VECTOR BIOLOGY & CONTROL

Vector Biology & Control Project
1611 North Kent Street, Suite 503
Arlington, Virginia 22209
(703) 527-6500

Telex: 248812 (MSCI UR)
Cable: MSCI Washington, D.C.

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ADULT ANOPHELINE DDT SUSCEPTIBILITY TRIALS AND ASSOCIATED ENTOMOLOGICAL EVALUATIONS AS RELATED TO MALARIA IN BOLIVIA

October 16 - November 30, 1986

by

Samuel G. Breeland, Ph.D.

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Author

Samuel G. Breeland, Ph.D., is a consulting entomologist based in Jacksonville, Florida.

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II. EXECUTIVE SUMMARY

This report summarizes the activities of Dr. Samuel G. Breeland and Government of Bolivia (GOB) counterparts under a consultant agreement for the purpose of evaluating the insecticide susceptibility of human disease vectors in Bolivia between October 16 and November 30, 1986.

After orientation meetings on October 17 with Dr. Robert Lennox, Dr. Andrew Arata and staff of the Vector Biology & Control Project (VBC), I traveled to La Paz on October 18 to begin work on October 20. The stated mission was to conduct anopheline susceptibility trials in the Departments of Beni, Pando, Santa Cruz, Tarija, Chuquisaca and Cochabamba in areas where, by GOB records, DDT utilization has been heaviest. Emphasis was to be placed on (a) zones of malaria hyperendemicity, (b) major vectors, Anopheles darlingi and An. pseudopunctipennis and (c) areas with known or suspected chloroquine-resistant strains of Plasmodium falciparum. Time permitting, studies were to be made on susceptibility of adult Aedes aegypti to malathion in the City of Santa Cruz. Along with susceptibility studies, ecological observations were to be made. Copies of pertinent data were to be left with USAID/La Paz and the Servicio Nacional de Control de la Malaria (SNEM).

The mission was accomplished to the extent allowed by logistics and the presence of anopheline mosquito populations. A preliminary draft summary report was left with both USAID and SNEM prior to my leaving Bolivia.

Meetings in La Paz on October 20 with appropriate officials of USAID, P.L. 480, Pan American Health Organization (PAHO), and the Ministry of Health, GOB, were followed by extensive planning and agreement sessions with Dr. Jorgé Mariscal, Director of Epidemiology and Chief, SNEM; and Dr. René Zumarán Varela, Chief, Communicable Diseases, P.L. 480. Areas selected for studies included Guayaramerín and Siglo XX in the Department of Beni; Las Araras and Ingavi in the Department of Pando; Mataral and General Roman in the Department of Cochabamba; Angostura, Surutu and the City of Santa Cruz in the Department of Santa Cruz; Estacion Caiza and Arrazayal in the Department of Tarija; and Situri and El Bañado in the Department of Chuquisaca. The plan was modified only in situations dictated by the lack of mosquitoes or problems of logistics.

Rarely were mosquitoes found in sufficient numbers for us to conduct susceptibility studies meeting WHO criteria. However, limited test results indicate probable susceptibility to DDT in the normal range for An. darlingi in Beni and Pando and for An. trinkae, the dominant anopheline in the Province of Chapare, Department of Cochabamba. Both species appear to be predominantly extradomiciliary and it is doubtful that there is a high degree of contact with residual deposits of DDT. Anopheles pseudopunctipennis shows signs of susceptibility to DDT ranging from poor to normal with varying degrees of tolerance by locality. The intradomiciliary habits of the species are also questionable in certain localities, making the general use of DDT wall spraying a question that must be evaluated locality by locality in relation to intradomiciliary habits. Aedes aegypti populations in Santa Cruz appear to be completely susceptible to both malathion and Sumithion. The GOB entomologists should make every effort to complete the planned baseline DDT susceptibility studies through the staff at the Cochabamba laboratory in cooperation with zone entomologists.

Vector biology and control investigations indicate the need for (1) a pilot conventional control program in Las Araras, Department of Pando, for the control of An. darlingi; (2) a study to determine the vector status and intradomiciliary habits of both An. trinkae and An. pseudopunctipennis in the Province of Chapare in the Department of Cochabamba; (3) a pilot vector control project in a community such as Pocitos in the Department of Tarija emphasizing larviciding of An. pseudopunctipennis habitats along with the intradomiciliary habits of adults; (4) the general development of methods of collecting both anophelines and Ae. aegypti as alternatives to human bait; (5) the stratification of malaria for the design of more effective vector control programs emanating from the local level; (6) the development of a contingency plan to meet potential vector control emergencies involving Ae. aegypti in Santa Cruz; and (7) the general improvement of entomological support for both malaria and Ae. aegypti control programs, perhaps through the establishment of a laboratory at Santa Cruz.

III. PRINCIPAL CONTACTS

USAID/La Paz	
Dr. Jerry Bowers	Health Development Officer
Mr. Timothy Bertotti	Executive Officer
Arq. Rafael Indaburu Q.	Project Manager
Lic. Elba Calero	Project Coordinator
P.L. 480	
Ing. Fedrique Muñoz Reyés	Executive Secretary
Dr. René Zumarán Varela	Chief, Communicable Diseases
Lic. José Sanjinez	Executive Under Secretary
PAHO	
Dr. Gustavo A. Mora	Bolivia Representative
Dr. Termistocles Sanchez	Malaria Advisor
Ministry of Health, GOB, La Paz	
Dr. Carlos Perez	Minister of Health
Dr. Angel Valencio	Director of Health
Dr. Jorgé Mariscal	Director of Epidemiology and Chief, SNEM
Sr. Nestor Prudencio C.	Chief, National Field Operations, SNEM
Sr. Federico Vargas Zabala	Chief, Central Office of Statistics, SNEM
Sr. Pepe Espinoza	Auxiliary Entomologist, Zone IV, SNEM
Sr. Roberto Rodriguez	Technical Entomologist, National Level, SNEM Cochabamba
Sr. Angel Rios Aroujo	Administrator, Communicable Diseases Project P.L. 480
Sr. René Mollinado Llave	General Administrator, Malaria, SNEM
SNEM, Zone I, Riberalta	
Dr. Jorgé Cuba	Chief Medical Officer
Sr. José Hashimoto	Administrator
Dra. Ruth Santivañez	Director, Sanitary Unit
Sr. Jaime Alvarez	Auxiliary Entomologist
Dr. Juan Carlos Avila M.	Chief Medical Officer (Guayaramerín)
Sr. Luis Crespo	Auxiliary Entomologist
SNEM, Zone II, Cochabamba	
Dr. Efraín Vallejo	Chief Medical Officer
Dr. Roberto Agudo	Malariologist
Sr. Demetrio Rivero	Auxiliary Entomologist
Sr. Orlando Murillo	Auxiliary Entomologist
Dr. Victor Lora Ponce	National Epidemiologist, Retired

SNEM, Zone VI, Santa Cruz

Dr. José Luis Cespedes	Chief Medical Officer, SNEM
Sr. Pablo Vaca	Auxiliary Entomologist, SNEM
Sr. Sergio Arteaga	Auxiliary Entomologist, SNEM
Dr. Wilfredo Vidal Boheme	Chief Medical Officer, Yellow Fever
Sr. Miguel Molina	Auxiliary Entomologist, Yellow Fever

SNEM, Zone VII, Tarija

Dr. Gustavo Quevedo	Chief Medical Officer
Sr. Ivar Colodro	Field Inspector

SNEM, Zone III, Chuquisaca

Dr. Freddy Lopez	Chief Medical Officer
Sr. Julio Lucuy	Auxiliary Entomologist
Sr. Julio Leon Laguna	Field Inspector
Sr. Mario Padilla	Administrator

IV. ACTIVITIES

Upon arrival in La Paz around 11:00 p.m. on Saturday, October 18, I was met by the USAID Executive Officer, Mr. Timothy Bertotti, who kindly helped me through customs and arranged my transport to the Hotel Plaza.

On Monday morning, October 20, I reported to Mr. Bertotti, who introduced me to my Control Officer, Mr. Rafael Indaburu. After a brief meeting with Mr. Indaburu and Dr. Jerry Bowers, USAID Health Development Officer, Mr. Indaburu and I met with P.L. 480 officials including Ing. Fadrique Muñoz Reyes, Executive Secretary; Lic. José Sanjinez, Executive Under Secretary; and Dr. René Zumarán, Chief of Communicable Diseases. This meeting involved a general discussion of my mission on behalf of VBC and its relationship to P.L. 480 representatives. This meeting was followed by very productive discussions with appropriate officials of the Ministry of Health including Dr. Angel Valencia, National Director of Health, and Dr. Jorgé Mariscal, Director of Epidemiology and Chief of SNEM. USAID participants included Mr. Indaburu and Lic. Elba Calero. Dr. Mariscal agreed to work with Dr. Zumarán and me in arranging a work plan, itinerary, and SNEM support for the VBC activities. In the afternoon, Mr. Indaburu and I met with Dr. Gustavo A. Mora, PAHO Country Representative, to discuss the project and its interrelationships with other groups. Dr. Zumarán and I spent the better part of the morning of October 21 in basic planning with Dr. Mariscal in the SNEM office. After completing agreements on the scope of work and establishing a plan, Dr. Mariscal assumed responsibility for a very detailed itinerary within the resources of the malaria program. By the end of a very busy day, he had made sufficient arrangements for the field work to commence immediately. Meanwhile, I apprised USAID officials of the plan and dispatched a telex to VBC.

The areas and localities selected for insecticide susceptibility trials are shown in Figure 1 and the planned itinerary is detailed in Table 1. Activities will be discussed chronologically, department by department.

Departments of Beni and Pando, October 23-29

On Wednesday, October 22, SNEM entomologists Roberto Rodriguez and Pepe Espinoza, and I traveled via air to Riberalta in the Beni/Pando area of northern Bolivia to commence field work. We were met by Jaime Alvarez, Auxiliary Entomologist, Zone I, Riberalta. Morning meetings were held with SNEM officials on

Thursday, October 23, to make specific arrangements for travel and work throughout the zone. In the afternoon we traveled to Guayaramerín with SNEM personnel including Dr. Jorgé Cuba, Chief Medical Officer, Zone I, Sr. Roberto Rodriguez, Sr. Pepe Espinoza and Sr. Jaime Alvarez. This group, along with others, participated in all activities in the area. Every effort was made to collect adult anophelines, principally Anopheles darlingi, in four localities (Guayaramerín, Siglo XX, Las Araras and Ingavi) representing the provinces of Vaca Diez (Guayaramerín and Siglo XX), Federico Roman (Las Araras) and Abuná (Ingavi) for use in the planned DDT susceptibility trials (Table 1). These localities had been selected on the basis of malaria hyperendemicity, the DDT-use pattern and the occurrence of chloroquine-resistant strains of Plasmodium falciparum. Further efforts were made to elucidate conditions affecting the transmission of malaria and implications of current and potential vector control approaches.

Guayaramerín. Exhaustive efforts in known malariagenic localities in and around Guayaramerín yielded a variety of mosquito species but few anophelines in seven nighttime captures over a two-day period (An. darlingi -- 5, An. albitarsis -- 4, An. trianulatus -- 4, An. peryasui -- 7) in spite of multiple efforts by a knowledgeable and well-trained team of specialists. Daytime activities included searches for larval mosquito habitats and associated sampling, the identification of natural or artificial resting shelters, placement of "red box" shelters, interviews with household occupants about mosquito activity, etc. Nighttime techniques included the use of "cebo humano" (human bait), animal bait, CDC light traps, "parked car" captures, etc., but all to little avail. Two full days and nights in the San Isidro locale of Guayaramerín yielded only the 20 specimens previously listed, only 5 of which were An. darlingi. Obviously, it was impossible to conduct insecticide susceptibility tests in the area. Results of all adult collections are given in Table 2.

Las Araras. On Saturday, October 25, a large contingent, including those previously listed, plus Dr. Juan Carlos Avila, Chief Medical Officer, Guayaramerín District, SNEM, traveled to Las Araras on the Bolivia side of Rio Madera on the border of Brazil. The group arrived in mid-afternoon in time to conduct daytime activities similar to those already described. Although this area has a history of malaria hyperendemicity and apparent active transmission, mosquito collecting results were extremely disappointing. In spite of diligent efforts and the general presence of mosquitoes, only 26 anophelines were collected (Table 2). One limited test with 4.0% DDT exposure was conducted on October 26 with a mixed group of specimens.

Although collections for susceptibility studies were nil, Las Araras provided a wealth of insight into the malaria control problem in Bolivia in general and into the compact village in particular. The malaria history and prevalence staggers the imagination. For example, the major (El Alcalde) of Las Araras has had malaria 40 times; a two-year old girl -- 8 times; an engineer -- 20 times in seven years; a frequent visitor -- 8 times in two years, etc. Every resident questioned reported having had frequent malaria attacks. As I questioned the mayor, his young daughter lay at my feet with peak symptoms of malaria. In the community of Las Araras, the chance of contacting malaria appears to be 100 percent. It seems that it is not a question of if, but when -- although SNEM records show only a 20% slide positivity rate. According to Zone 1 and Guayaramerín District Malaria Directors, both of whom were on the trip, there seem to be multiple strains of Plasmodium throughout the area. In spite of two SNEM workers being permanently based at Las Araras and every reasonable effort seemingly being made, the scourge goes on. However, I feel that Las Araras offers an ideal testing ground for real malaria control to accomplish what, perhaps, actions taken to support the eradication concept could not possibly achieve. Favorable results could influence modifications in other such communities.

Briefly, my reasons for believing that Las Araras offers much promise for yielding to conventional, longitudinal malaria control are the following: (1) the community is compact and isolated (ca. 200 houses, 500-600 residents); (2) the possible, even probable, anopheline breeding habitats are well defined and accessible; (3) housing would lend itself to exclusion mechanisms; (4) the people are knowledgeable about malaria and receptive to education, prevention and treatment activities. The creation of a sound, longitudinal vector control program would seem to be achievable under existing conditions. The situation at Las Araras leads one to surmise that An. darlingi might not be the exclusive vector, that outdoor transmission probably occurs at a high rate, and that if An. darlingi is, indeed, the major vector it is sparsely distributed, mostly exophilic and exophagic, wary of DDT-sprayed surfaces and extremely efficient as a vector of malaria in this area. Supportive entomological studies are strongly indicated.

The Las Araras group broke camp on Monday, October 27 to return to Guayaramerín to work again in that area and to make preparations for the trip to Siglo XX.

Siglo XX. After the work in Las Araras, the plan called for a return to base at Guayaramerín with two days at Siglo XX in the Province of Vaca Diez. However, previous experience in the area indicated that the absence of larval breeding or adult harborages such as cattle corrals near breeding sites would signal additional lost time. After a four-hour daytime search with negative results, it was decided to continue on to Riberalta for a planned flight to the isolated locality of Ingavi in the Province of Abuná. Extensive preparations had been made for a two-day stay in this area accessible only by air, but one with much interest to malaria officials in Bolivia. The flight was made with my Bolivian counterparts in a private "avioneta" flown by Bolivian Air Force pilots. Unfortunately, the proposed runway was too short and in no condition for a landing. As pre-planned, the group returned to Riberalta to resume work in that area.

The evening of October 29 yielded a few mosquitoes and some Phlebotomus, but only a single specimen of An. darlingi. The work in Zone 1 was reviewed with Dr. Cuba on October 30 and loose ends tied, marking an end to our activities in this area. The group departed for Cochabamba at 1:30 p.m., and arrived at about 5:00 p.m. We were met by Dr. Efrain Vallejo, Chief Medical Officer, Zone 11, SNEM.

Department of Cochabamba, October 30-November 8

The first day at Cochabamba, October 31, was spent at the SNEM laboratory/insectary in the identification of mosquitoes collected in Zone 1 and in observing colonies and learning something of the functions of the laboratory. The laboratory is impressive. There are five colonies of An. pseudopunctipennis (from La Paz, Santa Cruz, Sucre, and Cochabamba, 2) maintained by force-mating the parents of each generation. In addition, there is a single self-perpetuating colony of Aedes aegypti originating from Santa Cruz. An. darlingi has defied laboratory colonization. Emphasis seems to be on taxonomy and basic laboratory biology. The laboratory is capable of mass rearing specimens, at least, for bioassay work in the field. Fruitful discussions concerning the malaria and yellow fever problems in Bolivia were held with long-time specialist, Dr. Victor Lora Ponce, who shared with me a wealth of knowledge and insight.

On November 1, one susceptibility test was conducted with An. pseudopunctipennis, but the test must be considered informal in that specimens were available for only one replicate with DDT. At least the test served to supplement training provided earlier in the year by Glen Fleming of PAHO. A second test using Ae. aegypti from Santa Cruz was conducted using exposure times of

fifteen minutes, thirty minutes, one hour and two hours, plus control with malathion at 5.0% and two cylinders of Ae. aegypti from the same source using Sumithion at 1.0%. Again, the tests are informal in that only a few specimens were available.

A group of six persons traveled to Chapare in the Department of Cochabamba on November 3 for mosquito collections and insecticide susceptibility trials involving anophelines in several localities. Emphasis was placed on General Roman and Villa Tunavi. This is a particularly interesting area because of the occurrence of malaria without the noticeable presence of either An. darlingi or An. pseudopunctipennis, but with a dominance of An. trinkae (taxonomically close to An. rangeli), whose vector status is unknown. The group arrived in time to check for anopheline breeding places and potential foci of adults. The first sizable collection of any anopheline species in this study turned out to be An. trinkae from the locality of San Mateo, Province of Chapare, on the night of November 3. Females were biting cattle and resting in large numbers among foliage, trunks and limbs of citrus trees near the tethered cattle. A sufficient number was collected to permit a three-replicate test with the full range of DDT concentrations. Test results are probably questionable in that it was necessary to transport the tubes to Cochabamba after having set the holding chambers. One hundred percent mortality, however, was obtained at the 4.0% level. The area of General Roman was visited on November 4 for field investigations of malaria activity and searches for larval mosquitoes for a more positive identification of An. trinkae. Results of the tests with An. trinkae were read at the Cochabamba laboratory on November 5 after which some of the group departed for Mataral in hopes of finding a field source of An. pseudopunctipennis before leaving the Department of Cochabamba. The SNEM group was successful in collecting a sufficient number of the species for a two-replicate test from Mataral in the Province of Campero on the nights of November 6 and 7.

After a review of progress to date, I was sufficiently concerned about the probable failure of the basic mission, viz., baseline DDT tests, to telephone Mr. Indaburu of USAID to discuss alternatives. After explaining our progress in other areas of the work, it was decided to continue without change in plans or emphasis.

The group returning from Mataral reported that the best results in collecting An. pseudopunctipennis had come from specimens entering the tent and that the species had been active all night. This is not unlike the behavior of An.

pseudopunctipennis in Central America, and suggests the possibility of using artificial shelters, animal baits and other means of collecting all night. Much of my own time was spent preparing material for an impending review with Dr. Zumarán in Santa Cruz, our next destination.

Department of Santa Cruz, November 8-13

SNEM entomologists, Roberto Rodriguez and Pepe Espinoza, and I traveled to Santa Cruz on Saturday, November 8. Upon arrival we were met by Dr. René Zumarán, Chief, Communicable Diseases, P.L. 480. The four of us had a brief meeting at the SNEM office along with Dr. Wilfredo Vidal, Chief of Zone V, SNEM. The group met again on Sunday for a complete review of the work to this point. I expressed my concern to Dr. Zumarán over the scarcity of anopheline specimens for susceptibility tests, reviewed the work in detail and outlined projected plans.

As a result of the review, Dr. Zumarán strongly urged continuation of the work without modification. On Monday, November 10, orientation meetings were held with the staffs of both SNEM and Yellow Fever Control. Plans were solidified for the work from the Santa Cruz base. The group was split into two teams, one each for anopheline and Aedes aegypti activities. Collections were begun in the afternoon by both groups. As a member of the Ae. aegypti group, my time was spent in adult mosquito collections in the mornings and evenings and in searches for larval habitats and foci of activity during the day. These searches continued throughout the week. On Wednesday, November 12, there was a lengthy session at the SNEM/Yellow Fever headquarters with Dr. Jorgé Mariscal, Chief Epidemiologist of both groups, and Dr. Termistocles Sanchez, PAHO Malaria Advisor. Principal staff members from Santa Cruz also participated. A complete review of all activities was well received by the group in spite of the paucity of data on DDT susceptibility. In addition to the review of the work, many aspects of the programs were discussed, some in detail, with emphasis on the findings and implications to the Bolivia vector control programs.

Efforts to collect sizable numbers of Ae. aegypti in the Santa Cruz area were thwarted by heavy rains on the first day of work, November 10, followed by high winds and rather cold weather. The cold and wind continued through November 13, especially in the mornings and evenings when Ae. aegypti is normally active. Adding to the problem was the complete cleaning of cemeteries, normally good collecting sites, for All Soul's Day, celebrated on November 2. However, these factors were probably minimal in comparison to the general absence of breeding

foci in the city. Every tire yard and cemetery was extensively searched, as were other foci known to the Santa Cruz staff. The rain on November 10 was reported to have been the first in more than a month. In spite of the difficulties, a total of 65 females was painfully collected from several visits to one particular tire yard (Recauchutadora Potasi). All other probable sites identified by the staff of the Yellow Fever Program were negative for both larvae and adults. Sixty specimens were used for a limited test using 5.0% malathion papers at exposure times of thirty minutes (20 specimens) and one hour (20 specimens). Complete mortality was obtained at each concentration (Table 10).

Efforts to collect An. pseudopunctipennis in the Department of Santa Cruz were even more futile. A team composed of the SNEM director, his staff, and Roberto Rodriguez of our group searched diligently for specimens in malarious areas well known to the team. Searches in the localities of Villa Barrientos (500 m. alt.) and El Remanzo (220 m.), Province of Andresibanas, yielded 12 and 15 anopheline larvae, respectively. No adults were collected in biting collections at either site. After two days and nights, the team moved to the locality of San José (240 m.) in the Province of Ichilo, where only five females were collected. The general absence of An. pseudopunctipennis can only be explained by a prolonged drought in the area. Because mid-November is the beginning of the rainy season in the area, it stands to reason that major dry season habitats had already peaked, leaving only a minimal number of available sites with greatly reduced surface water.

Department of Tarija, November 14-20

The group -- Breeland, Rodriguez and Espinoza -- traveled to Tarija on Friday, November 14. Upon arrival it was found that we were totally unexpected and that SNEM was unprepared for our visit. After considerable discussion of options, it was decided that field work could not possibly commence before Monday. The weekend was spent working up data and drafting reports. On Monday our group was joined by Ivar Colodro of the Tarija SNEM staff for extensive field work in the Department. On the first day, we traveled by Jeep as far as Cerapari en route to Pocitos and Arrazayal. This was an 11-hour trip. After camping overnight, the group continued on to Pocitos on the Bolivia side of the Argentina border. At Pocitos we sampled likely breeding places and searched for anopheline foci during the day and made nighttime collections of An. pseudopunctipennis from two locations near Pocitos. On Wednesday the group traveled to Arrazayal, sampled breeding foci, set up camp for nighttime collections and susceptibility tests with specimens collected to

this point. All-night thunderstorms on Wednesday night precluded any further collecting or testing in the area of Arrazayal. The group returned to Tarija.

After having located rather prolific breeding sites of An. pseudopunctipennis on the outskirts of Pocitos, attempts to collect adults by human bait during the night yielded only 65 females. The specimens were saved for later testing to avoid losing even more time in the area. Subsequent collections all night at another breeding site in Arrayazal yielded only 20 specimens. Nevertheless, DDT susceptibility tests were conducted using specimens from Pocitos at 2.0% and 4.0% and with the specimens from Arrayazal at 4.0%, all with a common control group from Pocitos. Complete mortality was obtained at the 4.0% level among specimens from each locality and 85% at the 2.0% level for those from Arrayazal. Results are given in Tables 6 and 7.

Department of Chuquisaca, November 21-25

After finalizing and reviewing the work in Tarija with SNEM, the group traveled to Sucre to begin work in Zone III, Department of Chuquisaca. On the first day, Friday, November 21, orientation meetings were held with Dr. Freddy Lopez, Chief Medical Officer and former head of the national malaria program. On Saturday the group left for Suturi and El Bañado for one final attempt to collect An. pseudopunctipennis. Much of my time in Sucre was spent in consultation with SNEM officials and in preparing interim reports for GOB and USAID. SNEM, again, was on strike, leaving no secretarial support. Although the SNEM at Sucre was experiencing a strike during our visit, cooperation with our efforts was excellent. A team consisting of our group and Julio Lucuy and Julio Laguna of SNEM traveled to the area of Monteagudo for work in the localities of Candua, El Bañado and San Miguel, all in the Province of Hernando Siles, a trip of some eleven hours, each way.

Searches for An. pseudopunctipennis at Candua on November 23 (alt. 1,080 m.; temp. 16°C) yielded only 13 anophelines, 11 of which were An. pseudopunctipennis. Collections at two localities on November 24 yielded 17 An. pseudopunctipennis at El Bañado (alt. 1,120 m., temp. 22°C). Specimens were collected by human bait at Candua and El Bañado, but were not in sufficient numbers to conduct a susceptibility test. The females collected at San Miguel were in natural foliage, but refused to accept blood meals when offered. Thus, no tests were conducted in the Department of Chuquisaca. However, the group was to hand carry gravid females to Cochabamba for possible tests with progeny. From the entomological standpoint, studies recommended in section VI in

the Department of Tarija and Cochabamba should be applicable to this Department as well.

La Paz, November 27-30

Thursday, November 27, was spent at SNEM headquarters in La Paz in preparing an interim report to satisfy the needs of GOB, USAID, and P.L. 480 officials prior to my leaving Bolivia. The following day was spent in meetings with representatives of these groups including an audience with Dr. Carlos Perez, Minister of HEalth, and in appropriate protocol visits marking an end to the work in Bolivia. The return trip to Jacksonville was made as scheduled on Sunday, November 30. A telephone report was made to Dr. Andrew Arata, VBC, Arlington, VA, on December 1.

V. FINDINGS AND CONCLUSIONS

Insecticide Susceptibility Tests

As the work progressed, it was obvious that sufficient mosquito specimens would not be available for tests following the World Health Organization (WHO) criteria for establishing baseline levels of insecticide susceptibility for each department. Nevertheless, tests were conducted to the maximum extent possible, even when only a few specimens were available. Because specimens were rare, a major emphasis was placed on elucidating every aspect of vector biology and control relating to the control of malaria and to the role of SNEM in each locality, province and department visited. Throughout these studies, "mosquitoes" were plentiful as evidenced by the data in Table 2 showing results of collections in the Departments of Beni and Pando. However, the principal species being sought in these areas, An. darlingi, was rare. This situation also prevailed in the pursuit of An. pseudopunctipennis throughout its range even though collections were concentrated near breeding foci and in malariagenic communities.

An. darlingi. In DDT tests, the only trial involving An. darlingi was with only 12 specimens among a mixed group of anophelines collected from the community of Las Araras, Province of Abuna, Department of Pando, on October 26. A one-hour exposure to 4.0% DDT paper in the standard WHO exposure tube resulted in 100% mortality after 24 hours. One of six specimens in the control group died, giving an uncorrected control mortality of 17%. Results are shown in Table 3.

An. trinkae. This is a species about which little is known in Bolivia, except that it is quite common in the Province of Chapare in the Department of Cochabamba. The species was described in 1979 (Faran 1979) and the systematics is complex, making it difficult to separate from its near relatives, An. rangeli and An. nuneztovari, which according to Faran (1980) make up one phyletic line of the Oswaldoi complex in the Albimanus section of the subgenus Nyssorhynchus of Anopheles. Faran has described An. trinkae as a distinct species, but states that nothing is known of its vector status. Although it is possible that the entity known as An. trinkae might not be genetically isolated from its close relatives, because of apparent geographic isolation, this might be a moot point.

A large number of specimens, believed to be An. trinkae, was collected on the night of November 4 from the locality of San

Mateo Bajo, Province of Chapare, Department of Cochabamba, Bolivia. Females from the collection were used to conduct DDT susceptibility tests using the full range of concentrations (0.25, .5, 1.0, 2.0 and 4.0%) with three replicates. The results showed mortalities of 35%, 53%, 37%, 78%, and 100%, respectively. Although somewhat erratic, the population is probably within the normal range of susceptibility as indicated by the 100% kill at the 4.0% level. However, the species appears to be both exophilic and zoophilic, making its susceptibility to residual deposits of DDT relatively unimportant with regard to control. Results of the DDT trials are given in Table 4. The implications of the apparent dominance of this species in certain areas of the Department of Cochabamba are fully discussed later in this section.

An. pseudopunctipennis. Results of DDT tests involving An. pseudopunctipennis are given in Tables 5 through 7. Tests using the full range of DDT concentrations with one-hour exposure, two replicates, with specimens collected November 6-7 from Mataral, Province of Campero, Department of Cochabamba gave an overall mortality of 92.5% at the 4.0% level (Table 5). Results from exposure to DDT of An. pseudopunctipennis from Pocitos, Province of Gran Chaco, Department of Tarija on November 19 showed 85% mortality among 20 specimens at the 2.0% concentration and 100% at the 4.0% level (Table 6). Equal results were obtained, i.e., 100% mortality at the 4.0% level, among specimens collected from Arrazayal, Province of Arce, Department of Tarija (Table 7). There was no mortality among the common control group for either locality.

In addition to tests using field-collected An. pseudopunctipennis, one test was conducted with a group of specimens from the SNEM colony at Cochabamba. These specimens were in the 54th generation and represented the Río Pojo Colony, Department of Cochabamba. The results (Table 8) were extremely erratic, giving 60% mortality at the 2.0% level and only 35% at the 4.0% level. Such colony specimens are probably useless in DDT-susceptibility trials, or even for field bioassay studies. A review of the data of Table 9 showing results of DDT susceptibility tests using 4.0% exposures between 1982 and 1985 by the staff of the Cochabamba laboratory shows a similar inconsistency in results among specimens from the Río Pojo area.

Overall results from DDT susceptibility tests involving An. pseudopunctipennis, though quite limited, do indicate that the species probably has many different levels of susceptibility--depending upon locality -- making it impossible to judge the general usefulness of DDT. This observation points out the need

for stratification and vector control evaluations at the locality level.

Ae. aegypti. Results of the limited susceptibility tests involving Ae. aegypti in the Santa Cruz area reveal no existing or potential problem of resistance to malathion. Both 30-minute and one-hour exposures to 5.0% malathion papers gave 100% mortalities among specimens collected in the city (Table 10) and 85% and 100% mortalities among specimens from the Santa Cruz colony at the SNEM laboratory in Cochabamba (Table 11). Equal results were obtained with Sumithion at 1.0% among the colony mosquitoes (Table 12).

The failure to establish baseline levels of insecticide susceptibility during this period should not deter future efforts of GOB entomologists to accomplish this important goal on an opportunistic basis as mosquitoes become available in specific localities selected for this study (Fig. 1). The expertise and means to complete the studies are certainly available as a result of this effort and the earlier training provided by PAHO through Glen Fleming.

In summary, the limited insecticide trials do show probable susceptibility to DDT in the normal range for An. darlingi and An. trinkae, though habits of both species are probably such that little contact is made with interior deposits of the insecticide. An. pseudopunctipennis shows signs of susceptibility levels ranging from poor to normal with varying degrees of tolerance by locality. This species also shows a confusing pattern of behavior concerning its intradomiciliary nature, a factor that must be determined for each discrete population to give any meaning at all to the question of DDT susceptibility. There would seem to be no question of susceptibility of Ae. aegypti to malathion on an operational basis in Santa Cruz. This belief is based not only on Santa Cruz observations, but on my previous experiences in other areas of Latin America as well.

Vector Biology and Control

In a country such as Bolivia, except for large-scale national efforts with full outside support such as those previously offered by the Malaria Eradication Program and the Aedes aegypti Eradication Program, I do not foresee a workable national malaria control program emanating from the Capital. Given the general problem of logistics; a low density human population; locality variables -- topography, climate, rainfall, mosquito fauna, mosquito density, accessibility; Plasmodium variables; presence of trained personnel; and the existence of

other, sometimes more pressing, public health problems, mosquito control aspects of malaria control should be tailored to each locality by zone personnel.

Basically, the SNEM malaria team in each department should identify problem localities, design programs for each, and submit their plans from the locality -- district, zone levels -- to the central SNEM directorate for consideration, direction and support. Programs should be developed from the bottom of the pyramid rather than the apex. Given the current situation, I see no other effective way to control malaria in Bolivia. Except for the eradication concept, no longer workable for many well-known reasons, mosquito control has always been a local matter, albeit with hierarchal guidance and support. Consequently, I have chosen to style this report department by department, rather than from a national perspective.

Departments of Beni and Pando, Zones I and V. The reputed anopheline vector of malaria in this area of Bolivia is *Anopheles darlingi*. Observations indicate that, in Bolivia, *An. darlingi* is not particularly endophilic or endophagic. Both of these factors would impede success through residual wall spraying with DDT or any other residual compound. Likewise, entomologists in Zone I, as well as those at the national level, indicate that the species is seldom active after 8:30 p.m., and that it is mostly active at twilight, a time when very few people are indoors. In fact, all efforts to collect this species with "cebo humano" are outdoors, rather than indoors, and early in the evening. The only other practicable method of collecting biting or resting specimens is to use animal bait during early evening, again outdoors.

In spite of all the evidence pointing to outdoor biting activity, the basic attack has been, and is, wall spraying-- why? Further, *An. darlingi* has the reputation, even when indoors, of being too flighty and restless to spend much time resting on interior surfaces, even after feeding. Also, the species does not seem to concentrate in favored niches -- a requisite habitat for vulnerability to residual wall spraying-- as do many shelter-oriented anophelines. When confronted with these factors, statistically oriented specialists respond that malaria goes down with spraying and up in the absence of spraying. This seems to justify total reliance on the DDT wall spraying approach plus, of course, drug prophylaxis and treatment. Perhaps the answer is that concerted spray programs bring increased overall malaria control activity. To those who believe that wall spraying is effective, I would respond that in Guayaramerin there is a 60% slide positivity rate, yet there is

an unbroken record of wall spraying. The same is true, although at a lower recorded rate, for Las Araras, where everyone seems to have repeated malaria attacks.

The reasons usually given for failure of wall spraying programs to reduce malaria transmission are (1) resistance to the insecticide employed, (2) discontinuous walls, and (3) extradomiciliary transmission. Although we lack reliable data on susceptibility of An. darlingi to DDT in Zone I, the other two factors (discontinuous walls and extradomiciliary transmission) are more than likely. Thus, even if the vector is normally susceptible to DDT, I doubt its usefulness against An. darlingi in this area of Bolivia.

I offer Las Araras as an example of alternatives that might be considered. This locality would seem to be a closed transmission system, i.e., it probably engenders and perpetuates its own malaria, although there is a considerable amount of traffic in and out of the community. Because of its relative isolation, manageable size, and compactness, I would suggest the possibility of a model vector control program to include source reduction, larviciding, and crepuscular space spraying of such chemicals as malathion or nailed with portable hand equipment, especially during periods of high malaria transmission. Space spraying (fog or ULV) would circumvent (1) any resistance to DDT, (2) domiciles with discontinuous walls, and (3) outdoor biting at the critical time. Such a program could be easily mounted in that two SNEM workers reside in Las Araras. This activity likely would be both cost effective and efficacious for anopheline control. Bonus benefits would be derived through some control of a very serious black fly and noxious mosquito problem (Table 1). The medical aspects of the current activities would continue as at present and, if successful, a control program could gradually reduce or eliminate the need for DDT wall spraying.

This proposal is offered only as an example. Similar programs could, and should, be tailored to meet the varying conditions of other communities, and would seem to offer a viable alternative, or augmentation, to the sole use of DDT wall spraying for the vector control aspect of malaria control.

Department of Cochabamba, Zone II. Although An. pseudopunctipennis is the reputed vector of malaria throughout the Department of Cochabamba, the species is essentially absent in the Province of Chapare. Yet, there is malaria and a DDT spray operation. The SNEM staff at Cochabamba and workers in the field indicate that the most probable vector is An. trinkae, closely related to and difficult to distinguish in the adult

stage from An. rangeli.

The presence, distribution and density of An. trinkae in the Chapare area and its abundance during malaria activity, along with the virtual exclusion of known vectors, indicate an urgent need to clarify its status as a vector. There is sufficient interest and expertise among the group at the Cochabamba SNEM laboratory to lead me to expect that some type of study will be undertaken. At the minimum, I would suggest a study designed to determine the ability of An. trinkae to accept and develop the parasite to the infective stage (medical aspects under the guidance of Dr. Vallejo) with corollary collections and dissections of guts and glands by the entomological staff (under the guidance of Sr. Rodriguez). I also feel that An. pseudopunctipennis might be somewhat of an unknown quantity in the Chapare area of the Department. Indeed, it must be present. Perhaps it is either highly focalized or very evenly and sparingly distributed, but sufficiently so to transmit malaria, especially during periods of maximum inundation, reportedly in June or July.

There seems to be an inordinate amount of movement of people in the Chapare area -- hotels, restaurants and buses abound. It is indeed possible that people movement, rather than mosquito distribution and density, might explain malaria in the area. At any rate, the elucidation of the vector status of An. trinkae and the true role of An. pseudopunctipennis, including domiciliary habits, would seem to be important objectives in the Department of Cochabamba. The Department has the advantage of the SNEM laboratory, a viable asset, for finding solutions to entomological and epidemiological problems in much of the valley and tropical areas of Bolivia.

Department of Santa Cruz, Zone VI. There are two distinct vector control problems and two distinct programs in the Department of Santa Cruz: the yellow fever problem and program concentrated in the city, and the department-wide malaria problem and program. Although in the same quarters and under a common administration, these programs seem to have little else in common at the operational level. This is an observation and not a criticism or judgment. However, it was quite noticeable that both programs are strongly control-oriented with very little supportive entomology, which is badly needed for surveillance and evaluation. The organizational structure, staffing, facilities and current operations indicate both the need and resources for developing a more supportive entomological structure. During our visit, it was apparent that even the most basic entomological surveillance was virtually absent as indicated by the lack of

equipment or expertise in this area.

During a joint meeting with the malaria and yellow fever staffs, Dr. Mariscal suggested the possibility of establishing a laboratory/insectary at Santa Cruz along the lines of the one at Cochabamba. I think of no other single action that can contribute more to both programs. Such a laboratory, properly staffed, would greatly improve the entomological support so badly needed for the important Ae. aegypti control operations in Santa Cruz and for malaria control in those departments to the south which have An. pseudopunctipennis as a common vector. Further, a laboratory at Santa Cruz would allow the Cochabamba facility more concentration of supportive work and coordination in the northern areas which include the problems in Beni and Pando with An. darlingi and in Chapare (Cochabamba) with An. trinkae. Furthermore, 30 percent of the malaria in Bolivia comes from the area of Guayaramerín, which deserves special attention from the Cochabamba staff.

From the standpoint of malaria control, other than a need to define the problem areas spatiotemporally as a basis for designing effective localized control programs, I see no special problems unique to the Department of Santa Cruz.

With respect to the Yellow Fever Control Program, it appears that all that can be done routinely to control the vector is being done. The major weakness is in the area of surveillance and evaluation of control efforts, i.e., the need for more and better entomology. A glaring need, expressed by Dr. Mariscal, is in the development of a plan of action, including on-hand resources for dealing with any emergency situation in Santa Cruz. Perhaps this is an area in which the VBC Project could be of considerable assistance.

Department of Tarija, Zone VII. The anti-malaria activities in the Department of Tarija seem well organized and in the hands of an efficient staff in spite of mind-boggling logistics problems. When we -- Breeland, Rodriguez and Espinoza -- arrived in Tarija on Friday afternoon, November 14, we were provided transport and the expertise of Ivar Colodro for a rigorous period of field activities, which included the localities of Pocitos and Arrazayal. This trip, perhaps more than others, exemplified the extreme problem of logistics in malaria control in Bolivia. The trip required 11 hours of driving -- five hours after dark -- on treacherous mountain roads of dirt, rocks, mud and dust just to reach a point to pass the night in tents. At about mid-point, our Jeep was pinned to the side of a mountain cut-out by a loaded gasoline tank truck going in the opposite direction with a 1000-

foot drop to the outside. Further along, it was possible to pass a construction area only between the hours of 6:00 p.m. and 6:00 a.m. The second leg of the trip required four hours the following day on the same type of roads. After collections at Pocitos on the Argentina border, it was necessary to clear customs and make special arrangements to travel through Argentina for the six-hour trip to Arrazayal in Bolivia. All of this for 65 specimens in Pocitos and 20 additional specimens in Arrazayal. Nevertheless, some DDT susceptibility tests were completed and much was learned of the character of anophelism and malaria as well as problems of control in the Department.

Notwithstanding logistics, there are some interesting and important observations on vector biology and control in the Department of Tarija. Reportedly, there are a number of places with malaria cases but little evidence of anophelism, and others with anophelism but no malaria. Such situations cannot be, and their true status certainly should be clarified. Other observations are that (1) spraying is being carried out even in areas where there is no malaria, (2) in some localities, e.g., Tarairi, both DDT spraying and malaria treatments are accomplished, but malaria goes on, and (3) there is a policy of "emergency spraying" of walls with DDT upon request or indication of malaria. These factors combine to reflect a committed effort on the part of SNEM to contain, or even prevent malaria, but under a set of parameters not well defined. Successful stratification is unlikely in much of the Department in that outbreaks occur seemingly at random and in unpredictable fashion, much like brush fires; hence, the "emergency treatment" program.

Although not discussed in other departments, the feasibility of depending on other sources, e.g., military and community volunteers, for malaria control work was discussed to some extent by the SNEM staff in Tarija. A major problem in vector control is that military personnel, trained to operate "by the numbers," are technically good but are subject to being pulled from the job without notice. Likewise, community workers are not too dependable except for those in the voluntary collaborator system, which seems to work quite well.

The major problem in the Department of Tarija, I believe, is logistics. It is simply impossible to cover such an area in the absence of definite and accessible foci. However, there are areas that would lend themselves to local control. In fact, the town of Pocitos would seem to be ideal for a pilot study on conventional control, and such a study might well answer some of the above-posed questions regarding anophelism and malaria. Factors indicating a good study site include those of a

relatively compact community in proximity to well-defined breeding areas. Thus, breeding, dispersal, and domiciliary habits could be evaluated in one relatively small area and evaluated against conventional control methodology, emphasizing serial larviciding, i.e., every eight to ten days. Though difficult, stratification would seem to be the only logical approach to a more defined program in the Department.

Department of Chuquisaca (Sucre), Zone III. Very little was learned in this Department. It appears that *Anopheles pseudopunctipennis* is the only vector and that there is nothing unique to the Department. It is believed that the studies recommended to clarify the intradomiciliary habits and the conduct of some pilot conventional control projects in the Departments of Tarija and Cochabamba would be applicable in localities of Chuquisaca.

VI. ANNOTATED RECOMMENDATIONS

It is recommended that the intended baseline DDT susceptibility trials be completed on an opportunistic basis by GOB entomologists. Anopheline populations in tropical areas tend to be very evenly distributed during an annual cycle, with perhaps one or two peaks in a 52-week period. In Bolivia, with several regions of distinct climates and ecologies, as well as several vector species, it is unrealistic to expect that a specific anopheline species could be collected in large numbers at a specific time in a specific locality. Collections for four replicates of a DDT trial to establish a baseline meeting WHO criteria would require a minimum of 500 specimens from essentially the same population and of a similar physiological state. Given enough time and a laboratory/insectary this could be done. If not collectable at a given time, specimens could be reared from larvae or from eggs obtained from gravid females. Therefore, it is suggested that entomologists in each of the departments determine the time, place and manner of anopheline collections to permit a full series of DDT trials when specimens become available. The laboratory at Cochabamba should be contacted by the locality auxiliary entomologist at the right time. In turn, the Cochabamba entomologist should conduct the appropriate tests and distribute results as prescribed by WHO. VBC, perhaps through USAID and P.L. 480, should support the trials and coordinate the study.

It is recommended that a pilot program of conventional mosquito control aimed at Anopheles darlingi be conducted in Las Araras. The malaria situation and outlook for control in Las Araras are such that a pilot program of conventional control should be established for at least one year. Two years would be preferable.

It is recommended that the vector status of both An. trinkae and An. pseudopunctipennis be elucidated in the Province of Chapare, Department of Cochabamba. A unique problem exists in Chapare in that malaria is persistently present but the vector is either unknown or of doubtful status. Such a study has been fully discussed with the SNEM staff at Cochabamba.

It is recommended that a conventional anopheline control program, on a pilot basis, with associated studies on the domiciliary habits of An. pseudopunctipennis be conducted in the Department of Tarija. Elucidation of the true intradomiciliary habits of this species is basic to the use of DDT or other residual wall sprays as the principal vector control method of

interrupting malaria transmission in Bolivia. To understand this aspect of vector biology is perhaps more important than the determination of susceptibility to DDT -- a moot question if the species is not strongly inclined to transmit malaria indoors. The domiciliary habits should be determined in a locality where the vector status of the species is not questioned, e.g., in Pocitos in the area of Zone VII near the Argentina border. In addition, the same locality could be the site of a conventional malaria control and evaluation project. Results of such a study would contribute to the knowledge needed for dealing with malaria control problems associated with An. pseudopunctipennis throughout its range in Bolivia.

It is recommended that alternative methods of collecting female anophelines be developed. In that all collections for susceptibility studies are characteristically made by the "cebo humano" (human bait) method and early in the evening, it is suggested that SNEM conduct studies on the efficacy of animal baited traps, artificial resting stations and various types of light traps. Baited traps and light traps would work all night and artificial shelters during the day, giving 24-hour coverage while reducing the need for the dangerous and often ineffective practice of using human bait to capture anophelines.

It is recommended that alternatives to human bait captures be developed for Aedes aegypti. Because collection of Ae. aegypti in Santa Cruz requires a considerable number of man-hours at specific times, morning and evening, consideration should be given to acquiring and modifying a New Jersey-type light trap for the domicile collecting of this species. The details have been discussed with appropriate personnel of the Yellow Fever Project and the SNEM laboratory staff at Cochabamba.

It is recommended that a contingency plan for dealing with vector control emergencies be developed by the yellow fever group. The question posed by Dr. Mariscal as to what would or could be done to cope with a major outbreak of yellow fever or dengue in Santa Cruz deserves immediate attention. The PAHO has a manual on this subject which should serve as the basis for addressing this problem. The plan should provide for the immediate acquisition of insecticides and delivery equipment.

It is recommended that a laboratory/insectary be established at Santa Cruz. The establishment of an entomological support facility at Santa Cruz, patterned after the one at Cochabamba, as suggested by Dr. Mariscal, Chief Epidemiologist, GOB, is strongly endorsed.

It is recommended that vector control decisions, insofar as possible, emanate from the local level. Vector biology and control problems and recommended solutions should be identified at the locality level, and passed on to the district and zone levels for consideration by the national level of SNEM. The emphasis should be from the bottom of the pyramid to the apex with a stronger horizontal function. This suggestion is discussed more fully in the main body of this report.

It is recommended that SNEM, with cooperation from each zone, develop a workable malaria stratification program. Malaria control in Bolivia in the future will have to be locality-based. A common national program will not serve the needs of the people. This question is addressed fully in this report and essentially all of the observations herein point to the need for stratification and associated work plans.

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Figure 1

AREAS SELECCIONADAS PARA INVESTIGACION DE SUSCEPTIBILIDAD DE ANOPHELINOS AL D.D.T. BOLIVIA OCTUBRE-NOV. 1986

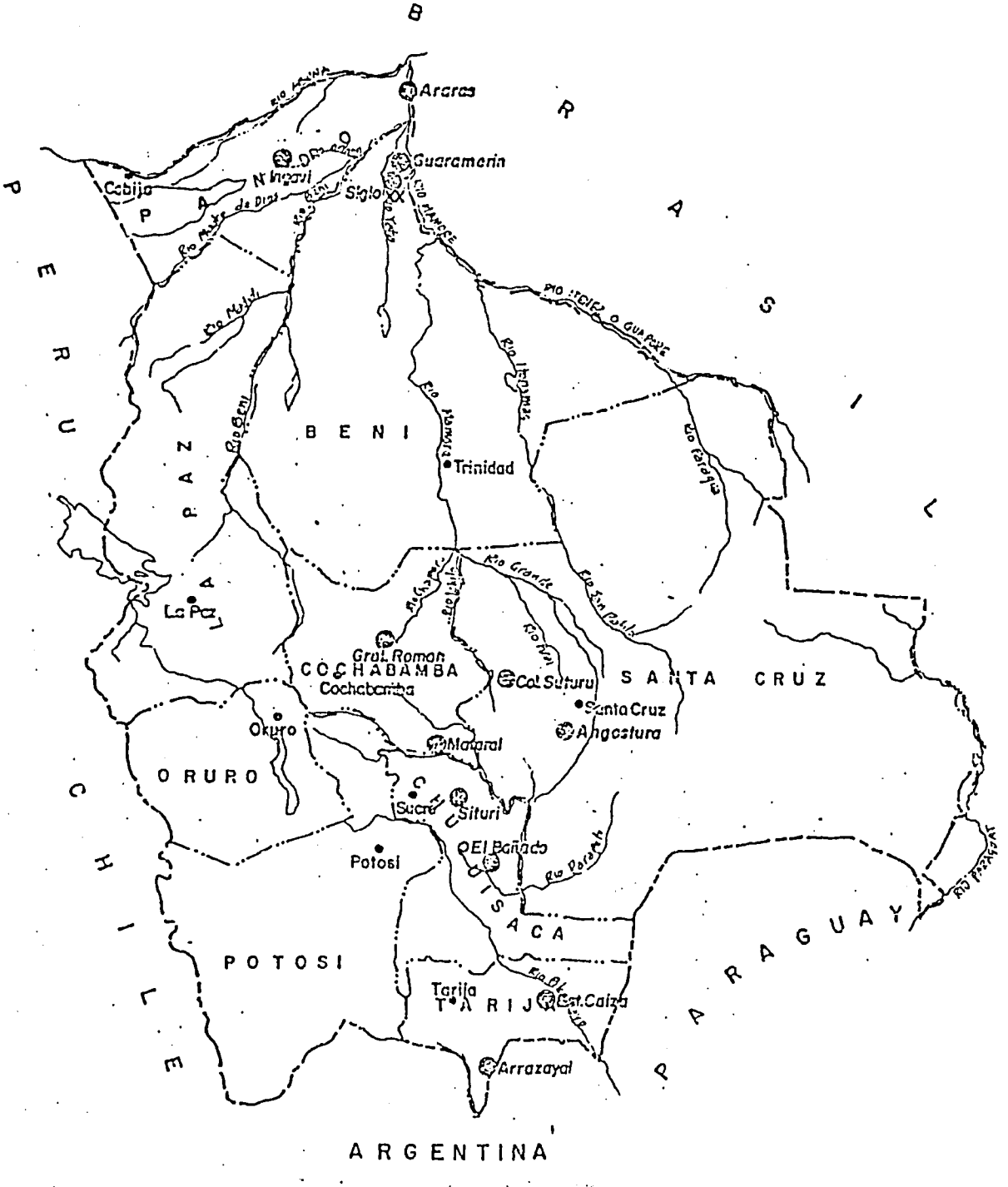

Table 1

LOCALIDADES SELECCIONADAS PARA LA
INVESTIGACION DE SUCEPTIBILIDAD DE ANOPHELINOS
AL DDT/ BOLIVIA OCTUBRE - NOVIEMBRE DE 1986

DEPARTAMENTO	PROVINCIA	LOCALIDAD	BASE DE OPERACIONES	DISTANCIA DE LA BASE DE OPERACIONES	VIA DE ACCESO	FECHAS DE TRABAJO
B E N I	Vaca Diez	Guayaramerín	Riberalta	90 Km.	Terrestre	23 - 25 oct.
B E N I	Vaca Diez	Siglo XX	Guayaramerín	35 Km.	Terrestre	26 - 27 oct.
P A N D O	Federico Román	Araras	Guayaramerín	85 Km.	Terrestre	28 - 30 oct.
P A N D O	Abuná	Ingavi	Riberalta	20 min.	Aérea (Avioneta)	31 - 2 Oct.-Nov.
COCHABAMBA	Campero	Mataral	Cochabamba	300 Km.	Terrestre	4 - 5 Nov.
COCHABAMBA	Chapare	General Roman	Cochabamba	150 Km.	Terrestre	6 - 7 Nov.
SANTA CRUZ	Andres Ibañez	Angostura	Santa Cruz	60 Km.	Terrestre	8 - 9 Nov.
SANTA CRUZ	Andres Ibañez	Surutu	Santa Cruz	70 Km.	Terrestre	10 - 11 Nov.
SANTA CRUZ	Andres Ibañez	Santa Cruz +	Santa Cruz	o		12 - 13 Nov.
T A R I J A	Gran Chaco	Estación Caiza	Tarija	280 Km.	Terrestre	14 - 16 Nov.
T A R I J A	Arce	Arrazayal	Bermejo	20 Km.	Terrestre	17 - 18 Nov.
CHUQUISACA	Zudañez	Situri	Sucre	140 Km.	Terrestre	19 - 21 Nov.
CHUQUISACA	Hernando Siles	El Bañado	Monteagudo	20 Km.	Terrestre	22 - 25 Nov.

+ Investigación de suceptibilidad del Aedes Aegypti al Malathion.

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Table 2

Results of Adult Mosquito Collections, Departments of Beni and Pando, Bolivia, October 23-29, 1986.

Locality	Date	Method	*Species Captured
Guayamerin (SNEM Capture Station #3) Beni	Oct. 23	Biting (26°C)	An. darlingi-1; An. albitarsis-1; Mansonia amazonensis-44; Mn.humeralis-5; Coquillettidia juxtamansonia-10; Cq. venezuelensis-4; Aedes scapularis-31; Psorophora cingulata-2; Ps. ferox-3; Culex quinquefasciatus-2; Cx. nigripalpus-6
Guayamerin (SNEM Capture Station #3)	Oct. 23	CDC LT	An. albitarsis-2; Mn. amaxonensis-1; (Simulium sp.-42)
Guayamerin (San Insidro)	Oct. 24	Biting (24°C)	An. darlingi-1; An. albitarsis-1; An. trianulatus-4; An. peryasui-7; Mn. amazonensis-93; Ae. scapularis-26; Cx. quinquefasciatus-6; Cx. nigripalpus-4
Las Araras Pando	Oct. 25	Biting (23°C)	An. darlingi-3; An. oswaldi-6; An. mediopunctatus-2; An. trinkae-2; Mn. amazonensis-12; Mn. humeralis-3; Ae. scapularis-6; Cx. quinquefasciatus-4
Las Araras	Oct. 25	CDC LT	Mn. amazonensis-1; (Simulium sp.-32)
Las Araras	Oct. 26	Biting (26°C)	An. darlingi-13; Mn. amazonensis-21; Ae. scapularis-3; Cx. quinquefasciatus-8; Cx. nigripalpus-4; Cq. juxtamansonia-1; Cq. venezuelensis-1; Ps. singulata-4
Alto Florida Vaca Diez, Beni	Oct. 29	Biting (25°C)	Negative for anophelines; Mn. amazonensis-72 Mn. humeralis-6; Ae. scapularis-46; Cx. nigripalpus-4; Cq. juxtamansonia-4; Cx. spp.-6

*Identifications made by SNEM Entomologists, Cochabamba Laboratory.

Table 3

Results of susceptibility test with 4% DDT, mixed group of anophelines, Las Araras, Province of Abuná, Department of Pando, Bolivia, October 26, 1986. Temp. 26° C.

	4.0% DDT *			Control		
Species	No. dead	Total tested	Percent mortality	No. dead	Total tested	Percent mortality
An. darlingi	12	12	100%	1	6	17%
An. albitarsis	3	3	100%	0	2	0
An. perasui	1	1	100%	0	2	0
An. trianulatus	4	4	100%	0	0	0
Tot. anophelines	20	20	100%	1	10	10%

* 1 hour exposure; 24 hour mortality.

Table 4

Results of susceptibility tests with DDT, *Anopheles trinkae*, San Mateo Bajo, Province of Chapare, Department of Cochabamba, Bolivia, November 4, 1986. Temp. 23° C., R.H. 56%.

% Concentration *	Rep. I			Rep. II			Rep. III			Total		
	No. dead	Total tested	% mort.	No. dead	Total tested	% mort.	No. dead	Total tested	% mort.	No. dead	Total tested	% mort.
0.25	12	20	60%	3	20	15%	6	20	30%	21	60	35%
0.5	15	20	75%	13	20	65%	4	20	20%	32	60	53%
1.0	8	20	40%	7	20	35%	7	20	35%	22	60	37%
2.0	15	20	75%	17	20	85%	15	20	75%	47	60	78%
4.0	20	20	100%	20	20	100%	20	20	100%	60	60	100%
Control	1	20	5%	5	20	25%	4	20	20%	10	60	17%

* 1 hour exposure; 24 hour mortality.

Table 5

Results of susceptibility tests with DDT, *Anopheles pseudopunctipennis*, Mataral, Province of Campero, Department of Cochabamba, Bolivia, November 7, 1986. Temp. 18° C. - 20° C., R.H. 20%.

% conc. *	Rep. I			Rep. II			Total		
	No. dead	Total tested	% mort.	No. dead	Total tested	% mort.	No. dead	Total tested	% mort.
0.25	0	20	0	0	20	0	0	40	0
0.5	1	20	5%	0	20	0	1	40	2.5%
1.0	1	20	5%	1	20	5%	2	40	5.0%
2.0	1	20	5%	13	20	65%	14	40	35.0%
4.0	18	20	90%	19	20	95%	37	40	92.5%
Control	0	20	0	0	20	0	0	40	0

* 1 hour exposure; 24 hour mortality.

Table 6

Results of susceptibility test with DDT, Anopheles pseudopunctipennis, Pocitos, Province of Gran Chaco, Department of Tarija, Bolivia, November 19, 1986. Temp. 30° C.

% concentration *	No. dead	Total tested	% mortality
2.0	17	20	85%
4.0	20	20	100%
Control	0	20	0

* 1 hour exposure; 24 hour mortality

Table 7

Results of susceptibility test with DDT, Anopheles pseudopunctipennis, Arrazayal, Province of Arce, Department of Tarija, Bolivia, Nov. 20, 1986. Temp. 32° C.

% concentration*	No. dead	Total tested	% mortality
4.0%	20	20	100%
Control	0	20	0

* 1 hour exposure; 24 hour mortality.

Table 8

Results of susceptibility test with DDT, Anopheles pseudopunctipennis, Rio Pojo colony (54th generation) SNEM insectary, Cochabamba, Bolivia, November 1, 1986. Temp. 24° C., R.H. 20%.

% concentration *	Number dead	Total tested	% mortality
0.25	1	20	5%
0.5	1	20	5%
1.0	4	20	20%
2.0	12	20	60%
4.0	7	20	35%

*1 hour exposure; 24 hour mortality.

Table 9

Susceptibilidad del Vector An. pseudopunctipennis al DDT.

Abreviaciones: F_1 = Primera Generacion Filial;
HdC = Las hembras del Campo.

Criterio para dosis diagnostica (DDT a 4%)

S = Susceptible Mort. 98 - 100%

V = Verificación requerida Mort. 80 - 97%

R = Resistente Mort. por debajo del 80%

Fecha	Localidad	Linaje	Mortalidad	Papel usado	Criterio
24-07-82	Chaco, Z-III, frente	HdC	100	2 veces	S
01-02-83	Chaco, Z-III, pueblo	F_6	86	1 vez	V
16-12-82	Estación Caiza, Z-VII	HdC	72	2 veces	R
01-02-83	Estación Caiza, Z-VII	F_2	77	1 vez	R
09-02-83	Río Pojo, Z-II	F_1	76	1 vez	R
14-09-83	La Aguada, Z-II	F_{10}	90	1 vez	V
14-09-83	Chaco, Z-III, pueblo	F_{22}	44	1 vez	R
16-09-83	Masicurí, Z-VI	F_1	75	1 vez	R
28-09-83	Chaco, Z-III, frente	HdC	93	1 vez	V
29-09-83	Chaco, Z-III, frente	HdC	97	1 vez	V
01-10-83	Chaco, Z-III, frente	HdC	100	1 vez	S
18-10-83	Chaco, Z-III, frente	F_1	93	1 vez	V
28-10-83	Río Pojo, Z-II	F_{14}	70	1 vez	R
18-11-83	Río Pojo, Z-II	F_{15}	97	1 vez	V
19-11-83	Río Pojo, Z-II	F_{15}	99	1 vez	S
19-11-83	Tatarenda, Z-VII	HdC	79	1 vez	R
07-12-83	Río Pojo, Z-II	F_{17}	90	1 vez	V
12-12-83	Palmar Chico, Z-VII	F_1	86.7	1 vez	V
22-01-84	Tatarenda, Z-VII	F_3	80	2 veces	V
25-05-84	Sarcocucho, Z-II	F_1	98	1 vez	S
01-10-84	Chaco, Z-III, frente	HdC	83	2 veces	V
16-10-84	Sarcocucho, Z-II	F_3	86	3 veces	V
27-11-84	Saipina, Z-VI	F_{18}	85	1 vez	V
06-11-84	Palmar Chico, Z-VII	F_2	85	1 vez	V
29-11-84	Palmar Chico, Z-VII	F_3	43	2 veces	R
12-12-84	Guayabos, Z-III	HdC	98	1 vez	S
03-01-85	Chaco, Z-III, pueblo	F_{43}	22	2 veces	R
04-01-85	Chaco, Z-III, pueblo	F_{43}	45	1 vez	R
10-01-85	Saipina, Z-VI	F_{20}	53	1 vez	R
15-01-85	La Aguada, Z-II	F_{30}	96	1 vez	V
23-01-85	Guayabos, Z-III	F_2	100	1 vez	S
30-01-85	La Plazuela, Z-IV	F_{26}	97	1 vez	V
05-02-85	La Aguada, Z-II	F_{31}	75	2 veces	R
14-02-85	Masicurí, Z-VI	F_4	99	1 vez	S
18-02-85	Pescadito, Z-VI	F_4	84	1 vez	V
21-02-85	Pescadito, Z-VI	F_4	83	1 vez	V
26-02-85	Mataral, Z-II	F_3	98	1 vez	S
02-04-85	Peñones, Z-VI	F_6	50	1 vez	R

Table 10

Results of susceptibility test with 5.0% malathion, Aedes aegypti, third Ring, City of Santa Cruz, Bolivia, November 13, 1986. Temp. 32° C.

Exposure time	Number dead	Total tested	% mortality
30 min.	20	20	100%
1 hr.	20	20	100%
Control	0	20	0

Specimens were collected over a 2-day period from Potosi Tire Service (Recauchutadora Potosi).

Table 11

Results of susceptibility tests with 5% malathion, Aedes aegypti, Santa Cruz colony (31st generation) SNEM insectary, Cochabamba, Bolivia, November 2, 1986. Temp. 22° C., R.H. 25%.

Exposure time	Number dead	Total tested	% mortality
15 min.	15	20	75%
30 min.	17	20	85%
1 hr.	20	20	100%
2 hrs.	20	20	100%
Control	0	20	0

Table 12

Results of susceptibility test with 1.0% Sumithion, Aedes aegypti, Santa Cruz colony (31st generation) SNEM insectary, Cochabamba, Bolivia, November 2, 1986. Temp. 22° C.; R.H. 25%.

Exposure time	Number dead	Total tested	% mortality
30 min.	20	20	100%
1 hr.	20	20	100%
Control*	0	20	0

*Shared with malathion test, Table 11.