ORIGINAL ARTICLE

Surveys for *Mansonella perstans* Filariasis in Kalabo, Kazungula, Choma and Kafue Districts of Zambia

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ABSTRACT

Background: Past case reports have documented *Mansonella perstans* infections in Zambia. However, knowledge on the epidemiology and geographical distribution of this infection in the country is lacking. This paper reports on surveys for *M. perstans* in communities in four districts(Kalabo, Kazungula, Choma and Kafue) in the Southern and Western parts of Zambia.

Design: The study was cross sectional. In the Kalabo District surveys, individuals aged one year and above had thick blood smears prepared and examined for *M. perstans* microfilariae (mf). In the other three districts the study design was retrospective and prospective, i.e. previously examined archived malaria slides from health centres were re-examined for *M. perstans* mf and at the same time individuals aged 15 years and above had thick blood spears prepared and examined for *M. perstans* mf.

Results: The retrospective study could only be undertaken in Choma District due to change in

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Department of Paraclinical Studies School of Veterinary Medicine University of Zambia Lusaka, Zambia Telephone: +260 966 757120 Email: <u>shawatam@yahoo.co.uk</u> or shawatam@gmail.com malaria examination method in the other districts from conventional preparation of Giemsa stained thick blood smears to use of rapid diagnostic tests. For the prospective surveys, out of the 1439 individuals recruited and examined, no *M. perstans* mf were seen in any of the blood smears.

Conclusions: The failure to find *M. perstans* mf was surprising considering previous case reports, even from some of surveyed areas. There is a need for more surveys to be carried in other parts of the country to ascertain the distribution of *M. perstans*. Health practitioners should moreover be informed about this infection, and trained to be able to accurately distinguish *M. perstans* infections from those of *W. bancrofti*, which are also endemic in Zambia.

INTRODUCTION

Mansonella perstans is a vector borne filarial nematode parasite of humans, transmitted by tiny flies of the genus *Culicoides* (biting midges). The adult females produce small larvae called microfilariae (mf) which find their way to the blood circulation. The mf are picked up by the vectors when they take a blood meal, and after a period of development in the thoracic muscles of the vector, the parasites may be transmitted onward to new human hosts when the vector bites again [1,2]. The development in the vector takes approximately one

to two weeks depending on the environmental conditions but the period required for further development to mature adult stages in the human host is unknown. Adult worms appear to live mainly in the serous body cavities, but have rarely been recovered[1].

Mansonellaperstans infections are widely distributed in Africa and also occur in parts of Central and South America and the Caribbean. Despite this, only a few studies have been carried out on the epidemiology and morbidity of this infection in endemic populations. This could probably be due to the lack of association with a distinct and specific clinical picture or lack of effective treatment for patients suffering from this infection [2,3]. Diagnosis of *M. perstans* infection is mostly by detection and identification of mf in peripheral blood. The microfilariae of *M. perstans* show a weak pattern of diurnal periodicity but are present in the peripheral blood both during the day and night[4].

Case reports from Zambia

The first cases of *M. perstans* infection in Zambia were reported by Buckley in 1946 from hospital patients in Lusaka, Ndola and Kasama[5]. Later on, more cases of *M. perstans* were reported by Barclay from the Luangwa basin when he carried out a survey for another filarial parasite, Wuchereria bancrofti [6]. More cases of individuals with M. perstans infection were recently seen in Chama District in the Luangwa valley, Eastern Province [7]. A few cases were moreover reported by medical personnel from district health centers in the recent past. Thus, two cases were reported from Mambova Health Centerin Kazungula District in 2012 by a medical officer, and in 2010 a woman from Shimabala area in Kafue District being investigated for trypanosomiasis was found with mf of M. perstansat the University Teaching Hospital in Lusaka (UTH records, unpublished findings). However, despite these reports of *M. perstans* since 1946, there has been no survey in any parts of the country to determine the occurrence and prevalence of M. perstans. Most of the reports of M. perstans infection were incidental findings as a result of other

surveys that were being carried out. Species of *Culicoides*, which are the vectors responsible for transmission of *M. perstans* infection, have been reported near Chilanga District in Lusaka Province [8].

Considering that Zambia is endemic for lymphatic filariasis caused by infection with the filarial parasite *Wuchereria bancrofti*[9,10], and that control activities to eliminate this disease as a public health problem are currently underway, it is important that medical personnel and scientists are aware of both parasites and are able to distinguish the mf of *M. perstans* from those of *W. bancrofti* for precise diagnosis and treatment. As a first step, the present survey was undertaken to determine the occurrence of *M. perstans* in some districts including those that had recent case reports of the parasites.

METHODS

Study sites

Spot check surveys were carried out in four districts. The first wasin August 2012 in Kalabo District, Western Province. Three villages at an altitude of 1000 - 1100 m above sea level, namely Sishekanu (14.84071S, 22.80762E), Lutwi (15.17347S, 22.38348E) and Liumena (14.98859S, 22.32845E) were selected on the basis of previous surveys that had recorded high prevalence of W. bancrofti infection [10]. The other surveys were carried out in March/April 2014 in three districts in Southern parts of Zambia. In Kazungula District, study participants were recruited from two villages at an altitude of 900-1000 m above sea level and located approximately 60 km apart, namely, Mukuni (17.90759S, 25.94151E) and Mambova (17.73088S, 25.19528E) under Chief Mukuni. Mambova had previously reported two cases of M. perstans infections. In Choma District, the study site was located approximately 20 km east of Choma town at an altitude of about 1200 m above sea level and in close proximity to Shamphande Health Centre (16.92256S, 26.99680E) which was among the few communities still reporting cases of malaria infection in the district. In Kafue District, study participants were recruited from three communities surrounding namely Nangongwe, Railways and Kafue Estates(15.78150S, 28.18368E) at an altitude of approximately 900 m above sea level.

Study design

A cross sectional survey was carried out in Kalabo District, where freshly prepared Giemsa stained thick blood smears were examined for *M. perstans* mf from individuals aged one year and above from Sishekanu, Lutwi and Liumena villages. In the other three districts, the surveys were initially designed to be both retrospective and prospective in nature, whereby previously examined archived malaria slides from health centres would be re-examined for M. perstans mf and at the same time, freshly prepared Giemsa stained thick blood smears from malaria suspected patients were also examined for mf. For the latter part, individuals aged 15 years and above from the surrounding communities reporting to the health centres during the survey and suspected to have malaria were recruited.

Ethical considerations

Permission to undertake the surveys was obtained from the Biomedical Ethics Committee (ref no. 007-06-11), the Ministry of Health, and the Provincial and District Health Offices. Permission was also sought from the local area chiefs and village headmen. Oral consent was obtained from the individuals before recruitment into the survey. Once they consented to participate, they were requested to provide a finger prick blood smear. Permission to reexamine the achieved slides were sought from the Ministry of Health, and the Provincial and District Health Offices.

Sample collection and analysis

In Kalabo, three villages with more than 200 individuals were selected. Community members aged one year and above were recruited into the study and examined for *M. perstans* mf by collection of small amounts of blood from a finger prick which was used to prepare a thick blood smear. In order to

recruit at least 300 individuals from each of the other three study districts (Kazungula, Choma and Kafue) to the prospective study, two or more communities were surveyed in each district. Community members aged 15 years above and from the surrounding community reporting to health centres and suspected to have malaria were recruited into the study and examined for *M. perstans* mf by preparing blood slides as described above for Kalabo. The slides were allowed to dry overnight and thereafter deheamoglobinized in clean tap water. The slides were then fixed with methanol, allowed to dry in the air and stored in slides boxes. Upon arrival at the parasitology laboratory at the University Teaching Hospital in Lusaka, the slides were stained using Giemsa and examined under a microscope for *M. perstans* mf [11]. For the retrospective study carried out in Choma district only, all archived malaria slides at Shamphande health centre were examined for M. perstans mf regardless of age, because only a few slides were available for examination

RESULTS

Although the study was designed to be both retrospective and prospective, it was only possible to carry out the retrospective part in one of the districts because the health centres in the other districts did not have archived malaria slides due to change in the malaria examination procedure from conventional preparation of Giemsa stained blood smears to using rapid diagnostic tests (RDTs). In Choma District, 64 archived slides(33 from females, 31 from males; mean age 16.4 years; range 1-53 years)were recovered at Shamphande health centre from individuals who had tested positive for malaria by the rapid diagnostic test. All 64 slides were examined for *M. perstans* mf and none were positive.

In the prospective study, a total of 1439 blood smears were screened for *M. perstans*mf in the four districts combined as shown in Table 1. The overall

mean age of the examined was 32.8 years and more females (65.5%) than males (34.5%) were examined. Microscopic examination revealed no M. *perstans* mf in any of the smears.

 Table 1: Study populations examined for *M. perstans*

 infection in the four districts

District	No. examined	Mean age (range) in years	Female : male ratio
Kalabo	425	31.0(1-82)	1.85
Kazungula	348	34.7 (15-83)	1.54
Choma	306	33.2(15-87)	1.66
Kafue	360	33.2 (15-84)	2.79
Total	1439	32.8(1-87)	1.90

DISCUSSION

The failure to find *M. perstans* mf in the examined individuals from the four districts was surprising as previous reports had indicated the presence of these parasites in Zambia[5-7]. Two of the four districts, namely Kazungula and Kafue, were selected based on previous finding of *M. perstans* mf there. However, it is possible that these mf positive individuals either had their homes in other parts of the districts or had travelled or resided in other areas endemic for *M. perstans* transmission. Unfortunately, the history and movements of the individuals had not been recorded.

The present findings could also be an indication of a recent decline in the transmission of *M. perstans* due to unfavourable environmental conditions. A recent study on human trypanosomiasis in the Luangwa valley [7], where a high prevalence of *M. perstans* microfilaraemia (23.4%) had previously been documented[6], only recorded five incidental cases of *M. perstans* microfilaraemia (0.8% of those examined) which might suggest a possible decline in transmission. Potential *Culicoides* vectors for *M. perstans* have been identified in some parts of the country [8], but it is possible that a decline in vector population due to changes in environmental conditions also could have affected transmission.

Considering that the number of districts surveyed was few, it is likely that *M. perstans* infection may occur in other areas that were not surveyed, but with

favourable environmental conditions. Hence, there is a need for more surveys to ascertain the presence of M. perstans parasites in other districts. Moreover, the surveys were undertaken in the southern and western parts of the country, and even though most cases have been reported from the southern parts, the individual may have acquired the infection in other parts where the infection is endemic.

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