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PLEBOTOMINE VECTORS OF HUMAN DISEASE(U) FLORIDA UNIV
GAINESVILLE DEPT OF ENTOMOLOGY AND MENARTOLOGY
D G YOUNG 30 DEC 84 DAMD17-82-C-2223

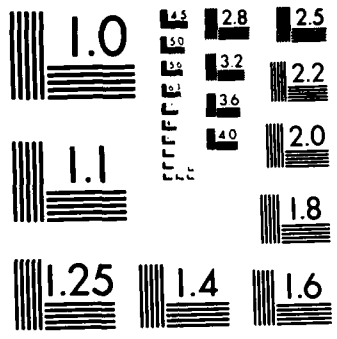
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PHLEBOTOMINE VECTORS OF HUMAN DISEASE

ANNUAL REPORT

DAMD 17-82-C-2223

By D.G. YOUNG

30 DECEMBER 1984

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Supported by

U. S. ARMY MEDICAL RESEARCH & DEVELOPMENT COMMAND
Fort Detrick, MD 21701 - 5012

DEPARTMENT OF ENTOMOLOGY & NEMATOLOGY
UNIVERSITY OF FLORIDA
Gainesville, Florida 32611

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REPORT DOCUMENTATION PAGE		READ INSTRUCTIONS BEFORE COMPLETING FORM
1. REPORT NUMBER	2. GOVT ACCESSION NO. ADA175046	3. RECIPIENT'S CATALOG NUMBER
4. TITLE (and Subtitle) Phlebotomine Vectors of Human Disease		5. TYPE OF REPORT & PERIOD COVERED Annual 30 Dec. 1983 - 30 Dec. 1984
		6. PERFORMING ORG. REPORT NUMBER
7. AUTHOR(s) D.G. Young		8. CONTRACT OR GRANT NUMBER(s) DAMD 17-82-C-2223
9. PERFORMING ORGANIZATION NAME AND ADDRESS University of Florida Gainesville, FL 32611		10. PROGRAM ELEMENT, PROJECT, TASK AREA & WORK UNIT NUMBERS
11. CONTROLLING OFFICE NAME AND ADDRESS U.S. Army Medical Research & Development Command Ft. Detrick, MD 21701-5012		12. REPORT DATE 30 Dec. 1984
		13. NUMBER OF PAGES
14. MONITORING AGENCY NAME & ADDRESS (if different from Controlling Office)		15. SECURITY CLASS. (of this report) Unclassified
		15a. DECLASSIFICATION/DOWNGRADING SCHEDULE
16. DISTRIBUTION STATEMENT (of this Report) Approved for public release; distribution unlimited		
17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)		
18. SUPPLEMENTARY NOTES		
19. KEY WORDS (Continue on reverse side if necessary and identify by block number) Sand fly <u>Lutzomyia</u> Leishmaniasis <u>Phlebotominae</u>		
20. ABSTRACT (Continue on reverse side if necessary and identify by block number) An extensive collection of Phlebotomine from Guatemala and Honduras was studied in 1984, in collaboration with Dr. C.H. Porter. A paper describing one new species was completed; others will treat new country records and additional new species. In Costa Rica, a highly anthropophilic and undescribed <u>verrucarum</u> group species was discovered in a coffee growing region where cutaneous leishmaniasis is endemic; other recently-collected sand flies from that country (<u>L. olmeca</u> complex) and from Panama were identified. The only female of <u>L. ovallesi</u> (also in the <u>verrucarum</u> group) captured at Ft. Sherman by WRAIR		

Investigators and dissected, had promastigotes in the hindgut. A hamster was inoculated with these parasites which may or may not prove to be Leishmania. An identification key to the sand flies of Tres Bracos, Brazil, a mucocutaneous leishmaniasis endemic site currently being studied by investigators at USAMRU-Brasilia, was completed. Two other papers dealing with biosystematics of South American Phlebotominae (Surinam and Bolivia) were also completed, during this time. Field work in Colombia yielded 35 species of Lutzomyia, one of which represents an undescribed species in the longispina species group. Of greater interest, was the discovery of natural promastigote infections in females of L. spinicrassa (verrucarum group) and L. erwindonaldoi (vexator group), both of which are avid man-biters in a leishmaniasis focus in NE Colombia. A laboratory colony of the former species and L. gomezi was established from females originating from this site (Arboledas).

An up-to-date summary of American sand flies, covering their medical importance, taxonomy, biology and other information, was written for general medical entomologists and is included in this report. A more detailed handbook on these species is in preparation; distribution maps were completed for each species and subspecies. The taxa will be treated in the format similar to that given for the species group baityi included in this report.

Additional phlebotomines from Tanzania, Brazil, Peru and Venezuela were slide-mounted and added to the reference collection. A paper on the laboratory colonization of Phlebotomus duboscqi, an important vector of cutaneous leishmaniasis in Africa, was submitted for publication.

For the first time, studies were begun to assess the ability of a USA sand fly species to maintain and transmit visceral leishmaniasis. Of 57 females of lab-bred L. diabolica that fed on a naturally infected dog, 51 (89.5%) subsequently became infected. Massive infections were observed. Twelve of the infected flies took second bloodmeals on laboratory hamsters. Results of these transmission trials will be reported later due to the long incubation period of

visceral leishmaniasis in hamsters.

Unclassified

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PERSONNEL SUPPORTED ON PROJECT

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QUALITY INSPECTED

ABSTRACT

1. Preparing Institution: University of Florida
2. Title of Report: Phlebotomine Vectors of Human Disease
3. Principal Investigator: David G. Young
4. 30 December 1984
5. Contract Number: DAMD 17-82-C-2223
6. Supported by: U.S. Army Medical Research and Development Command,
Fort Detrick, Frederick, MD 21701-5012

An extensive collection of Phlebotomine from Guatemala and Honduras was studied in 1984, in collaboration with Dr. C.H. Porter. A paper describing one new species was completed; others will treat new country records and additional new species. In Costa Rica, a highly anthropophilic and undescribed verrucarum group species was discovered in a coffee growing region where cutaneous leishmaniasis is endemic; other recently-collected sand flies from that country (L. olmeca complex) and from Panama were identified. The only female of L. ovallesi (also in the verrucarum group) captured at Ft. Sherman by WRAIR investigators and dissected, had promastigotes in the hindgut. A hamster was inoculated with these parasites which may or may not prove to be Leishmania. An identification key to the sand flies of Tres Bracos, Brazil, a mucocutaneous leishmaniasis endemic site currently being studied by investigators at USAMRU-Brasilia, was completed. Two other papers dealing with biosystematics of South American Phlebotominae (Surinam and Bolivia) were also completed during this time. Field work in Colombia yielded 35 species of Lutzomyia, one of which represents an undescribed species in

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PROGRESS REPORT

DAMD 27-82-C-2223

Introduction

This report covers the period from 30 Dec. 1983 to 30 Dec. 1984. Results of this research will be given according to the objectives that are listed below.

Objectives

1. To study the taxonomy of sand flies from the Neotropical Region, with special emphasis on the fauna of Mexico and Central America, and to provide a handbook on the entire fauna.
2. To study the taxonomy of sand flies from Africa and the Near East, providing identification keys, reference collections (based mainly on newly collected material); and to evaluate future needs for study in these areas.
3. To colonize in the laboratory, different species of Phlebotominae from Africa, the Near East and the Neotropical Region, with special emphasis on vector species.
4. To assess the susceptibility of colonized species to aetiologic agents of disease.

Results

1. Taxonomic studies of New World Phlebotominae

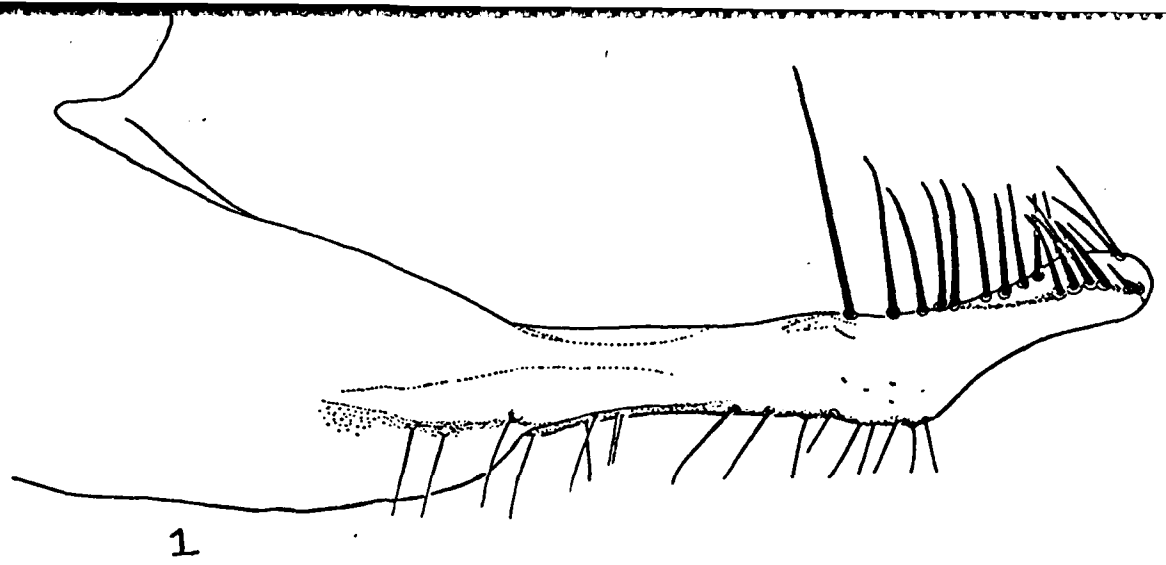
a. Central America

Examination of phlebotomines, collected by Dr. C.H. Porter, in Guatemala and Honduras in the early 1980's, revealed the presence of 4

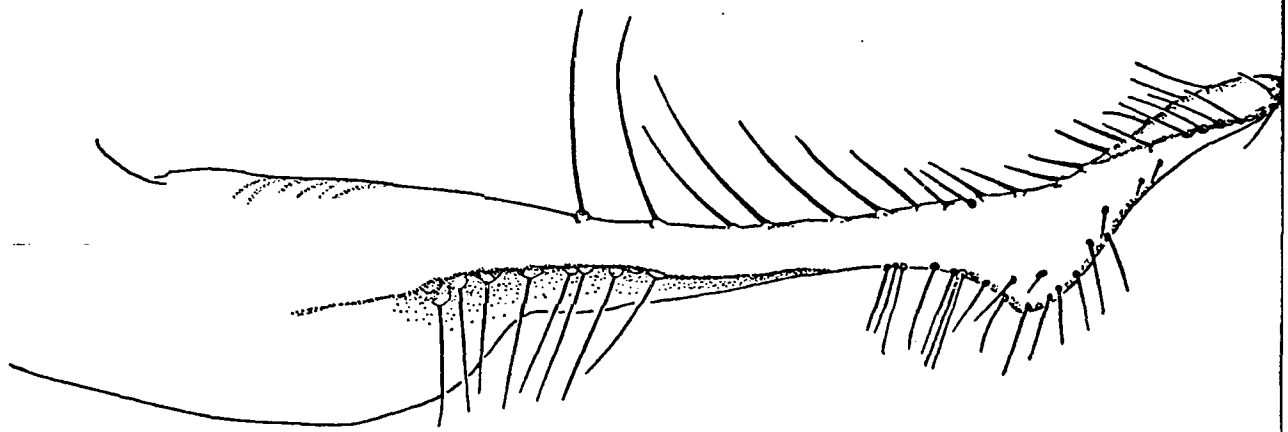
undescribed species. A paper (1) describing one of these species was completed in October. More than 10 other Lutzomyia species, previously unknown in Guatemala and Honduras, were identified. Further studies of the late Dr. deLeon's private collection of Guatemalan sand flies were made in collaboration with C.H. Porter in Feb. 1984. We concluded that L. edentula, L. piedraferroi and L. atulapai, all poorly described by deLeon, are valid species. Subsequent papers dealing with these and other Guatemalan sand flies should be completed in 1985.

A new focus of cutaneous leishmaniasis about 40 km S of San Jose, Costa Rica was discovered by Rodrigo Zeledón in 1984. The endemic area, about 1000 m above sea level, is mountainous. Houses are situated among coffee plantations that are common in the area. The PI was invited to Costa Rica in November to study sand flies from this and other foci with travel support from the UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases.

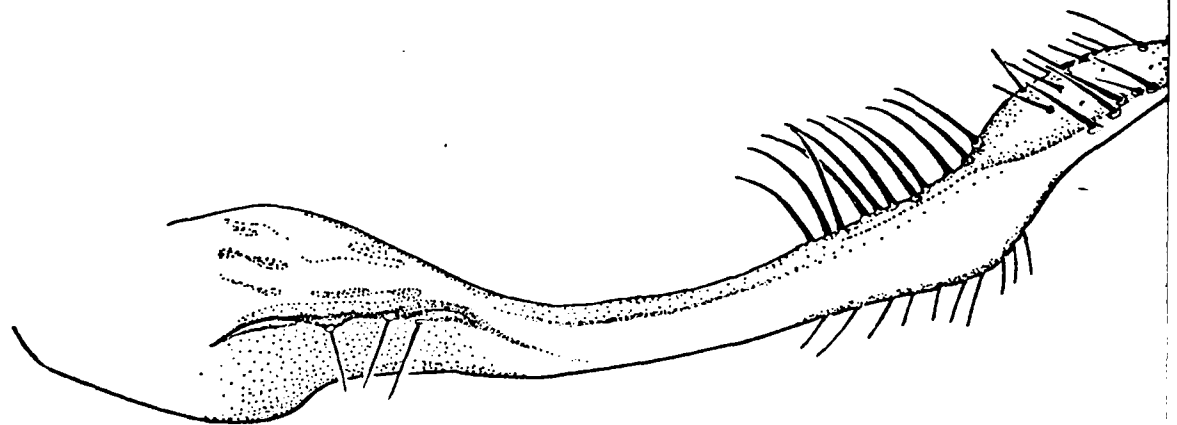
The most abundant anthropophilic sand fly at the newly discovered site is undescribed but is closely related to L. townsendi and L. quasitownsendi from Venezuela and Colombia, respectively. This discovery led to further studies of the 3 sibling species by the PI, Dr. Dora Pinero from Venezuela and J. Murillo from Costa Rica. Minor, but consistent differences, of the male parameres (Fig. 1-3), serve to distinguish these taxa. It was concluded that L. "townsendi" -- a species incriminated as the vector of cutaneous leishmaniasis in western Venezuela -- is not that species but instead is conspecific with the new species from Costa Rica. There are no records of any of these verrucarum group species in Panama so the geographic distribution of the new species, at least, appears to be disjunct.



1



2



3

Fig. 1-3. Parameres of *Lutzomyia n.sp.* (1), *L. townsendi* (2), *L. quasitownsendi* (3)

Transmission of leishmaniasis to humans apparently occurs in and near houses located among coffee groves in Costa Rica. Bloodfed females of the new species, collected by the PI and colleagues, were maintained alive in plastic vials for egg deposition in Costa Rica.

The PI examined females of the L. olmeca complex from Costa Rica. Members of this group have been incriminated as vectors of Leishmania mexicana among rodents and/or humans from Mexico to the Amazon Basin. Specimens referable to L. olmeca olmeca, identified by the PI, were collected only 30 km from the Costa Rica-Panama border on the Atlantic side; thus suggesting that populations of this subspecies and those of L. olmeca bicolor may meet in western Panama, not in Costa Rica or Nicaragua as was previously suspected.

In Panama at Fort Sherman where military personnel contracted cutaneous leishmaniasis in July, 1984, Lutzomyia panamensis was the most common sand fly collected in light traps or from human bait in Dec. 1984. Other anthropophilic species, identified by the PI, included L. trapidoi, L. ylephiletor, L. gomezi, L. bispinosa, W. rotundipennis and L. ovallesi. The latter species, represented by a single female from this Panama collection, is not considered a vector of leishmaniasis but promastigotes were found in this single female when dissected in Gainesville in December. The flagellates were inoculated into the nose and one foot of a hamster. Results of this inoculation will not be known for several months.

b. South America

A short paper with J. Hudson on new records of phlebotomines from Surinam (2) was completed and submitted to the Trans. R. Soc. Trop. Med. during this period.

A longer publication (3) on the phlebotomine fauna of Bolivia with H. Bermudez was also completed. Twelve species are recorded for the first time in the Republic. Locality records for these and all others are given. The sand fly fauna of Bolivia remains poorly known but continued collaboration with Dr. Bermudez, a Bolivian who spent 3 months in the PI's laboratory in 1984, should add considerable information in the next few years.

A paper on the sand flies of Ecuador was published this year, and another dealing with the entire Peruvian phlebotomine fauna was accepted by the Intl. J. Ent. for publication (April, 1985 issue). One of the new Lutzomyia species from Peru belongs in the newly created microps group, a review of which will be published in the Mem. Oswaldo Cruz in 1985.

In Colombia, with the support of this and a NIAID contract, over 35 species of sand flies were collected from April to May and August, 1984. An undescribed species related to L. longispina was captured at Arboledas in Norte de Santander Department where human cutaneous leishmaniasis is common. The previously unknown female of L. erwindonaldi, was also taken and it and L. spinicrassa (verrucarum group) were noted for the first time to be highly anthropophilic. Promastigotes were found in one wild-caught female of the former species; in 2 females of L. spinicrassa, and in 1 female of L. gomezi. Characterization of these flagellates will be made following results of hamster inoculations in Gainesville.

c. General

A book chapter on phlebotomine sand flies in the Americas, supported by this contract, was completed during this period. Because it will be translated into Spanish for a PAHO publication, it is included

here in English (4) as an up-to-date summary that may be helpful to medical entomologists.

All of the distribution maps for each described Lutzomyia species were completed. These will be included in the handbook of American sand flies that should be finished in 1985. A sample format for treating a species group is given in Appendix V.

2. Old World sand flies

A paper on the laboratory colonization of Phlebotomus duboscqi from Kenya was submitted to the Trans. R. Soc. Trop. Med. Hyg. (with R.F. Beach and G. Kiilu). Additional sand flies from Tanzania (collected by the PI in 1982) were slide mounted, but no new material from the Old World was studied this year.

3. Colonization attempts

From sand flies we collected in Colombia in 1984, two species were successfully colonized in the laboratory at Yale University. Both species, L. gomezi and L. spinicrassa, are now suspected vectors of cutaneous leishmaniasis at Arboledas, Colombia. The F₁ generation females deposited about 1000 eggs so in subsequent generations, enough material should be available for subcolonization attempts at WRAIR. Natural promastigote infections were found in dissected females of these species from Arboledas. Lutzomyia spinicrassa has not been previously colonized and there are no other existing laboratory colonies of L. gomezi, a more widespread species.

Laboratory colonies of L. anthophora and L. vexator continue to be maintained at the University of Florida.

4. Vector potential of sand flies

The ability of sand flies from the USA to maintain and transmit

Leishmania causing visceral disease has not been evaluated. Infected dogs imported from endemic areas in other countries are reported each year in the USA; and, near Oklahoma City, Oklahoma, there is a focus of canine leishmaniasis that may have originated from imported dogs.

On 4 June 1984 a 7 year old golden retriever dog was admitted to the Veterinary Medical Teaching Hospital, University of Florida. The dog had previously lived in Sicily where it had presumably contracted Leishmania. Amastigotes, believed to be Le. donovani infantum, were abundant in the bone marrow and skin. A submitted paper by M. Shaer, D.J. Meyer and D.G. Young (1985, a dual infection of Leishmania donovani and Erichia canis in a dog) will give additional clinical details of this case.

Fifty-seven lab-bred L. diabolica sand flies from our colony fed on the stomach, back or right ear, of this dog on 7 June 1984. This anthropophilic sand fly occurs in Texas and Mexico.

Fifty-one (89.5%) of these flies, when dissected 3 to 11 days following the infecting bloodmeal, were positive for leishmanial promastigotes. Massive infections were observed at the stomodeal valve beginning on day 6 post feed (at 27°C). Twelve of 51 infected flies survived oviposition to take second bloodmeals on hamsters on days 6 to 11 after the infecting bloodmeal. Results of these transmission attempts will be given in a later report due to the long incubation period of visceral leishmaniasis in these animals. However, the high proportion of L. diabolica females that became infected after feeding on this dog was unexpected and suggests that this species may be a suitable host for Leishmania donovani under certain conditions.

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4. Young, David G. Phlebotomine Sandflies in the Americas.

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