Short Report

First Report on Natural Promastigote Infection of *Phlebotomus* caucasicus in a New Focus of Visceral Leishmaniasis in North West of Iran

*Y Rassi¹, F Kaverizadeh¹, E Javadian¹, M Mohebali²

Abstract

An investigation was carried out in Ahar district, (a new focus of visceral leishmaniasis), Eastern Azerbaijan, North-West of Iran in order to determine the vector(s) of the disease, during the summer of 2003 sand-flies were collected using sticky papers, CDC light traps and aspirators. All of the collected sand-flies were dissected in a drop of normal saline for surveying of promastigotes. Head and two last abdomen segments of-sandflies were mounted in a drop of Puri's Medium for species identification. Totally 215 sandflies were collected including: *Phlebotomus kandelakii* (57.2%), *P. perfiliewi* (18.1%), *P. caucasicus* (11.2%), *P. sergenti* (7.9%), *P. papatasi* (3.3%) and *P. (adlerius)* sp.(2.3%). Only 1 of 24 *P. caucasicus* species (4.2%) was found naturally infected with parasite. This is the first infection report of *P. caucasicus* in this region. Some behavioral aspects of the mentioned sandfly have been discussed in details.

Keywords: Phlebotomus caucasicus, Visceral leishmaniasis, Iran

Introduction

Leishmaniases are parasitic diseases of multifacted clinical manifestations, widespread in the old and new world with a great epidemiological diversity. At least 20 *Leismania* species are transmitted by female sand flies (*Phlebotomus* in the old world and *Lutzomyia* in the new world), which cause the disease. About 30 species of sandflies are proven vectors (1-2). Leishmaniases are important in terms of morbidity and some in terms of mortality. In man the disease occurs in at least four major forms: Cutaneous, mucocutaneous, diffuse cutaneous and visceral leishmaniasis. More than 90% of visceral leishmaniasis (VL) cases in the world were reported from Bangladesh, Brazil, India

and Sudan (1). Currently the global annual incidence of (VI) is estimated about 500000 cases (2). There are four important foci of VL in Iran including Ardebil, East Azerrbaijan (North West), Fars and Bushehr (South) provinces. The disease occurs sporadically in the other parts of the country. In East Azerbaijan more than 44 new cases of kala-azar have been reported in 2003(unpublished data). Eighteen out of mentioned cases (49%) belonged to Ahar (study area) district. The recent reports were indicated of increasing new cases of disease in East Azerbaijan as well as in Ahar (unpublished data). The last recent investigations on vectors of VL in Ardabil province (adjacent to East Azerbaijan) showed two species of P. kande-

¹Dept. of Medical Entomology and Vector Control, School of Public Health and Institute of Health Research, Tehran University of Medical Sciences, Iran

²Dept. of Medical Parasitology, School of Public Health and Institute of Health Research, Tehran University of Medical Sciences, Iran

lakii and *P. perfiliewi* were main probable vectors of the disease as they were found naturally infected by promastigotes(3-5). There was no classical study on vectors of VL in East Azerbaijan and this paper is the first one

Materials and Methods

Study area The study was carried out in Ahar county, north-west of Iran. Its Altitude is 1600 meter above the sea level. The total population of the county was about 158581 in 2002. The climatic condition was very hot (up to 40) in summer and quite cold (-20) during winter. The main activities of the people are agriculture and veterinary farming.

Identification of host preference The smear of blood meals of each engorged female sandflies was prepared on Whatman No.1 filter paper, which was then marked with the number of sandflies, date and place of collection. The papers were sent to the parasitology Department of Medical Parasitology and Mycology in Tehran University of Medical Sciences for ELISA testing (6).

Sand-flies collection Sand-flies were collected from indoors (bedroom, guestroom, toilet and stable) as well as outdoors (wall cracks and animal burrows) biweekly using sticky papers, CDC light traps and aspirators. Five villages called Aghakandi, Afil, Moradloo, Chinab and Navassar were selected and sand-flies were colleted during August and September (the activity peak of sand- flies) in 2002-2003. Sand-flies were dissected in a drop of normal salin. Theodor and Mesghali (1964) systematic identification key was employed for species identification (7).

Results

Totally 215 female sand-flies were collected and dissected. They were *Phlebotomus kandelakii* (57.25%), *P.perfiliewi* (18.1%), *P.caucasicus* (11.2%), *P.sregenti* (7.9%), *P.papatasi* (3.3%) and *P. (adlerius)* sp. (2.3%). Among the

dissected sand-flies, 65% were parous. Result showed that only 1 out of 24 *P.caucasicus* (4.4%) was infected by promastigotes. The activity of this species was started in early August and ended in early September. ELISA test on host preference of *P.caucasicus* showed that 25% of them were positive with dog antiserum and 75% with human.

Discussion

Among the dissected sandflies, the species of P.kandelakii with high density of population (57.25%) were collected during the activity of sand flies. This species has been reported as the probable vector of visceral leishmaniasis in Afghanistan (8). During the recent investigations in Meshginshahr district, north-west of Iran (The most important focus of VL in Iran) P.kandelakii was found naturally infected by promasigotes with high preference to man (4-5) Meshginshahr has located in adjacent to Ahar, so it seems that *P.kandelakii* may play a role as vector in Ahar. Naturally promastigote infection of *P.caucasicus* with pretty high density of population (11.2%) was the most important finding of this study. This species is proven vector of zoonotic cutaneous leishmaniasis in central Iran. Promastigote infection of P.caucasicus in this area without any reporting of cutaneous leishmaniasis cases and much interesting to human and dogs (the domestic reservoir of VL)was very wonderful and needs more future studies. This is the first record of promastigote infection of *P.caucasicus* in the north west of Iran.

Acknowledgements

The authors are grateful to authorities of Institute of Public Health Researches, Tehran University of Medical Sciences, Iran for supporting of this investigation and Mr. Zare the staff of Training and Health Station of Meshginshahr, for his assistance in the field work.

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