiasis (HAT) or sleeping sickness in South Sudan (Snow, 1983). Tsetse flies have been associated with the transmission of African trypanosomes since a century ago (Aksoy *et al.*, 2003). They infest an approximate total area of 10 million km² in sub-Saharan Africa including 250,000 km² of mostly fertile agricultural land in South Sudan, exposing a population of 65 million people at risk of infection in 38 African countries (PATTEC, 2012).

The distribution and prevalence of tsetse flies are most influenced by spatial factors including climate, vegetation and land utilization (Rogers *et al.*, 1996). It has been found that the tsetse fly apparent density (mean catch/trap/day+S.E) and infection rate are the main entomological factors related to transmission and risk (WHO, 1986). Nevertheless, the knowledge of the population dynamics is imperative for vector control strategies (Hay *et al.*, 1998). Consequently, monitoring the vector apparent density and infection rate are of paramount importance to identify and determine risky area in order to develop vector control strategy for effective control of the disease (Cano *et al.*, 2007).

This study describes and discusses some ecological factors that influence the seasonal abundance, apparent density, male/female ratio and infection rate of *G. f. fuscipes* in Kajo-Keji County (KKC), South Sudan.

Corresponding Author: Yatta S. Lukaw, Department of Basic Sciences, College of Natural Resources and Environmental Studies, University of Juba, P.O. Box 82, Juba, South Sudan

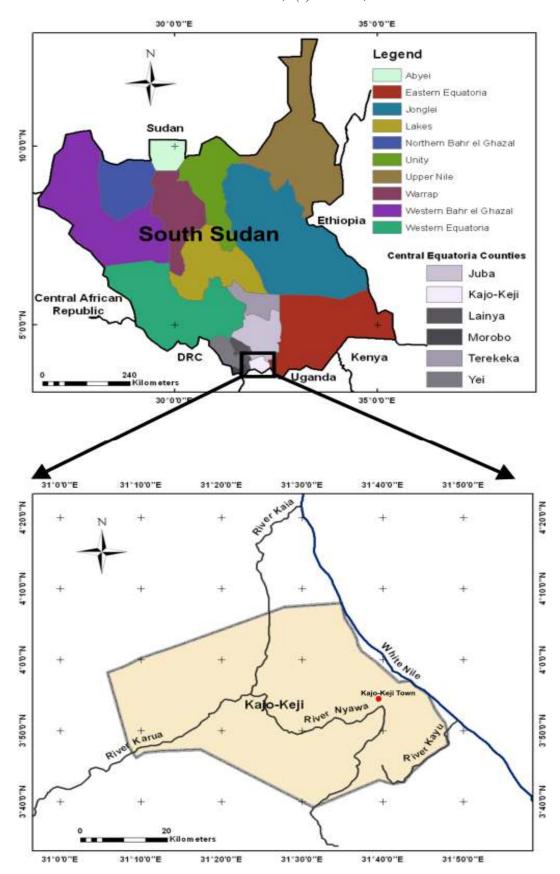


Fig. 1: Map of Kajo Keji County, Central Equatoria State, South Sudan

MATERIALS AND METHODS

The study area: This study was conducted as from January 2011 to December 2012 in KKC, Central Equatoria State (CES), South Sudan. KKC is located between geographical latitudes 3.67203- 4.13238°N and longitudes 31.1004 -31.8172°E. The County covers an area of approximately 113,000 km² bordering Uganda in the South, Yei River County in the West, Juba County in the North and the River Nile in the East (Fig. 1). KKC is an area of the tropical rainforest with moderate soil fertility and the climate is marked by minimal variations in seasonal temperatures. The annual rainfall ranges between 1,200 and 2,000 mm for about 8 months from March to late October. The dry season starts from late November to early March.

Entomological survey: Sampling of tsetse flies was carried out once every week from 8:00 a.m to 4:00 p.m h for 24 months during the wet and the dry seasons. Unbaited biconical traps were deployed in seven sites along the banks of eight streams 150 m apart and 5 m distant from the streams. Caught flies were stored in cool boxes, differentiated into males and females, counted and identified (Brunhes *et al.*, 1998). Nonteneral *G. f. fuscipes* flies were dissected and examined under microscope to check for trypanosomes (Clloyd and Johnson, 1924). Fly Apparent Density (AD) was estimated as described by Dede *et al.* (2005) and the infection rate (IR) was determined as follows:

The Infection Rate (IR) was determined as $\frac{\text{Number of the infected flies}}{\text{Total number of dissected flies}} = \times 100$

Data management and statistical analysis: Meteorological data for KKC on rainfall, atmospheric temperature and Relative Humidity (RH%) for the two-year sampling period were obtained from Juba Meteorological Station. Graphs drawn and tables depicted using Microsoft Excel 2010 for Windows. Comparison of monthly AD of *G. f. fuscipes* between the dry and the wet seasons was made.

Male/female catches at the streams of four Payams (Districts) of Lire, Kangapo I, Kangapo II and Liwolo of the KKC were analyzed using Student t-test. Seasonal variation in *Trypanosoma* infection rate in *G. f. fuscipes* was assessed using χ^2 -statistics and the relation between the fly AD and the ecological abiotic factors was analyzed using Spearman (R) Correlation Matrix. All the statistical analyses were made using SPSS 16.00 statistical package compatible to Microsoft Windows.

RESULTS

G. f. fuscipes population: In four out of the five Payams of KKC, CES a total of 6,509 flies were caught in the year 2011, of which 57 and 43% were male and female flies at the ratio of 1.3: 1, respectively.

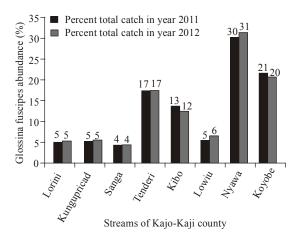


Fig. 2: Glossina fuscipes fuscipes abundance % in the streams of Kajo-Kaji County, Central Equatoria State South Sudan

However, in the second year, 2012, a total of 6,814 flies were caught, of which 60 % and 40% were male and female flies at the ratio of 1.5:1, respectively (Table 1).

G.f. fuscipes abundance in eight streams of KKC: Nyawa stream had the highest percentage of G. f. fuscipes where 31% of the fly caught in the year 2012 and 30% in 2011; Koyobe, 21% in the year 2011 and 20% in 2012; Tenderi, 17% in both years; Kibo, 13% in the year 2011 and 12% in 2012 and Lowiu, 5% in 2011 and 6% in 2012. However, Lorini and Kungupricad streams had the same proportion of 5% in both years whereas the smallest proportion (4%) of the fly was caught in Sanga stream in both years (Fig. 2).

Seasonal variations in *Glossina fuscipes fuscipes* **abundance:** The abundance *of* G. f. f *uscipes* in the dry and wet seasons showed that the dry season fly catches were significantly higher (p<0.01) than the wet season catches in a 2-year period in the study area. In the year 2011 the difference was at t-value = 5.224, DF = 10, P = 0.0004 compared to the year 2012 which was at t-Value = 5.227; DF = 10 and P = 0.0004. The AD of G. f. f *uscipes* in the dry season was significantly higher than the wet season (p<0.05). The significant differences were read at DF = 92, t-Value = 2.147, p<0.03; DF = 92, t-Value = 3.764, p<0.0003 in 2011 and 2012, respectively (Table 2).

Seasonal variations in *Glossina fuscipes fuscipes* total catches during the study period: The total catches of G. f. fuscipes in the dry season were significantly higher (p<0.05) than the wet season catches in both years. The significant differences were read at DF = 10, t = 3.477 and P = 0.006 in the year 2011; DF = 10, t = 7.943 and p<0.0001 in 2012 (Table 3).

Table 1: Glossina fuscipes fuscipes catches composition in Kajo-Keji County, Central Equatoria State South Sudan

Year	M	F	Total	M (%)	F (%)	M : F
2011	3.698	2.811	6.509	57	43	1.3:1
2012	4.085	2.729	6.814	60	40	1.5:1
Total	7 783	5 540	13 323			

M: male: F: female

Table 2: Seasonal variations in Glossina fuscipes fuscipes monthly apparent density during the study period in Kajo-Kaji County South Sudan				
Year 2011	DF	t-value	p-value	
Dry-wet seasons	92	2.147	0.03*	
Year 2012	DF	t-value	p-value	
D	02	2.764	0.0002**	

^{*:} Significant (p<0.05); **: highly significant (p<0.01); NS: Non significant (p>0.05) one-tailed P level

Table 3: Comparison (un paired t-Test) of the total catches of Glossina fuscipes fuscipes between dry and wet season in Kajo-Keji County, South Sudan

Year	DF	t-value	p-value
2011 (Dry wet season)	10	3.477	0.006**
2012 (Dry-wet season)	10	7.943	<0.0001**

^{*:} Significant (p<0.05); **: highly significant (p<0.01); NS: Non significant (p>0.05) one-tailed P level

Table 4: Comparison (paired t-Test) between male and female catches of Glossina fuscipes fuscipes in the years 2011 and 2012 in Kajo-Keji County, South Sudan

Year/season	DF	t-value	Significant (2-tailed)
2011 (Dry-wet)	15	4.26	0.001
2012 (Dry-wet)	15	5.492	0.000

^{*:} Significant (p<0.05); **: highly significant (p<0.01); NS: Non Significant (p>0.05) one-tailed P level

Table 5: Assessment of seasonal variations in Trypanosoma infection rate in Glossina fuscipes fuscipes in Kajo-Keji County South Sudan

Payam	Infection rate (IR %)			
	Dry season	Wet season	χ2	Probability-P
Lire	0% (0/107)	0% (0/69)	162	p<0.000
Kangapo (I)	0% (0/59)	0% (0/69)	124	p<0.000
Kangapo (II)	7 (12/168)	10 (23/205)	280	p<0.000
Liwolo	5 (3/57)	11 (2/16)	52	p<0.000
K.K.C.	4.9 (15/391)	9 (25/389)	602	p<0.000
Over all IR%	4.9	,		1

^{*:} Significant (p<0.05); **: highly significant (p<0.01); NS: Non significant (p>0.05) two-tailed P level

Table 6: Correlation of Glossina fuscipes fuscipes apparent density with some ecological factors

	Year 2011 G. f. fuscipes AD		Year 2012 G. f. fuscipes AD		
Ecological factors	Spearman's R	p-value (one-tailed)	Spearman's R	p-value (one-tailed)	
Temperature (°C)	R = 0.72	P = 0.004**	R = 0.311	P = 0.163N	
Rainfall (mm)	R = 0.71	P = 0.005**	R = 0.55	P = 0.031*	
Relative Humidity (RH %)	R = 0.82	P = 0.001**	R = 0.85	P = 0.00**	

^{*:} Significant (p<0.05); **: highly significant (p<0.01); NS: Non significant (p>0.05) one-tailed P level

Seasonal variations in male and female fly catches during the study period in Kajo-Keji county: Variations in the number of male and female flies caught in the dry and wet seasons in KKC during the study period showed that the number of male fly catches was significantly higher (p<0.05) than that of female fly catches (Table 4).

Correlation of Glossina fuscipes fuscipes apparent density/trap/day with some ecological factors: It is apparent that only atmospheric temperature showed statistically significant positive correlation with G. f.

fuscipes AD in the year 2011 (Spearman's R = 0.72, P = 0.004). On the contrary, both rainfall and RH% showed statistically significant negative correlation with *G. f. fuscipes* AD; rainfall (Spearman's R = -71, P = 0.004) and RH% (Spearman's R = -0.82, P = 0.001). In the year 2012, only atmospheric temperature showed no correlation with *G. f. fuscipes* AD (Spearman's R = 0.31, P = 0.163>0.05). Similarly, both rainfall and RH% showed statistically inverse correlation with *G. f. fuscipes* AD; rainfall (Spearman's R = -0.55, P = 0.031<p<0.05) and RH% (Spearman's R = -0.85, P = 0.00<p<0.01) as depicted in Table 6.

DISCUSSION

A study of ecological factors influencing the seasonal abundance of tsetse flies is imperative. *Glossina. f. fuscipes* identified in this study has confirmed the results obtained by Mohammed *et al.*

(2008). The existence of a large fly catches caught in Nyawa stream, KKC during the study period might be due to the presence of dense vegetation of tall shaded trees along this stream which provides shade and maintains a suitable microclimate for tsetse as well as a habitat for their vertebrate hosts. Such environmental parameters have created suitable conditions for the survival and flourishing of flies (Rogers and Randolph, 1985; Okoch *et al.*, 2011). This study showed a disparity in the higher number of male fly catches as compared with the number of female fly catches. Such disparity might be attributed to the response of the male flies to stationary objects and to traps, suggesting the high percentage of male catches (Okoch *et al.*, 2011).

Likewise, flies caught during dry season were significantly more in number than those caught during wet season which shows the typical behaviour of *G. palpalis* group (Ahmed *et al.*, 2000). This might be attributed to the concentration of such tsetse within riverine gallery forest or high capture performance of the trap during the dry season (Vale *et al.*, 1985). However, some studies reported that the number of fly catches in the wet season were significantly higher than those caught in the dry season (Baldry, 1970). The disparity in tsetse populations between the dry and wet seasons is well-documented in literature (Jordan, 1974). But, the increase in temperature was found to associate with increase in fly apparent density (Page, 1959).

The low fly abundance in the wet season compared with the fly abundance in the dry season might be due to lower temperatures which provide lower rate of breeding (Ahmed, 2004). However, a decrease in temperature slows down the rate of eggs development in the ovaries and increases the larval period leading to the inactivation of Glossina males rendering them unable to fertilize Glossina females which eventually could cause to population density drop-off (Mellanby, 1937); Moreover, the low fly apparent density in the wet season might be due to the presence of full leafed vegetation, which implies that many areas became water-logged and could attract predators such as spiders, dragonflies and ants on tsetse fly. Consequently, this might have forced the flies away from riverine vegetation into the open woodland (Okoch et al., 2011). Flies disperse beyond the water courses during the wet season is on contrary to the report in which flies disperse along river banks during wet seasons (Cano et al., 2007).

The high correlations of fly AD with atmospheric temperature, rainfall and RH% could explain the environmental conditions as the suitable range that favours the increment in the fly ADs. Rainfall has no direct effect on tsetse activity, but on the fly AD by affecting the humidity. Rainfall could cause local flooding that may lead to the drowning and submerging many pupae, but floods contribute to flourishing of different vegetation zones (FAO, 1982). This might

explain the lower fly AD in the wet season than that in the dry season. However, it was found that all environmental variables had shown statistically significant relationships with the rhythms of *G. f. fuscipes* (Mohamed-Ahmed and Wynholds, 1997). The critical climatic factors are the mean annual rainfall and temperature; the maximum temperature is the dominant factor determining longevity of tsetse stages rather than humidity although it is important. The longevity is negatively correlated with the temperature (Nash, 1948; Buxton, 1955).

The infection rates of 4.7 and 9.0 % which were determined in the dry season and the wet season, respectively are more than an infection rate of 1.55% observed by Waisa et al. (2006) and less than that of 18.2% reported by Ahmed (2004). The infection rate observed during the wet and dry seasons could be correlated with availability of infected host being fed on, tsetse-age and length of time taken for trypanosomes to complete development in tsetse; the later factors are quite variable and temperature dependent. Seasonal fluctuations in infection rates are caused by seasonal variations in the mean ages of the tsetse flies. During the rainy season, the flies have longer life span due to the favorable conditions, which also leads to a shortening of the trypanosome development cycle; consequently high infection rate was scored. Tsetse population with a high proportion of old flies usually has the highest mean infection rates, because they have had opportunities to develop infections (Tarimo et al., 1985). During the hot-dry season the high temperature is unfavorable for tsetse flies, consequently their life span became too short to allow trypanosomes to reach infective stage, which culminated in low infection rate (Jordan, 1986). Yet it was observed that high ambient temperatures shortened the duration of the development cycle within tsetse (Fairbairn and Culwick, 1950). Moreover, it was proved that infection rates in tsetse flies are positively correlated with ambient temperature to which puparians were exposed. Thus, the high infection rate observed during the wet season might be due to the flies emerged from pupae deposited in the late hot dry season (Ndegwa et al., 1992). Otherwise such flies might be primarily more likely to feed on infected hosts with trypanosomes than those of the dry-season flies which might feed on less satisfactory trypanosome reservoirs (Leak, 1999).

CONCLUSION

Some ecological factors which included atmospheric temperature, rainfall and relative humidity might be responsible for the seasonal variations in the fly total catches, male/female abundance, AD and the infection rate. These ecological factors could be used in developing tsetse control strategies.

ACKNOWLEDGMENT

We would like to acknowledge Bill and Melinda Gates Foundation (BMGF) for funding this research activity through Makerere University. Sincere thanks are due to technical staff at the National Ministry of Animal Resources and Fisheries, South Sudan for technical assistance.

REFERENCES

- Ahmed, A.B., 2004. A peridomestic population of the tsetse fly *Glossina palpalis palpalis* Robineaudesvoidy, 1830 (Diptera: Glossinidae) at Kontagora, Niger State, Nigeria. Entomol. Vect., 11(4): 599-610.
- Ahmed, A.B., G.A. Omoogun and S.S. Shaida, 2000. Trypanosome infections in Glossina species at the Kainji Wild Life Park, Nigeria. Entomol. Soc. Nig. Occ. Publ., 32: 57-62.
- Aksoy, S., W.C. Gibson and M. Lehane, 2003. Interactions between tsetse and trypanosomes with implications for the control of trypanosomiasis. Adv. Parasit., 53: 81-83.
- Baldry, D., 1970. Observations on peridomestic breeding behaviour and resting sites of *Glossina tachinoides* Westown near Nsukka, East Central State, Nigeria. B. Entomol. Res., 59: 585-593.
- Brunhes, J., D. Cuisance, B. Geoffroy, J.P. Hervy and J. Lebbe, 1998. Logiciel d'identification glossine expert. Manuel illustré d'utilisation. Les glossines ou mouches tsé-tsé. Didactiques, ISSN: 1142-2580.
- Buxton, P.A., 1955. The Natural History of Tsetse Flies, an Account of the Biology of the Genus *Glossina* (Diptera). In: Lewis, H.K. (Ed.), Memoir No. 10 of the London School of Hygiene and Tropical Medicine. Maclehass and Co. Ltd., the University Press, Glasgow, pp: 816.
- Cano, J., M.A. Descalzo, N. Ndong-Mabale, P. Ndongo-Asumu, L. Bobuakasi, N.J. Buatiche, S. Nzambo-Ondo, M. Esono, A. Benito and Roche, 2007. Spatial and temporal variability of *Glossina palpalis palpalis* population in the Mbini focus (*Equatoria Guninea*). Int. J. Health Geogr., 6: 36-42.
- Clloyd, L.L. and W.B. Johnson, 1924. Alternative approach to detect *Trypanosoma* in Glossina. B. Entomol. Res., 14: 265-288.
- Dede, P.M., I. Halid, G.A. Omoogun, N.R. Nziogwe, C.I. Njoku, A.D. Daniel and A.J. Dadah, 2005. Revue ÉlevMédVét. Pays. Trop., 58(1-2): 31-35.
- Fairbairn, H. and A.T. Culwick, 1950. The transmission of polymorphic trypanosomes. Acta Trop., 7: 19-47.
- FAO, 1982. Ecology and Behaviour of Tsetse Fly. In: Training Manual for Tsetse Control Personnel. FAO, Vialedelle Terme di Caracalla, 00100 Rome, Italy, 2: 100.

- Hay, S.I., R.W. Snow and D.J. Rogers, 1998. From predicting mosquito habitat to malaria seasons using remotely sensed data: Practice, problems and perspectives. Parasitol. Today, 14: 306-313.
- Jordan, A.M., 1974. Recent developments in the ecology and methods of control of tsetse flies (*Glossina* spp.) (Diptera: Glossinidae): A review. B. Entomol. Res., 63: 361-399.
- Jordan, A.M., 1986. Trypanosomiasis Control and African Rural Development. Longman, London, pp. 343.
- Leak, S.G.A., 1999. Tsetse Biology and Ecology: Their Role in the Epidemiology and Control of Trypanosomosis. CABI Publishing/ILRI, Nairobi, Kenya, pp: 568.
- Mellanby, H., 1937. Reproduction in the tsetse fly *Glossina palpalis*. Parasitology, 29: 131-141.
- Mohamed-Ahmed, M. and Y. Wynholds, 1997. Effects of vegetation and weather on trap catches of *Glossina fuscipes fuscipes* near Lake Victoria, Kenya. Entomo/Oria Exp. Appl., 85: 231-236.
- Mohammed, Y.O., M.M. Mohammed-Ahmed, K.H. El Malik and I.E. El Rayah, 2008. Distribution pattern of *Glossina fuscipes fuscipes* (Diptera: Glossinidae) in Bahr el Jebel State, southern Sudan. Int. J. Trop. Insect Sci., 27(3/4): 210-215.
- Nash, T.A.M., 1948. Tsetse flies in British West Africa. Colonial Office, 1-75, HMSO, London.
- Ndegwa, P.N., L.W. Irungu and S.K. Moloo, 1992. Effect of puparia incubation temperature: increased infection rates of *Trypanosoma congolense* in *Glossina morsitans centralis*, *G. fuscipes fuscipes* and *G. brevipalpis*. Med. Vet. Entomol., 6: 127-130.
- Okoch, K.E., I.S. Ndams, E. Kogi and C.G. Vajime, 2011. Catch composition of Tsetse flies. Am. J. Appl. Sci., 8(11): 1067-1072.
- Page, W.A., 1959. The ecology of *Glossina palpalis* (R-D) in southern Nigeria. B. Entomol. Res., 50: 617-631.
- PATTEC (Pan African Tsetse and Trypanosomiasis Eradication Campaign) 2012. Background and Justification Tsetse Flies are Reported from 37 AU-Member States and Causes Enormous Suffering and Death Both to Human. Milestones for 2012. rea.au.int/en/RO/PATTEC .Retrieved from:
 - https://www.google.com/#q=Pan+African+Tsetse+and+Trypanosomiasis+Eradication+Campaign+(20 12) (Accessed on: March 26th, 2014)
- Rogers, D.J. and S.E. Randolph, 1985. Population ecology of tsetse. Ann. Rev. Entomol., 30: 197-216.
- Rogers, D.J., S.I. Hay and M.J. Packer, 1996.

 Predicting the distribution of tsetse flies in West
 Africa using temporal fourier processed
 meteorological satellite data. Ann. Trop. Med.
 Parasit., 96(3): 225-241.

- Snow, W.F., 1983. Assignment report: Tsetse distribution and ecology in relation to sleeping sickness in Southern Sudan. WHO, EM/PD/5: EM/SUD/MPD/005/ FR (UNHCR) (2301).
- Tarimo, S.R.A., W.F. Snow, L. Butler and R.O. Ransfield, 1985. The probability of tsetse acquiring trypanosome infection from a single blood meal in different locations in Kenya. Acta Trop., 42: 199-207.
- Vale, G.A., E. Bursell and J.W. Hargrove, 1985. Catching-out the tsetse fly. Parasitol. Today, 1: 106-110.
- Waisa, C., E. Picozzi, W. Olaho-Mukani, R.A. Musoke and S.C. Welburn, 2006. *Glossina fuscipes fuscipes* in the trypanosomiasis endemic areas of south eastern Uganda: Apparent density, trypanosome infection rates and host feeding preferences, Acta Trop., 90(1): 23-29.
- WHO (World Health Organization), 1986. Epidemiology and control of human African trypanosomiasis. Report of a WHO Expert Committee on Sleeping Sickness, World Health Organization Technical Report Series No. 739, World Health Organization, Geneva.