Iranian J. Publ. Health , 1995 , Vol. 24 , Nos. 1-2

PHLEBOTOMUS (LARROUSSIUS) KESHISHIANI SHCHURENKOVA 1936 ANOTHER VECTOR OF VISCERAL LEISHMANIASIS IN IRAN.

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Key words: *Visceral leishmaniasis , Phlebotomus , Iran.*

Abstract

In a sand-fly survey , conducted for two years (1992-93) in Ghir-Karzin , Fars province , a total of 1020 female *Phlebotomus keshishiani* were dissected , of which 12 females were found to have promastigotes. Promastigotes of six *Ph. keshishiani* were inoculated to six hamsters and one hamster became infected. Amastigotes were observed in the spleen , but the culture of parasite was not possible due to death of the hamster. Of 141 blood meals tested , 28.5% and 57.7% of *Ph.keshishiani* females were fed on human and dog respectively. This is the first report in the world about the role of *Ph.keshishiani* as a probable vector of thinfantile type of visceral leishmaniasis.

Introduction

Since 1949 , several reports have been published about human infection and reservoir hosts (13,15,17) of visceral leishmaniasis (VL) in Iran , but studies about vectors were limited and only on the basis of epidemiological evidence it was reported that *Phlebotomus major* is suspected to be the vector of VL (13).

In the last decade VL has become an important endemic disease in East Azerbaijan , in north west and in Fars province , in south of Iran(3,20).

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Confirmed cases of kala-azar have been reported sporadically and in endemic form from different regions of Iran. In Meshkin-shahr which is an endemic focus in the palearctic region , *Ph kandelakii* and *Ph.perfiliewi transcaucasicus* were found infected with flagellates (11). In the oriental region of Iran , where VL is endemic , the role of another probable vector *Ph. major* has been reported (18).

Phlebotomus keshishiani has a limited distribution in Pakistan (8) , Tajikestan (16) , Afghanistan (2,6,14) and Iran. In Iran , it has been found with a very low population in Fars (9). Bandar Abbass (10) , Gorgan (19) , Khuzistan (5,12) and Azerbaijan (1).

Investigation on vectors was continued during 1992-1993. The present paper shows the results of studies done on natural promatigote infection of *Ph.keshishiani* in the town of Ghir-karzin in Fars province.

Materials and methods

Five study sites were chosen within one km of the town of Ghir (53^0. 10' east longitude and 28^0.25' north latitude). Outdoor collections were made in 3 locations , common to these 3 sites were an altitude about 850 , presence of *Zizyphus vulgaris* and Proximity to rocky mountains. Two sites for indoor collections were chosen in courtyards on houses , where inhabitants used for sleeping. These two study sites , near the edge of rocky mountains , contained VL patients.

Two methods of trapping sand-flies were used: a) sticky paper traps, and b) CDC miniature light traps(21). Sticky paper traps were placed before dusk in the collection sites and were removed early in the next morning before sunrise. CDC miniature light traps were hung one meter above the ground. The traps were operated 30 minutes after sunset to 30 minutes before sunrise. Electricity for the traps was provided by a 6 volt battery. The gathered sand-flies were collected before sunrise. Living sand-flies were picked off the sticky paper traps and aspirated from collection bags. Blood fed and gravid females were dissected and examined microscopically for promastigote infection and identification of specimen.

In order to test the infectivity of promastigotes they were inoculated to hamsters. The hamsters were killed 3-4 months after inoculation to

examine their liver , spleen and bone marrow for parasites. Sand-flies used for blood meal identification were collected , using the CDC light trap.

Blood meals of identified sand-flies were smeard on whatman filter paper and dried at room temperature. They interleaved with non absorbent paper and were packed inside plastic bags.

Blood meals were tested by enzyme-linked immunosorbent assay (4) in the Protozoology unit , Department of Medical Parasitology and Mycology , School of Public Health.

Results and discussion

Twelve of 1020 (1.1%) *Ph.Keshishiani* dissected were infected with flagellates (Table 1). The infected sand-flies were collected only by the CDC light traps.

From our outdoor collections 850 females were dissected , of which 10 were infected , nine had promastigotes only in midgut and one in midgut , oesophagus and head. From the indoor collections 170 *Ph.keshishiani* were dissected and two of them had promastigotes infection in midgut and foregut. These two sand-flies with flagellate were collected from two houses with VL patients.

Flagellates from 6 infected flies were inoculated to hamsters (each to one animal). One of these 6 hamsters became infected. This hamster died 3 months after inoculation and amastigotes were observed in the spleen , but we were not able to isolate the parasite in the culture.

70 and 71 blood meals were tested against human and dog antisera , respectively. The results showed that 28.5% of the *Ph.keshishiani* females examined had fed on humans and 57.7% on dogs.

During our present study , *Ph.keshishiani* was collected with very high population and anthro population index in mountainous areas of Ghir , Fars province and Sohu , Bushehr provicne , where VL is endemic , indicating that feeding habits and host preference of *Ph.keshishiani* in this area enables it to be a good and suitable vector of VL.

The list of proven and suspected vectors of L.(L.) infantum includes 16 species (7,11,18,22). *Phlebotomus keshishiani* has not yet been reported to be a vector of leishmaniasis and this is the first report in the world about

the role of *Ph.keshishiani* as a probable vector of the infantile type of VL.

Acknowledgment

The authors gratefully acknowledge Mr. M. Kazemeini who carried out the field work., Dr. Nadim and Dr. Javadian for their critical suggestions during the period of this study and for reviewing this manuscript. Thanks also are due to Dr. Mesdaghinia , the Dean , and Dr. Ghiasseddin , Vice Dean of the School of Public Health , for their help in implementing this project.

Table 1- Natural promastigote infection of *Phlebotomus (L.) keshishiani* Shchurenkova 1936 , collected in town of Ghir (April 1992-January 1994).

Months	Outdoor Collection						Indoor Collection					
	Number	Number Positive					Number	Number Positive				
		G	O	H	T	%		G	O	H	T	%
Aprill 1992	20	0	0	0	0	0	0	0	0	0	0	0
May												
June	54	0	0	0	0	0	3	0	0	0	0	0
July	25	0	0	0	0	0	0	0	0	0	0	0
August	9	0	0	0	0	0	2	0	0	0	0	0
Sep	16	0	0	0	0	0	4	0	0	0	0	0
Oct	12	0	0	0	0	0	11	0	0	0	0	0
Nov	93	2	0	0	2	2.3	40	0	0	0	0	0
Dec	31	2	0	0	2	6.6	50	2	2	2	2	4
Feb 1993	45	1	1	1	1	0.5	28	0	0	0	0	0
March	1	0	0	0	0	0	0	0	0	0	0	0
May	2	0	0	0	0	0	0	0	0	0	0	0
June	97	0	0	0	0	0	0	0	0	0	0	0
July	15	0	0	0	0	0	1	0	0	0	0	0
Sep	99	0	0	0	0	0	6	0	0	0	0	0
Oct	85	0	0	0	0	0	5	0	0	0	0	0
Dec	108	5	0	0	5	4.6	17	0	0	0	0	0
Jan 1994	88	0	0	0	0	0	0	0	0	0	0	0
	50	0	0	0	0	0	3	0	0	0	0	0
Total	850	10	1	1	10	1.17	170	2	2	2	2	1.17

G: gut O: oesophagus H: head T: total

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