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Vector Biology
and Control Project

Trip Report:
Training Course on Sand Fly and *Leishmania* Surveillance in Nepal

March 20-April 19, 1992

by

Phillip G. Lawyer, Ph.D.

VBC Report No. 81324

Managed by Medical Service Corporation International • Sponsored by the Agency for International Development
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Author

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Acknowledgments

The author wishes to thank Dr. Richard Andre, Vector Biologist, Vector Biology and Control Project, and Dr. Jerald Sadoff, Director, Division of Communicable Diseases and Immunology, Walter Reed Army Institute of Research, for the opportunity to participate in this activity. Thanks also go to Dr. B.L. Shrestha, Chief, Malaria Control Division, and Mr. Riddhi G. Vaidya, Chief Entomology Section, National Malaria Eradication Division, Nepal Ministry of Health, for their cooperation and in-country support, and for the latter's participation in and assistance with the course. Finally, special thanks go to Mr. Shreedhar P. Pradhan, Medical Entomologist/Malaria Advisor, Office of Health and Family Planning, USAID/Kathmandu, whose close coordination, organization, management and translating skills ensured the success of the course.

Preparation of this document was sponsored by the Vector Biology and Control Project under Contract No. DPE-5948-C-00-9030-00 to Medical Service Corporation International, Arlington, Virginia, USA, for the Agency for International Development, Office of Health, Bureau for Research and Development.

Executive Summary

His Majesty's Government of Nepal, through the Ministry of Health's Malaria Control Division, requested assistance from USAID/Kathmandu in providing training in sand fly and *Leishmania* surveillance for control of kala azar transmission. In response to this request, VBC asked the author to serve as a consultant to organize and present a training course.

The goal of this activity was to provide training that would enable the Nepal Ministry of Health (MOH) to investigate an epidemic focus of visceral leishmaniasis (kala azar) in the eastern half of the Terai plain.

The author arrived in Nepal March 22, 1992, three days before the course began to allow for initial briefings and logistical preparations, and departed the country April 19 following appropriate debriefings (see p. 17 for detailed itinerary). An intensive training course was conducted in Janakpur Dham, Dhanusha District, Central Region, from March 25 to April 15, 1992, with the able and much appreciated assistance of Mr. Shreedhar Pradhan and Mr. Riddhi Vaidya.

Instruction was presented in two phases: Phase I, March 26 to 31, intensive classroom instruction and laboratory and field demonstrations; Phase II, March 31 to April 15, practical field experience in designing, conducting, and evaluating sand fly and *Leishmania* surveys. Twenty-five students from the Nepal Ministry of Health's Malaria Control and Epidemiology Divisions participated in the course. Participants worked very diligently in every aspect of the course. All passed and are now qualified to conduct sand fly and *Leishmania* surveillance.

During the Phase II practical field experience, the students conducted parallel surveys in three villages in the vicinity of Janakpur. While processing specimens from 266 separate collections, they gained extensive experience in identifying sand flies, dissecting and examining them for parasites, and keeping and interpreting collection records.

Using a variety of methods, training course participants collected nearly 5,000 sand flies, representing three species. The relative abundance of females by species was 21 percent *Phlebotomus argentipes,* 68 percent *P. papatasi* and 11 percent *Sergentomyia babu.* In a 10-day survey of resting sites in the three villages, only 11 percent of female *P. argentipes* were collected inside human dwellings. Fifty percent were collected inside mixed dwellings where people and animals were living under the same roof and 39 percent were found inside animal dwellings. By comparison, 54 percent of *P. papatasi* females were collected inside human dwellings, 17 percent in animal dwellings and 29 percent in mixed dwellings.

Approximately 2,000 females (550 *P. argentipes* and 1,400 *P. papatasi*) were dissected and examined by light microscopy for *Leishmania*-like flagellates. Infections in two *P. argentipes* (0.4 percent) were confirmed by whole kinetoplast-DNA probes as *L. donovani*. This finding represents the first confirmation of *L. donovani* infections in *P. argentipes* in Nepal and illustrates the utility of this bio-molecular diagnostic technique in field studies. *Leishmania* infections were not found in 1,400 *P. papatasi* dissected and examined.

Host blood-meal ELISA results on 492 blood meals of feral flies indicate that while *P. argentipes* are reported to be strongly zoophilic, in mixed dwellings they are in fact rather opportunistic feeders and feed about equally on domestic animals (cattle) and humans. Thus, humans who sleep under the same roof as their livestock are at much greater risk of being bitten by infected *P. argentipes* than those who sleep in separate dwellings. *P. papatasi*, on the other hand, were found to be strongly anthropophilic and should not be ruled out as potential vectors of kala azar.

Based on this initial training visit, the following actions are recommended:

1. Draft a detailed proposal and seek financial and administrative support for a two-year longitudinal survey of *Leishmania* vectors in kala azar-endemic areas of Nepal.

2. Conduct a follow-up training session September 1-30, 1992 (including travel) to augment and reinforce the initial training and to organize the proposed longitudinal survey.

3. Conduct laboratory experiments to investigate the vectorial competence of *P. argentipes* and *P. papatasi* for *L. donovani.*

4. Continue and intensify the existing program of residual spraying of human dwellings in affected areas and also include spraying of mixed and strictly animal dwellings.

5. Initiate a program to educate the general public in affected areas about factors that contribute to epidemic transmission of kala azar and ways to reduce contact with the vectors (such as sleeping away from their animals).

6. Initiate a program of active case finding in villages where index cases have been identified through passive detection.

1. Introduction

A World Health Organization (WHO) information release dated March 24, 1990, reported that "Leishmaniasis currently infects 12,000,000 people in the 80 [affected] countries with an additional 90,000,000 at risk."[1] Because this complex of diseases most severely affects the poorest and most underprivileged peoples of the world, support is generally lacking for leishmaniasis prevention and control programs.

Although visceral leishmaniasis has probably been endemic in Nepal for many years, it was first recorded in the country in 1980 and is presumed to have been introduced during population migrations from India.[2] Information about its current status in Nepal is rather limited.[3] Recent reports (personal communications, Dr. Richard Andre and Mr. Shreedhar Pradhan) and observations made during this training activity indicate that since 1987, the number of cases of kala azar in rural villages of the eastern Terai plain has increased sharply (Figures 1 and 2). This resurgence is attributed to the cessation of routine house spraying for malaria control with DDT, which was evidently effective against sand flies as well as mosquitoes.

There is a need to investigate current trends in leishmaniasis transmission in rural villages in Nepal, with a view to developing practical, low-cost, community-based kala azar surveillance, prevention and control programs. Little is known of the diversity and distribution of sand fly species in the country. Prior to this activity, *P. argentipes* was the presumed but unconfirmed vector of kala azar in the Terai.

---

[1] Anonymous, 1990. WHO Office of Information Issues, "World Report on Tropical Diseases." **Trop. Med. Hyg. News**, June 1990: 72.

[2] Joshi, D.D., Shresta, J.D., Pradhan, S.P., Joshi, A.B., 1990. Kala-azar in Morang District: Epidemiological situation. **J. Inst. Med.** 12: 205-209.

[3] Anonymous, 1984. The Leishmaniases. Report of a WHO Expert Committee. Technical Report Series 793. WHO, Geneva.

Figure 1
Trend of Case Incidence and Case Fatality Graph

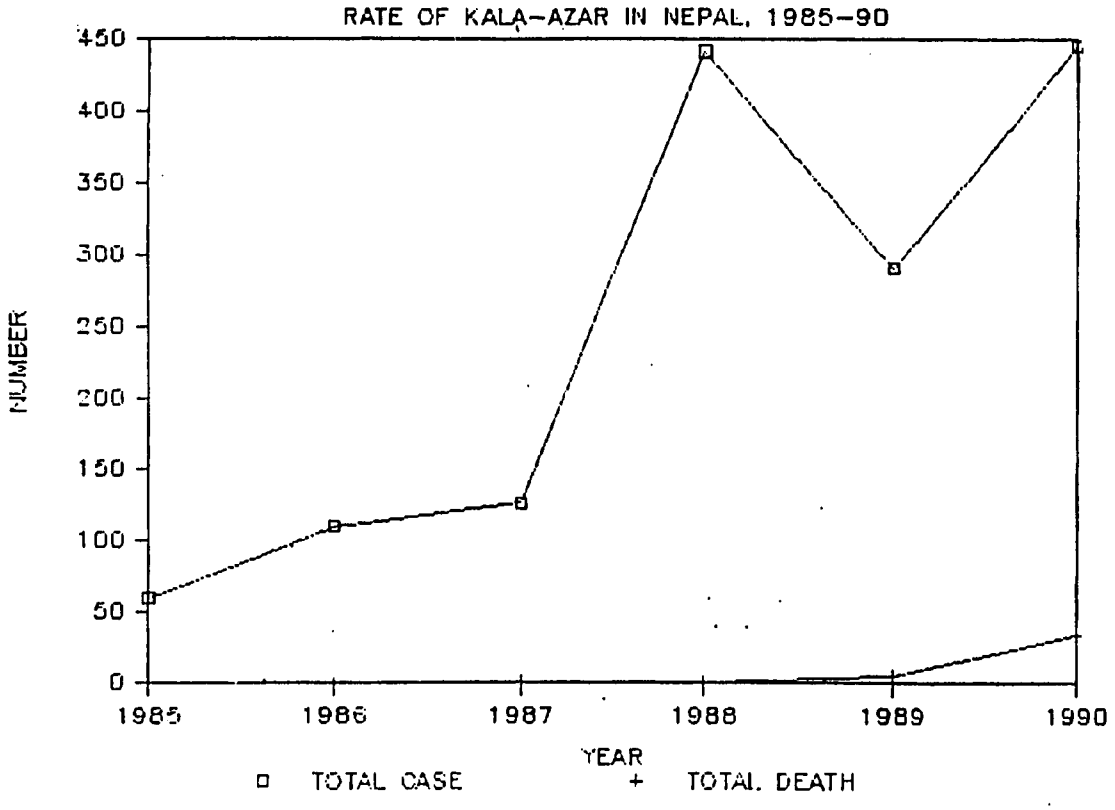
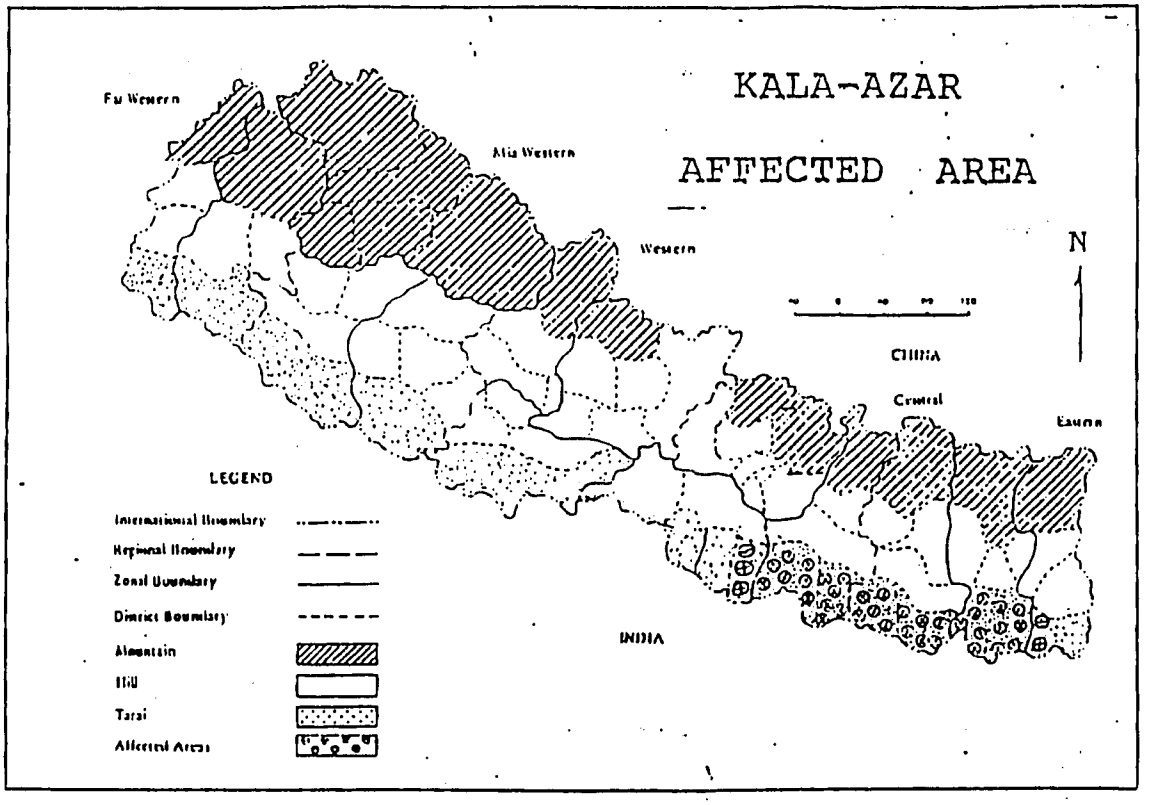


Figure 1. Trend of kala azar case incidence and fatalities in Nepal, 1985-1990.

Figure 2
Distribution of Kala azar Cases



Prepared by: S.P. Pradhan
HFP, USAID/N

The purpose of this activity was to assist Nepal's Ministry of Health in investigating an epidemic focus of visceral leishmaniasis (kala azar) in the eastern Terai plain. Specific objectives were to:

1. train a cadre of entomologists and epidemiologists in identification and surveillance of sand flies and in techniques for investigating the epidemiology of leishmaniasis transmission;

2. initiate a longitudinal sand fly and *Leishmania* survey; and

3. recommend practical prevention and control measures that could be implemented by local public health workers to reduce human-sand fly contact and thus the incidence of kala azar.

The training visit began March 20, 1992, and continued through April 19, 1992. The scope of work included the following steps:

1. obtaining and preparing training materials;

2. presenting an intensive course on identification of vector sand flies in Nepal and on epidemiological survey methods for leishmaniasis, including laboratory and field techniques;

3. initiating preparation of a reference teaching collection of the sand flies of Nepal to be used at the Hetauda Research and Training Center;

4. assisting the Ministry of Health in establishing a program for monitoring leishmaniasis transmission risk by determining vector parameters (e.g., species abundance, host preference, resting behavior, and natural longevity), infection rates in sand flies, and human prevalence of leishmaniasis in selected villages;

5. providing information to the Ministry of Health on possible diagnostic procedures for identifying *Leishmania* in sand flies and mammalian hosts; and

6. assisting the Ministry of Health in developing possible leishmaniasis control strategies.

2. Findings and Results

The course was presented in English with simultaneous translation provided as needed by Mr. Pradhan and Mr. Vaidya. With a few modifications, the classroom facility rented from the Janakpur Public Health Office adequately accommodated the lecture and laboratory portions of the training course. Ten compound and 10 stereoscopic dissecting microscopes, as well as some chemicals, were provided by the Malaria Control Division. A portable, gasoline-powered electricity generator, also furnished by the Malaria Control Division, was invaluable for operating the microscope illuminators during daily power outages.

Instruction was presented in two phases: Phase I, March 26 to 31, intensive classroom instruction and laboratory and field demonstrations; Phase II, March 31 to April 15, practical field experience in designing, conducting, and evaluating sand fly and *Leishmania* surveys. The course was introduced with a written pre-examination and culminated in practical and written final examinations. A detailed training schedule is included as Annex A. Twenty-five students from the MOH's Malaria Control and Epidemiology Divisions participated in the course (Annex B). Participants worked very diligently in every aspect of the course. All passed and are now qualified to conduct sand fly and *Leishmania* surveillance.

During the Phase II practical field experience, the students selected three villages in the vicinity of Janakpur in which to conduct parallel surveys. They inspected each village, determined the best collection methods for the situation and selected specific trapping/collecting sites. Collection sites were: 1) inside human dwellings; 2) inside animal dwellings; 3) inside mixed dwellings (animal and human); and 4) outside in peridomestic locations.

Using a variety of methods, training course participants collected nearly 5,000 sand flies. The overall relative abundance of females by species was 25 percent *P. argentipes,* 63 percent *P. papatasi* and 12 percent *S. babu* (Table 1). In a 10-day survey of resting sites in the three

Table 1
Summary of Sand Fly Collections in Janakpur Area

Species	No. Female	% Total Female	No. Male	% Total Male
P. argentipes	703	25	337	16
P. papatasi	1,726	63	1,645	76
Sergentomyia	330	12	197	9
Total	2,759		2,179	

villages, the following observations on relative abundance and distribution of local sand fly populations were made (Tables 2 & 3.):

1. *P. argentipes* were most abundant in mixed dwellings where animals and humans slept in close proximity (50 percent of all female *P. argentipes* collected), followed by strictly animal dwellings (39 percent) and human dwellings (11 percent). This species predominated in strictly animal dwellings. Blood-fed females were collected in abundance with mouth aspirators (sucking tubes) in the early mornings and late afternoon while resting on the interior walls of all three types of dwelling. The density of flies in exterior collections was very low. *Phlebotomus argentipes* were collected in biting/landing collections from humans and animals both inside and outside of dwellings throughout the night.

2. *P. papatasi* were found in greatest abundance in human dwellings (54 percent of all female *P. papatasi* collected). This species was also abundant in mixed dwellings (29 percent), but less abundant in animal dwellings (17 percent). As with *P. argentipes*, blood-fed *P. papatasi* were collected with mouth aspirators in early mornings and late afternoons while they rested on the interior walls of all three types of dwelling. Exterior biting density was also very low, and *P. papatasi* were collected throughout the night in biting/landing collections from humans and animals.

3. A nonvector species, *Sergentomyia babu*, was collected in greatest abundance in mixed dwellings (62 percent of all female *S. babu* collected), but was also found in human dwellings (19 percent) and in strictly animal dwellings (19 percent). Blood-fed *S. babu* were found resting on the walls of human dwellings, as well as in strictly animal and mixed dwellings, indicating that this species may bite humans.

While processing specimens from 266 separate collections, the students gained extensive experience in identifying sand flies, dissecting and examining them for parasites, and in keeping and interpreting collection and dissection records. Approximately 2,000 females (550 *P. argentipes* and 1,400 *P. papatasi*) were dissected and examined by light microscopy for *Leishmania*-like flagellates. Six *P. argentipes* and one *P. papatasi* were found with suspected flagellates in their midguts. One of

Table 2

Relative Abundance of Sand Fly Species Collected at Resting Sites*

	Type of Dwelling											
	Human				Animal				Mixed			
Sand Fly Species	No. Female	% Female	No. Male	% Male	No. Female	% Female	No. Male	% Male	No. Female	% Female	No. Male	% Male
P. argentipes	86	8	39	4	268	49	88	25	288	35	117	21
P. papatasi	875	81	839	89	228	42	235	67	407	50	342	63
Sergentomyia	121	11	64	7	48	9	30	9	123	15	88	16
Totals	1,082		942		544		353		818		547	

*Collected in the vicinity of Janakpur Dham, Dhanusha District, Nepal, April 1-14, 1992.

Table 3
Distribution of Sand Flies by Species in Human, Animal and Mixed Dwellings*

Phlebotomus argentipes

Habitat	No. Female	% Female	No. Male	% Male
Human	60	11	27	13
Animal	210	39	72	34
Mixed	265	50	113	53
Total	535		212	

Phlebotomus papatasi

Habitat	No. Female	% Female	No. Male	% Male
Human	598	54	601	55
Animal	191	17	193	18
Mixed	327	29	281	27
Total	1,116		1,075	

Sergentomyia babu

Habitat	No. Female	% Female	No. Male	% Male
Human	59	29	26	19
Animal	38	19	25	19
Mixed	106	52	83	62
Total	203		134	

*Collected during 10 days of parallel resting site surveys in three rural villages in the vicinity of Janakpur Dham, Dhanusha District, Nepal.

the *P. argentipes* had a massive mature infection of *Leishmania* flagellates that packed the midgut and blocked the juncture between the esophagus and the stomodeal valve (opening to the thoracic midgut). This infection included metacyclic (infective-stage) parasites.

Samples of midgut contents of these putatively infected flies were preserved on nylon paper and/or dried on microslides for kinetoplast-DNA hybridization and enzyme-linked immunosorbent assay (ELISA) using monoclonal antibodies. *Leishmania donovani* infections were confirmed in two of the *P. argentipes* females by hybridization techniques using whole kinetoplast-DNA probes. This finding represents the first confirmation of *L. donovani* infections in *P. argentipes* in Nepal and illustrates the utility of this bio-molecular diagnostic technique in field studies. *Leishmania* infections were not found in 1,400 *P. papatasi* dissected and examined.

Almost 500 feral, blood-fed flies, collected inside dwellings, were squashed on filter paper to preserve the blood meals, which were later identified by ELISA (Table 4). Results of blood-meal analyses show that both *P. argentipes* and *P. papatasi* are endophagic and endophilic. The former is more zoophilic and opportunistic, however, and the latter highly anthropophilic. In mixed dwellings, *P. argentipes* feed on humans with about the same frequency as they feed on cattle. Thus, humans who sleep in the same dwellings as their livestock are at much greater risk of being bitten by infected *P. argentipes* than if they sleep in a separate dwelling. About 11 percent of blood-fed *P. argentipes* showed evidence of multiple-source blood meals (usually human and bovine), compared to 5 percent in *P. papatasi*. *P. papatasi* were found to be strongly anthropophilic, but showed a tendency to move to other dwellings to rest after a blood meal, as evidenced by the high proportion of these flies with human blood meals found resting in animal dwellings.

Table 4
Results of Blood-Meal ELISA Run on 492 Blood Meals From Feral Sand Flies*

Phlebotomus argentipes

		Blood Meal Identification					
Dwelling Type	**n**	**H (%)**	**HB (%)**	**B (%)**	**S (%)**	**HS (%)**	**? (%)**
Animal	61	2(3)	11(18)	47(77)	---	---	1(1)
Human	14	5(36)	----	7(50)	---	---	2(14)
Mixed	81	36(44)	4(5)	38(47)	---	1(1)	2(3)
Total	156	43(28)	15(10)	92(59)	---	1(1)	5(3)

Phlebotomus papatasi

		Blood Meal Identification					
Dwelling Type	**n**	**H (%)**	**HB (%)**	**B (%)**	**S (%)**	**HS (%)**	**? (%)**
Animal	31	25(81)	2(7)	4(13)	---	---	---
Human	168	156(93)	3(2)	7(4)	---	---	2(1)
Mixed	121	99(82)	9(7)	10(8)	---	3(3)	---
Total	320	280(88)	14(4)	21(7)	---	3(1)	2(1)

*Collected in rural villages in the vicinity of Janakpur Dham, Nepal.

Legend:

n = Total number of flies of a species from a particular dwelling type
H = Human
HB = Human and bovine
B = Bovine
S = Sheep or goat
HS = Human and sheep or goat
? = Unidentified

Table 4
Results of Blood-Meal ELISA Run on 492 Blood Meals From Feral Sand Flies (Continued)*

Sergentomyia babu

		Blood Meal Identification					
Dwelling Type	**n**	**H (%)**	**HB (%)**	**B (%)**	**S (%)**	**HS (%)**	**? (%)**
Animal	8	1(13)	---	1(13)	1(13)	---	5(63)
Human	2	---	---	---	---	---	2(100)
Mixed	6	---	---	---	---	---	6(100)
Total	16	1(6)	---	1(6)	1(6)	---	13(82)

*Collected in rural villages in the vicinity of Janakpur Dham, Nepal.

Legend:

n = Total number of flies of a species from a particular dwelling type
H = Human
HB = Human and bovine
B = Bovine
S = Sheep or goat
HS = Human and sheep or goat
? = Unidentified

3. Recommendations

The following actions are recommended as a result of this initial training visit:

1. Draft a detailed proposal and seek financial and administrative support for a two-year longitudinal survey of *Leishmania* vectors in kala azar-endemic areas of Nepal. This should be a joint proposal between USAID, associated consultants, and the Nepal Ministry of Health, Malaria Control and Epidemiology Divisions.

2. Conduct a follow-up training session September 1-20, 1992, (including travel), to augment and reinforce the initial training and to organize the proposed longitudinal survey. This training should include instruction on specific methodologies for locating vector breeding sites and determining post-feeding dispersion behavior.

3. Conduct laboratory experiments to investigate the vector competence of *P. argentipes* and *P. papatasi* for *L. donovani*. This work can begin at the Department of Entomology, Walter Reed Army Institute of Research, but should also be conducted at the Hetauda Training Center after construction of the center has been completed.

4. Continue and intensify the existing program of residual spraying of human dwellings in affected areas and also include spraying of mixed and strictly animal dwellings. Assess the efficacy of spraying activities with follow-up sand fly population density surveys.

5. Initiate a program to educate the general public in affected areas about factors that contribute to epidemic transmission of kala azar and ways to reduce contact with the vector (such as sleeping away from their animals).

6. Initiate a program of active case finding in villages where index cases have been identified through passive detection.

4. Itinerary and People Contacted

March 22 - April 18, 1992

Date	Time	Particular
03/22/92	09:55	Arrived in Kathmandu from Washington.
	13:30	Met with Shreedhar P. Pradhan, Medical Entomologist/Malaria Advisor, Division of Health and Family Planning, USAID/Kathmandu, to discuss the day-to-day course schedule.
03/23/92	08:30	Met and briefed with Mr. Matthew Friedman, Technical Advisor, and Mr. S.P. Pradhan, Office of Health and Family Planning, USAID/Kathmandu.
	10:30	Met wth Dr. Badri L. Shrestha, Chief, Malaria Control Division, and staff, accompanied by Mr. S. P. Pradhan, Office of Health and Family Planning, to discuss training courses.
	13:00	Attempted to meet with Dr. Yogendra M.S. Pradhan, Additional Secretary, MOH, to brief him on the training course, accompanied by S.P. Pradhan, Office of Health and Family Planning. Meeting was cancelled due to other commitments of the Additional Secretary.
	15:00	Prepared a list of supplies for local procurement.
03/24/92	all day	Procured supplies. Packed and rechecked supplies and equipment brought from the USA and those that were purchased locally. Made arrangements for proceeding to the Dhanusha training site.

Date	Time	Particular
03/25/92	08:30	Proceeded to the Dhanusha training site accompanied by S.P. Pradhan, Office of Health and Family Planning, and Riddhi Vaidya, Malaria Control Division.
	20:30	Arrived in Janakpur Dham.
3/26/92 - 4/15/92		Conducted the training course.
4/15/92	15:00	Departed Janakpur for Kathmandu.
	20:00	Arrived in Hetauda; toured Hetauda Training Center.
04/16/92	05:30	Departed Hetauda for Kathmandu.
	15:30	Arrived in Kathmandu.
04/17/92	09:00	Held debriefing with Dr. Matthew Friedman, Technical Advisor, Office of Health and Family Planning, USAID/Kathmandu.
	10:00	Held debriefing with Theodora Wood-Stervinou, Deputy Mission Director, USAID/Kathmandu, accompanied by S.P. Pradhan, Office of Health and Family Planning.
	10:45	Held debriefing with Dr. Badri L. Shrestha, Chief, Malaria Control Division, and his senior staff; discussed a follow-up trip in September 1992.
	13:15	Planned to debrief with Dr. Yogendra M.S. Pradhan, Additional Secretary, MOH, accompanied by S.P. Pradhan, Office of Health and Family Planning. Meeting cancelled due to other committments of the Additional Secretary.
	14:00	Packed equipment for return to USA.
04/18/92	1:55	Departed Kathmandu for Washington, D.C.

Annex A

Nepal Leishmaniasis Project Sand Fly and *Leishmania* Surveillance Training Schedule

Thursday, March 26

AM	10:00 - 11:00	Registration/Welcome/Orientation
	11:00 - 11:30	Pre-test on material to be covered
	11:30 - 11:40	Break
	(Beginning of Phase I)	
PM	11:40 - 01:00	Sand Fly Morphology/Classification/Biology
	01:00 - 02:00	Lunch
	02:00 - 03:00	Medical Importance of Sand Flies/Distribution of Leishmaniases
	03:00 - 03:10	Break
	03:10 - 05:00	Population Dynamics/Factors Affecting Rates of Transmission/*Leishmania* Development in Sand Flies

Friday, March 27

AM	10:00 - 11:00	Factors Affecting Rates of Transmission
	11:00 - 11:30	Criteria for Incriminating a Vector
	11:30 - 11:40	Break
	11:00 - 01:00	Sand Fly Surveillance Methods
PM	01:00 - 02:00	Lunch
	02:00 - 03:00	Processing Collected Material/Record Keeping
	03:00 - 03:10	Break
	03:00 - until*	Sand Fly Surveillance Methods (Setting traps, bite-landing collections)

*Times varied.

Saturday, March 28

AM	06:00 - 08:00	Sand Fly Surveillance Methods (retrieving traps, resting-site collections)
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing collected material

Sunday, March 29

AM	10:00 - 11:30	Specimen preservation, clearing and mounting techniques
	11:30 - 11:40	Break
PM	11:40 - 01:00	Specimen preservation, clearing and mounting techniques
	01:00 - 02:00	Lunch
	02:00 - 03:00	Sand Fly Taxonomy (using a dichotomous key structural terminology)
	03:00 - 03:00	Break
	03:10 - 05:00	Sand Fly Taxonomy
	05:00 - until*	Sand Fly Surveillance Methods

Monday, March 30

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 02:30	Breakfast
	08:30 - until*	Processing Collections/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping
PM	01:00 - 02:00	Lunch
	02:00 - 02:30	Host Preference Studies
	02:30 - 03:00	Mass-rearing of Sand Flies
	03:00 - 03:10	Break
	03:10 - 04:00	Prevention and Control
	04:00 - until*	Sand Fly Surveillance Methods

Tuesday, March 31 (Beginning of Phase II)

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30: - until*	Processing Collections/Sand Fly dissection and Examination/Sand Fly Taxonomy/Record Keeping
PM	01:00: - 02:00	Lunch
	02:00 - 03:00	Selection of a Study Site (Desk study, on-site inspection, survey design and layout)
	03:00 - 03:10	Break
	03:10 - 04:00	Selection of a Study site
	04:00 - until*	Sand Fly Surveillance Methods

Wednesday, April 1

AM	01:00 - 02:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Brea -fast
	08:30 - until*	Processing Collections/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping
PM	01:00 - 02:00	Lunch
	04:00 - until*	Sand Fly Surveillance Methods

Thursday, April *2*

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing Collection/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping
PM	01:00 - 02:00	Lunch
	04:00 - until*	Sand Fly Surveillance Methods

Friday, April 3

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing Collections/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping
PM	01:00 - 02:00	Lunch
	04:00 - until*	Sand Fly Surveillance Methods

Saturday, April 4

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing Collection/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping

Sunday, April 5

PM	04:00 - until*	Sand Fly Surveillance Methods

Monday, April 6	Sand Fly Surveillance Methods
Tuesday, April 7	Sand Fly Surveillance Methods
Wednesday, April 8	Sand Fly Surveillance Methods
Thursday, April 9	Sand Fly Surveillance Methods
Friday, April 10	Holiday (Participation optional)

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing Collection/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping

Saturday, April 11

PM	04:00 - until*	Sand Fly Surveillance Methods

Sunday, April 12

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing Collection/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping

Monday, April 13 Holiday (Participation Optional)

	04:00 - until*	Sand Fly Surveillance Methods

Tuesday, April 14

AM	06:00 - 08:00	Sand Fly Surveillance Methods
	08:00 - 08:30	Breakfast
	08:30 - until*	Processing Collection/Sand Fly Dissection and Examination/Sand Fly Taxonomy/Record Keeping
PM	01:00 - 02:00	Lunch
	04:00 - until*	Sand Fly Surveillance Methods

Wednesday, April 15

AM	10:00 - 12:00	Final Examination
	12:00- until*	Group Photo/Service and Pack Equipment and Supplies/Correct Written Examination

Annex B

Training in Sand Fly Identification, Techniques on Surveillance and Investigation of Leishmaniasis and its Vectors

Student

No.	Name of Participant	Designation	Division
1.	Mr. Chiranjibi Shah	DPHO	MCD
2.	Mr. Shambhu Raj Shrestha	Entomologist	MCD
3.	Mrs. Nanda Palikhe	Parasitologist	MCD
4.	Mr. Shambhu Nath Jha	Parasitologist	MCD
5.	Mr. Murari Lal Das	Entomologist	MCD
6.	Mr. Ramananda Prasad Mandal	Entomologist	MCD
7.	Mr. Ganga Lal Shrestha	Asst. Entomologist	MCD
8.	Mr. Purushottam Gautam	Asst. Entomologist	EPD
9.	Mr. Shishir Kumar Pant	Asst. Entomologist	EPD
10.	Mr. Tribikram Shrestha	Asst. Entomologist	MCD
11.	Mr. Badri Bahadur Thapa	Malaria Inspector	MCD
12.	Mr. Bishnu Bahadur Shrestha	Malaria Inspector	MCD
13.	Mr. Shyam Kumar Lama	Malaria Inspector	MCD
14.	Mr. Bhanu Prasad Aryal	Malaria Inspector	MCD
15.	Mr. Kamal Khadka	Malaria Inspector	MCD
16.	Mr. Hem Prasad Poudel	Malaria Inspector	MCD
17.	Mr. Madhav Pyakurel	Malaria Inspector	MCD
18.	Mr. Keshav Prasad Upadhyaya	Malaria Inspector	MCD
19.	Mr. Narayan Prasad Shrestha	Malaria Inspector	MCD
20.	Mr. Pramod Kumar Shrestha	Malaria Inspector	MCD
21.	Mr. Harsha Nath Jha	Malaria Inspector	MCD
22.	Mr. Ram Sinhashan Shah	Malaria Inspector	MCD
23.	Mr. Ghanashyam Nepal	Malaria Inspector	MCD
24.	Mr. Ram Bahdur Thapa	Malaria Inspector	MCD
25.	Mr. Gautam Raj Shaema	Malaria Inspector	MCD

MCD = Malaria Control Division
EPD = Epidemiology Division

Mr. Riddi G. Vaidya, Senior Entomologist, MCD, and Mr. Shreedhar P. Pradhan, Medical Entomologist/Malaria Advisor, USAID/Nepal, were local coordinators for the training course.