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Diversity and altitudinal distribution of phlebotomine sand flies (Diptera: Psychodidae) in visceral leishmaniasis endemic areas of northwest Ethiopia

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ABSTRACT

Background: The Leishmaniases are caused by the protozoan parasites of the genus *Leishmania* and are transmitted to humans by the bite of infected female phlebotomine sand flies. Both visceral and cutaneous leishmaniases are widely distributed in different parts of Ethiopia. The aim of this study was to determine the diversity and altitudinal distribution of phlebotomine sand flies from Kafta Humera to Gondar town in northwest Ethiopia.

Methods: Seven localities were selected with distinct altitudinal variations between 550 m above sea level (m a.s.l) and 2300 m a.s.l. In each locality, sand flies were collected using standard CDC light traps and sticky traps during the active sand fly season from December 2012 to May 2013. Shannon-Weiner species diversity index and Jaccard's coefficient were used to estimate species diversity and similarity between altitudes and localities, respectively.

Results: A total of 89,044 sand flies (41,798 males and 47, 246 females) were collected from the seven localities/towns throughout the study period. Twenty-two species belonging to 11 species in the genus *Phlebotomus* and 11 species in the genus *Sergentomyia* were documented. Of these, *Sergentomyia clydei* (25.87%), *S. schwetzi* (25.21%), *S. africana* (24.65%), *S. bedfordi* (8.89%), *Phlebotomus orientalis* (6.43%), and *S. antennata* (4.8%) were the most prevalent species. The remaining 10 *Phlebotomus* species and six *Sergentomyia* were less frequent catches. In CDC light trap and sticky trap, higher species diversity and richness for both male and female sand flies was observed at low altitude ranging from 550 to 699 m a.s.l in Adebay village in Kafta Humera district whereas low species richness and high evenness of both sexes were also observed in an altitude 1950–2300 m a.s.l.

Conclusion: The results revealed that the presence of leishmaniasis vectors such as *P. orientalis, P. longipes, P. papatasi,* and *P. duboscqi* in different altitudes in northwest Ethiopia. *P. orientalis* a vector of *L. donovani*, occurred between altitude 500–1100 m a.s.l, the area could be at high risk of VL. *P. longipes* a vector of *L. aethiopica*, was recorded in the highland area in Tikil-Dingay and Gondar town, implicating the possibility of CL transmission. Hence, further investigation into vector competence in relation to leishmaniasis (VL and CL) in the region is very vital.

1. Introduction

Phlebotomine sand flies are involved in the transmission of leishmaniases, sand fly fever and bartonellosis in the tropical and subtropical regions of the Old and New Worlds. In the Old World, phlebotomine sand flies belong to three genera: *Phlebotomus*, *Sergentomyia* and *Chinius*. Only species occurring in the genus *Phlebotomus* are responsible in the transmission of leishmaniasis and

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sand fly fever in the Old World (Lane, 1993; WHO, 2010).

Leishmaniasis exist in two main clinical forms: visceral leishmaniasis (VL) and cutaneous leishmaniasis (CL) which both occur in different parts of Ethiopia. Cutaneous leishmaniasis is caused by three different *Leishmania* species: *L. aethiopica*, *L. major* and *L tropica*, however, CL due to *L. aethiopica* is by far the most widespread and important disease in Ethiopia (Hailu et al., 2006). The estimated annual incidence of CL is about 20,000–50,000 cases with a population of about 28 million at risk (Alvar et al., 2012; Tsegaw et al., 2013). It mainly occurs between 1400 and 2900 m a.s.l (Wilkins, 1972; Ashford et al., 1973; Hailu et al., 2006). In contrast, VL is mainly found in the peripheral low land areas of the south, southwest, north and northwest Ethiopia (1500 m a.s.l) (Hailu et al., 2006) but has recently been emerged in highland areas ranging from 1800 to 2000 m a.s.l (Alvar et al., 2007; Herrero et al., 2009). The estimated annual incidence of VL is between 3700 and 7400 cases (Alvar et al., 2012; Deribe et al., 2012), and the population at risk is about 3.3 million in Ethiopia (Tsegaw et al., 2013).

In Ethiopia, at least 22 species of *Phlebotomus* have been documented, of which eight species have been proved or suspected as vectors of CL and VL, *P. martini*, *P. orientalis* and *P. celiae* are vectors of VL due to *L. donovani* while *P. longipes*, *P. pedifer* are vectors of *L. aethiopica* (CL), *P. duboscqi* is vector of *L. major* (CL) and *L. tropica* (CL) transmission is associated with *P. sergenti* and *P. saevus* (Gebre-Michael et al., 2004a; Hailu et al., 2006; WHO, 2010). With regards to the distribution of vectors of VL, they are widely spread in different ecological settings. Two species of the subgenus *Synphlebotomus P. martini* and *P. celiae* and another related species *P. vansomernae* (its role yet unknown) are associated with *Macrotermes* termite mounds (Minter, 1964; Gebre-Gebre-Michael and Lane, 1996; Marlet et al., 2003). These are generally found in the southern parts of the country, but *P. martini* have been found as far north as Thatay Adiabo district in Tigray Region (Gebresilassie et al., 2015). *Phlebotomus orientalis* which is the most likely VL vector in north and northwest Ethiopia (Hailu et al., 1995; Gebre-Michael et al., 2010) is mostly distributed in areas where there are *Acacia-Balanites* vegetation and cracking black cotton clay soil (verstisol) as in Sudan and South Sudan (Hoogstraal and Heyneman, 1969; Elnaiem et al., 1999). In Ethiopia, *P. orientalis* has an extensive geographical distribution much more than the distribution of the disease (Gebre-Michael et al., 2004b).

Phlebotomine sand flies are widely distributed along different elevation gradients. The density of sand flies at higher and lower altitudes is dependent on different environmental and climatic conditions. The distribution and abundance of sand fly vectors and human and/or reservoir hosts are affected by various physical factors (temperature, rain fall, humidity, altitude, latitude, surface water and wind) and biotic factors (vegetation, host species, predators, competitors, parasites and human interventions) (Lane, 1993; Rohr et al., 2011). All of these factors also affect the spatial and temporal distribution of vectors and reservoirs, which in turn affect the epidemiology and dynamics of pathogen transmission to the human population (Rohr et al., 2011). Knowledge on diversity and altitudinal distribution of sand flies are very vital to predict the impact of environmental modification, the increasing seasonal labourer migration (from non-endemic to endemic areas and the vice versa) and climate change on the dynamics of vector population. Currently, cases of VL and CL have been reported in different parts of the country, showing that both diseases are spreading in previously non-endemic areas. A few years ago, few cases of VL were diagnosed in Gondar town (2300 m a.s.l) in children who have never been out of the town (Prof A. Hailu). VL cases have also been detected in some districts between Kafta Humera and towns near Gondar towns (e.g. Dansha, Sorkoa, Sanja) (Humera Hospital, unpublished), 1000–2000 cases occur annually in Kafta Humera-Metema plains (Ngure et al., 2009). In Kafta Humera, a case-control study showed that daily individual activities around home and farm fields during night times and poor housing conditions are important for VL transmission (Yared et al., 2014). The spreading of VL and CL in the country may be due to environmental change, demographic, host and human activity factors (Desjex, 2001). As well, the distribution and abundance of the sand fly vector are also enhancing the transmission of the disease (Desjex, 2001).

Determining, the faunistic composition and distribution patterns of phlebotomine sand flies can indicate the possible presence of autochthonous transmission and/or can aid in the incrimination of the vector species. It is essential to know the distribution of sand flies along various altitudinal gradients for continuous monitoring of VL and CL vectors in the study areas. Therefore, the present study was undertaken to determine species composition, distribution, diversity, altitudinal and ecological relationships of sand flies in representative localities between Kafta Humera (lowland) and Gondar town(highland)in northwest Ethiopia.

2. Material and methods

2.1. Study localities between Kafta Humera and Gondar

The study was conducted in a transect from Kafta Humera district to Gondar town (Fig. 1) because the sites represents a wide altitudinal range in a relatively compact and well defined geographical area. For this purpose, seven localities (Adebay, Dansha, Soroka, Ashere, Sanja, Tikil-Dingay and Gondar) were selected with distinct altitudinal variations between the two major localities along a transect on the main paved road between Humera and Gondar towns (Table 1). Additional criteria for selection were accessibility, security and availability of minimal accommodation/subsistence facilities. The altitude varied from 550 m a.s.l in Kafta Humera to 2300 m a.s.l in Gondar.

2.1.1. Adebay (Kafta Humera district)

Kafta Humera district is found in Western Tigray Zone, Northwest Ethiopia. It is situated at about 967 km from Addis Ababa. The district is mostly flat plain at altitudes of 550–699 m a.s.l. The villages are surrounded by uniform agricultural fields. Most of the natural vegetation in the Humera lowlands has been cleared for the extensive commercial agricultural practices, leaving only scattered *Acacia* and *Balanites* trees with neem (*Azardirachta indica*) grown commonly in urban areas as shade trees. Temperature rises to a maximum average of 42 °C between April and June and falls to between 25 and 35 °C during the moderate months between June and February. Crop production is exclusively dependent on the unimodal precipitation (average annual rainfall is 400–650 mm), which runs from July to September (Gemetchu et al., 1975).

2.1.2. Dansha town (Tsegede district, Western Tigray)

This is a small town situated about100kms from Setit Humera and 150 km from Gondar at altitude range of 700–799 m a.s.l. It is semi-arid with vertisol type of soil. Sesame and livestock are main the income sources in the area. The vegetation comprises *Acacia* trees and mixed forest.

2.1.3. Soroka (Tegede district, North Gondar Zone, Amhara region)

It is a semi- urban area about 125 km from Setit Humera and 125 km from Gondar town. It is located at altitude 800–849 m a.s.l. Livestock and sesame and sorghum farming are the main economic activities. *Acacia*, *Balanites* and *Boswellia* (incense) trees are common, although much of the natural vegetation has been cleared for farming, fuelwood and constructions (huts and fences). Angerib River passes through the village to join the Atbara River system in the eastern Sudan.

2.1.4. Ashere (Tach Armachiho district, North Gondar Zone, Amhara region)

This is a small town about 155 km from Setit Humera and 95 km from Gondar. It is located at altitudes of 850–920 m a.s.l. The