FULL LENGTH RESEARCH ARTICLE

ACTIVE TRANSMISSION OF *Trypanosoma brucei gambiense* Dutton, 1902 SLEEPING SICKNESS IN ABRAKA, DELTA STATE, NIGERIA.

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ABSTRACT

Active surveillance of Human African trypanosomiasis (HAT) or sleeping sickness was undertaken in 3 agrarian villages in Ethiope East Local Government Area of Delta State, Nigeria. Card Agglutination Trypanosomiasis Test (CATT) was used qualitatively for mass screening with undiluted fresh whole blood (WB) and quantitatively for diagnosis in serum dilution tests. Thereafter, palpation for enlarged cervical lymph gland (ECLG) was followed by parasitological examination of aspirate using wet film, haematocrit centrifugation technique (HCT) and mini-anion exchange centrifugation technique (mAECT). Only one confirmed case of sleeping sickness was diagnosed out of the 491 samples screened. The results showed 43 (9.8%) serological positive cases in WB/ CATT test. 12 (27.9%) suspected cases that reacted at ≥1/4 titre in serum dilution test were highly suspected serological positive but parasitological negative cases. The study indicates that there is ongoing active transmission of Gambian type sleeping sickness in Abraka focus of Nigeria. The highly suspected cases will be followed up. Many cases might have gone undetected and more villages within the same focus were not covered. Moreover, a large-scale multi-disciplinary disease surveillance, vector and animal reservoir studies are required to determine the true situation of HAT in this focus.

KEY-WORDS: Transmission, Gambian Trypanosomiasis, Screening, Blood, Serum, Diagnosis.

INTRODUCTION

Human African Trypanosomiasis is a serious neglected tropical disease transmitted by the tsetse fly (Glossina spp.) with an estimated clinical transmission of about 300,000 to 500,000 cases yearly. Prompt diagnosis and staging are essential, as the disease is difficult to treat if not detected early and fatal if left untreated (Chretien & Smoak 2005). The clinical signs associated with the initial or haemolymphatic stage are chancre, fever, and general malaise, adenopathy, skin rash and pruritus, local eodema, cardiovascular disturbances, endocrine dysfunction, and even neurological disorders. As the disease progresses to the late or meningoencephalitic stage, the nervous system becomes affected. The clinical signs associated with this stage include disturbance of consciousness and sleep; disorder of tonus, motility and abnormal movements; mental changes including psychiatric problems (Buscher & Lejon 2004).

The resurgence of the disease has been reported in areas where population migration has been induced by conflicts, with the most affected countries being Democratic Republic of Congo, Angola, Central African Republic, and Southern Sudan. Its re-emergence is attributed to long years of geopolitical instability and a breakdown of control programmes. However, in Cameroun, Chad, Cote d' Ivoire, Gambia and Nigeria, the disease has re-emerged after long years of control (Moore & Richer 2001 ; Stanghellini & Josenando 2001 ; van Nieuwenhove *et al*. 2001).

Reports of passive cases are on the increase from the Abraka focus in Delta State (Airauhi *et al.* 2000, Enwezor *et al.* 2000, Halid *et al.* 2001). Recent case involved a child in advance stage of the disease. Over the years, the situation of sleeping sickness in Nigeria has been under reported due to absence of active surveillance. There is need for the magnitude of the disease spread to be ascertained in order to delineate and curtail any impending epidemic from ensuing. More importantly, the disease cannot be easily diagnosed at the level of available rural health care institutions with the facilities of Rural Health Centre, Clinic and General Hospital. There is need to create an awareness among the health care provider for the disease to be included as one of the likely disease when attending to patients presenting with those diseases that share similar clinical signs with HAT.

MATERIALS AND METHODS

Study Area: The Abraka focus in Delta State, Nigeria has been described (Weir *et al*. 1985; Edeghere *et al*. 1989). It lies between latitude 5° 47' and 6° 15 E and longitude 5° 42-6°N. The indigenous inhabitants are mostly Urhobo ethnic extraction whose main occupation is farming and related activities.

Training of Personnel at the Grassroot: A one week intensive training workshop was undertaken to transfer basic skills and techniques for mass screening and diagnosis of the disease to participants.

The participants included officers drawn from the Federal, State and the Local Government Area Councils (LGA) involved in public and private health care delivery system, country representative of World Health Organisation (WHO) Communicable Disease Surveillance, WHO African Regional Office, and WHO Head Office in Geneva. Also involved is the Prince Leopold Institute for Tropical Medicine, Antwerp, Belgium, the Manufacturer of CATT *T. b. gambiense* kit. The work will form the basis for further research initiative aimed specifically at the control of the disease in this focus and enhancing the capacity of frontline players for both passive and active disease surveillance, reporting, treatment and case management.

Epidemiological Protocol and Field Studies: Individuals aged 3 years and above of both sexes were screened as described by Edeghere *et al.* (1989) except that all those presenting for screening were finger pricked to collect blood in heparinised capillary tubes.

Qualitative Serology: The CATT kit used for this study was developed at the Institute for Tropical Medicine, Antwerp, Belgium. Only those whose fresh whole blood (undiluted) that showed reaction in direct agglutination test were subjected to further examination as described by Lejon *et al.* (2003).

Quantitative Serology: Titration of serum samples was performed in the field on patients who were whole blood CATT positive. About 5ml blood samples were collected by venesection into ethylene diamine tetra acetic acid (EDTA) containing bottles using sterile disposable syringes and needles from patients who showed very strong reaction of >1/4 titre. This set of people was subjected to further parasitological examination.

Parasitology: Palpation for enlarged cervical-lymph gland (ECG) was carried out and those found positive had lymph aspirate collected for wet film microscopy. Haematocrit centrifugation techniques (HCT) buffy coat examinations (Woo 1970) were performed on blood of those with 1/8 titre but were ECG negative. Mini-anion exchange centrifugation technique (mAECT) was used for the serum dilutions that were positive at 1/32 titre.

RESULTS

The survey of three villages (Urhouka, Oria and Ugono) in Abraka focus of Delta State using the CATT screening kit revealed only 1 positive parasitological case out of the 300 screened at Urhuoka with 26 seropositve in whole blood test (Table 1). No positive case was recorded among the 3 and 14 seopositives from Oria and Ugono drawn from the 71 and 120 sample populations respectively. The only confirmed parasitological positive case was from aspirate of enlarged cervical lymph gland of a 46 years old woman with evidence of overt signs indicative of clear case of sleeping sickness. The patient could not walk nor stand on her own, and she was sub-conscious and dropping saliva from the mouth. Both her son and daughter who brought her showed strong serological reaction of 1/8 and 1/32 respectively. Two other females showed a titre of 1/32, indicating highly suspected HAT cases. These highly suspected cases were both haematocrit and minianion centrifugation technique (mAECT) negative. The confirmed case was referred to Eku Baptist Hospital where clinical staging will be carried out and treatment instituted accordingly.

DISCUSSION

A spot active survey of HAT in Abraka focus, Delta State, Nigeria was undertaken using CATT kit for serological screening and parasitological confirmation by wet film microscopy of lymph aspirates, HCT and mAECT of blood samples. The study area is a relatively active sleeping sickness endemic focus first reported by Francis (1972) and Olowe (1975). Abraka focus has since remained one of the most active HAT transmission focus compared to the old foci in the North.

The epidemiological importance of the only parasitological positive case of a woman from Uruoka Village was re-enforced by the seropositivity of her daughter and son. It showed that they could have contracted the infection within the same location. Information from both passive and active surveys undertaken in this area in recent past (Elhassan *et al*. 1997; Airauhi *et al*. 2000) confirmed that the risk of infection of a family is determined by type of occupation, particularly farming and source of water. The consequences of the disease therefore on a family will be more devastating where two or more family members are infected as may be the case in this study.

TABLE 1. RESULTS OF CATT REACTION WITH WHOLE BLOOD AND SERUM PARASITOLOGICAL DIAGNOSIS OF HAT

VILLAGES	Screened population	CATT					Parasitology				Patients			Active Prev
		Whole blood		Serum dilutions			ECLG (%)		HCT (%)					
		+	%	<1/4	= 1/4	>1/4	-	+	-	+	Total	P1	P2	
Urhuoka	300	26	8.7	17	4	5	8 (2.7)	1(0.3)	8(2.7)	0	1			0.33%
Oria	71	3	4.2	2	0	1	1	0	1	0	0			0
Ugono	120	14	11.7	12	2	0	0	0	0	0	0			0
Total	491	43	8.8	31	6	6	9	1	9	0	1			0.2%
%				**6.3%**	**1.22%**	**1.22%**	**1.8%**	**0.2%**	**1.8%**		**0.2%**			

The CATT kit could differentiate past infection from on-going infection. This is demonstrated by the results of the few old cases (n=4) that had been treated and cured among the sample population. Three (3) were negative and one (1) was slightly positive by the WB screening test. The dilution test was capable of differentiating active infection from past exposure to infection. But the period when this can occur has to be determined, as most of the old cases (n=3) that were more than 12 years post treatment (since 1993) based on oral information obtained from them were negative.

The WB positive patient WHO was only slightly positive by dilution test with 1/2 titre had been treated in 1999. Similarly, a pregnant young woman that was WB positive was only slightly positive in dilution test (1/2 titre). The later might be indicative of cross-reactivity attributable to pregnancy and other parasitic diseases like malaria and schistosomiasis have been documented elsewhere using this kit (Lejon unpublished personal Communication).

The need for follow up survey to monitor the parasitological status of the positive cases (n=3) having serum dilutions of ≥1/16 titres is recognised These were highly suspected serological positive cases that were apparently parasitologically negative. Using the diagnostic algorithms of Chappuis *et al*. (2005), all cases that were ≥1/4 (n=12) as shown on the Table, should be followed up for both clinical and parasitological diagnoses. Lumber puncture was not performed during this study. The Eku Baptist Hospital where the patient was referred for treatment will handle the staging of the parasitological case.

In the interpretation of this data, we have to be cautious of the fact that the CATT kit has been reported to be less sensitive (Lejon *et al.* 2003). Its sensitivity is dependent upon geographical region: it may be lower in Fontem focus in Cameroun (Dukes *et al.* 1992) and Ethiope East focus of Nigeria (Buscher *et al.* 1999) compared to result obtained from elsewhere. While it is possible to miss some cases which cannot be determined, it is plausible that some of the WB positives were due to cross reaction judging from the dilution tests. The 12 (2.44%) serological suspected cases of the disease call for concern. The situation is worrisome and calls for urgent action to prevent a catastrophe.

During this spot survey that followed the training workshop, participants had practical field experience on how to use both CATT kit and the reagents. Some kits and reagents were handed over to the State Ministry of Health to facilitate passive and active case detection in the area. This has partially addressed some of the concerns expressed (Airauhi & Halid 2001) on how to eliminate the disease in this focus.

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