Characterization of sand fly breeding sites in vertisols in North-Western Ethiopia

Ecology and Transmission Dynamics of Visceral Leishmaniasis in Ethiopia

BILL & MELINDA GATES foundation

Aviad Moncaz[1] , Moshe Shenker[2] Araya Gebre Sellassie[3] , Oscar Kirstein[1], Ibrahim Abbasi[1] Teshome Gebre-Michael[3], Asrat Hailu[4] and Alon Warburg[1]

[1]The Faculty of Medicine, the Hebrew University, Hadassah Medical School, Jerusalem, Israel.
[2]Department of Soil and Water, The Faculty of Agriculture, the Hebrew University, Rehovot, Israel.
[3] Aklilu Lemma Institute of Pathobiology, Addis Ababa University, Addis Ababa, Ethiopia
[4]Department of Microbiology, Immunology & Parasitology, Addis Ababa University, Addis Ababa, Ethiopia
e-mail contact: adi.moncaz@mail.huji.ac.il

The Hebrew University of Jerusalem

Addis Ababa University

Introduction

Phlebotomine sand flies are the principal vectors of *Leishmania* Ross, 1903 (Kinetoplastida: Trypanosomatidae) , a group of protozoan parasites infecting populations exceeding 12 million worldwide During recent years foci of VL have been described in the Sheraro district near villages surrounded by vertisol fields. The successful identification and characterization of breeding and resting sites of leishmaniasis vectors in vertisol in North East Ethiopia may provide essential information for the development of measures for their control

Fig. 1 – Cracked vertisol – Putative breeding site of phlebotomine sand flies

Objectives

- To identify putative sand fly breeding sites in vertisols in rural areas in NW Ethiopia.
- To characterize the ambient conditions inside vertisol cracks including microclimate and physico/chemical parameters.

Materials and Methods

Putative breeding sites in cracked vertislol were covered with sand-fly proof net . Emerging sand flies were trapped on large sticky traps placed inside the net. Traps were baited with incandescence light or fermenting sugar solution (Fig 2). Temperature, relative humidity and soil volumetric water content were monitored continuously in two habitats: area shaded by trees canopy and open vertisol field.

Air samples were drawn and oxygen concentrations were measured at two different depths during low and high sand fly season. . Soil samples were analyzed for organic mater, pH, and electrical conductivity. DNA was extracted from blood-fed sand fly females and the blood host were determined by PCR and sequencing of the *cyt b* gene.

Fig. 2 – Emergence Traps baited with fermenting sugar sol.

Fig. 3– Micro – climate monitoring system

Results

Vast majority of sand flies trapped belonged to the genus *Sergentomyia* (2568/2592). Significantly more *Sergentomyia* spp. were captured in emergence traps placed under trees than in open fields (514±180 and 64 ± 29 sand flies/trap/night respectively), P=0.002 (Fig 4). Initial readings taken inside he soil at the end of rainy season showed stable temperature and humidity. Oxygen concentrations in shaded areas (0.7±13.5%) differed significantly from those in open fields (19.19±0.6%, P>0.0001) (Fig. 5). Subterranean temperature readings in shaded areas were stable and lower (26°C) than open fields (28°C). The relative humidity in subterranean habitats stabilized at 100% in all habitats. Microclimatic conditions were measured repeatedly during early and mid dry season. While Oxygen concentration had stabilized around atmospheric values, temperatures in deep cracks decreased with increased depth.

Mean volumetric water content was high in both habitats reaching 17.79±2.77% under tree canopies, and 20.37±1.21% in open fields. Volumetric water content increased significantly with the increase in depth (P< 0.0001, fig. 6). Organic matter content reached 5.38±0.14% under tree canopy, and 5.49±0.08% in open field. However, the difference was insignificant for different habitats (P= 0.579) and different depths (P=0.950) (Fig. 7).

Fig. 4 - Sand fly trapped at putative vertisol habitats

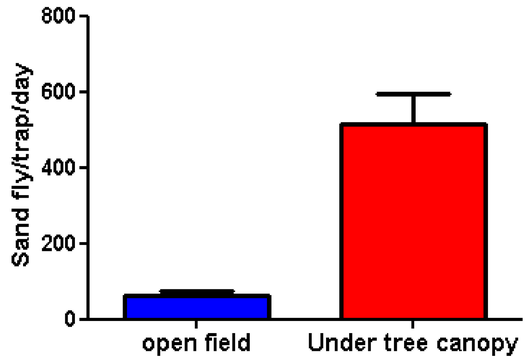


Fig. 5 - Oxygen Concentration at two habitatand two sempling depth

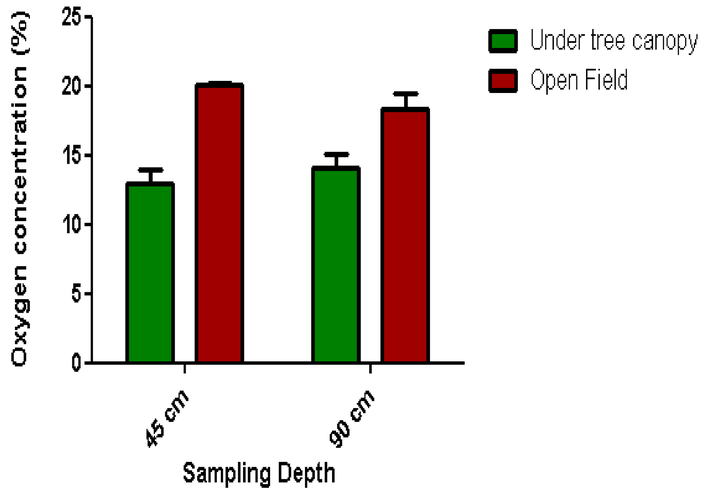


Fig. 6 - Volumetric water content at two habitats and four sampling depths

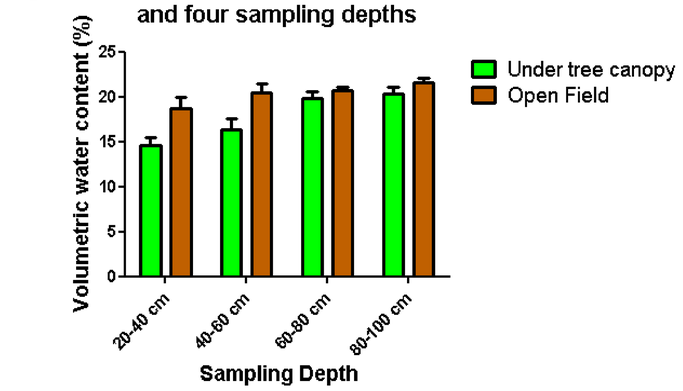
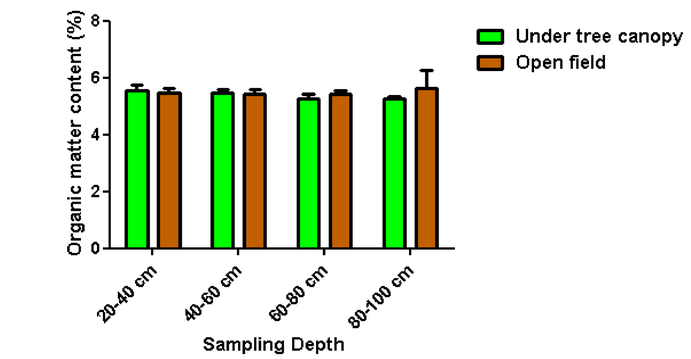


Fig. 7 - Organic matter content in the two habitats and four depths



Blood engorged females were tested for host blood meal (n=90). Human blood was identified in 9 *Phlebotomus orientalis* (putative vector of VL in the region). Human and bovine or camel blood were identified in 6 *Phlebotomus orientalis* . Interestingly, human blood was also identified in 34 of the 43 engorged *Sergentomyia* spp. females (79%).

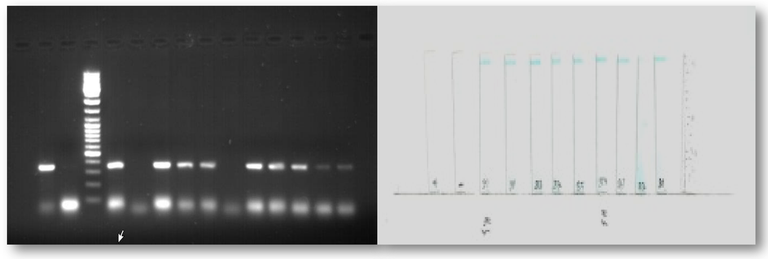


Fig. 8– PCR amplification with cytB primer (left side) and RLB (right side) of *Sergentomyia spp*. sand flies

Discussion

The trapping of phlebotomine sand flies emerging from cracked vertisols supports our assumptions that vertisols serve as sand fly breeding sites. Furthermore, the micro-climatic conditions inside cracks were stable with high relative humidity and soil moisture throughout the year, making them suitable for development of sand flies . Larval food may be provided by decomposing organic matter, demonstrated by both rich organic matter content and the low oxygen concentrations sampled during rainy seasons. The low oxygen concentration may be caused by microbial metabolic activity depleting oxygen . The proximity to villages apparently provides human and live-stock blood sources for questing females.