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Annual Progress Report

Year 1 June 71 - June 72

PD-AAL-140

TO: Office of Science and Technology
U.S. Agency for International Development
Washington, D.C. 20523

SUBJECT: Annual Research Report

1. Project Title: Ecological studies on *Aedes aegypti* in East Africa Preliminary to Genetic Control. Contract No. AID/csd-3159

2. Principal Investigator and Contractor:
Profs. George B. Craig, Jr. and K. S. Rai
Department of Biology
University of Notre Dame
Notre Dame, Indiana 46556

3. Contract Period: 3 years, 1 July 1971 - 30 June 1974

4. Period Covered by Report: 1st year, 1 July 1971 - 30 June 1972

5. Total A.I.D. Funding of Contract: $258,900.00

6. Expenditures - Previous Year: None

7. Expenditures - Current Year: $86,300.00

8. Expenditures - Next Contract Year: $86,300.00

9. Narrative Summary - Accomplishments:

In its first year, the project concentrated on recruitment and training of personnel, establishment of a laboratory and development of a suitable field site for ecological investigations. All have been accomplished. The project has 3 scientists and 10 Kenyan technicians and field aides. A splendid laboratory at Nyali, just north of Mombasa, has been leased and equipped. The field site at Rabai, just west of Mombasa, consists of numerous rural villages, with indoor-breeding *A. aegypti*, plus adjacent forests and rivers, with treehole-breeding *A. aegypti*. Both populations are under intensive monitoring; marking experiments have shown that the two populations are distinct and do not mix. Considerable life table data for the two forms was developed. Chromosome translocations, potentially useful in genetic control, were induced and isolated from local domestic populations. Finally, we have had marked assistance and cooperation from the Rabai people; we are especially grateful both to them and to the Kenya Medical Department for friendly relationships and significant material aid given to this project.

ANNUAL RESEARCH REPORT

A. GENERAL BACKGROUND

Aedes aegypti is a pervasive and significant vector of human disease through most of the tropical and subtropical world. It transmits yellow fever, dengue, Asian hemorrhagic fever, Chikungunya and other viruses to man; it may also play a role in epidemiology of filariasis (elephantiasis). Larvae breed in containers that are inaccessible to conventional mosquito control techniques such as drainage and larviciding. Human activities such as urbanization, water storage and poor sanitation have increased the threat of this mosquito. Vast eradication campaigns directed against this species have failed dismally, in large part because control weapons are inadequate.

In East Africa, Aedes aegypti is widespread and abundant; at present, even minimal control is not feasible in most areas. To compound the problem, there is evidence of two populations within the species. One occurs in villages, breeding in domestic water containers and throw-away containers such as tin cans and rubber tires. The other is feral, breeding in tree-holes and sylvan situations that are far from human habitation. The relative importance of these two forms to disease transmition and control and the genetic relationships between them have never been established.

In recent years, entomologists have been searching for alternative, non-chemical control methods which might avoid the twin hazards of insecticide resistance and environmental pollution. Among the most promising biological approaches is genetic control, which involves turning a species against itself. While the best-known method is the "sterile-male technique," where released radiation-sterilized males compete for field females, there are many other methods of genetic manipulation which could achieve a similar end. For Aedes aegypti, chromosome translocations, sex ratio distortion, genes for sterility and other genetic devices have shown marked promise in the laboratory. However, field research and trials are required to allow proper evaluation of these newer methods.

B. PROJECT OBJECTIVES

The long-term goal of this project is to discover whether genetic control of Aedes aegypti is feasible under East African conditions. However, initial research must be primarily ecological and bionomic. Before we attempt genetic manipulation, we must accumulate information on population dynamics, dispersal and reproductive biology of the specific population(s) under study. The following are specific research objectives for the three-year period of this contract.

1. Develop a study area where the population of A. aegypti is thoroughly, intensively and minutely known. (By analogy, one might point to the careful, long-term, intimate observations of the mountain gorilla by Schaller and of the chimpanzee by Lawick-Goodall).
   a. Discover the absolute number of mosquitoes in the area and determine fluctuation on a weekly, seasonal and annual basis.

b. Develop predictive life tables on a specific population, using data on fecundity, mortality, growth rate and ecological parameters.

2. Establish 10-20 replicate villages (ecological islands) which can be used for later releases. Monitor each population for at least one year.
3. Determine the genetic and ecological relationships between indoor-breeding and feral populations of A. aegypti.
4. Develop chromosome translocations and other genetic mechanisms potentially useful for genetic control.
5. Develop facilities for mass-production of mosquitoes.
6. Make preliminary field releases of laboratory-reared mosquitoes to develop technical methods.
7. Conduct subsidiary bionomic research on related mosquito disease vectors, especially Aedes simpsoni.

ANNEX 1 gives a breakdown of 9 specific projects designed to accomplish these objectives. The list includes project description, staff assignments, consultants required and supportive research to be conducted at the University of Notre Dame.

In addition to scientific objectives as listed above, other objectives of the contract include:

1. Develop a major research component, the Mosquito Biology Unit (MBU), of the International Centre for Insect Physiology and Ecology (ICIPE).
2. Provide training opportunities for African scientists and technicians.
3. Serve as a mechanism for strenghtening the East African Community by affiliation with both research institutes and universities.

C. CONTINUED RELEVANCE OF OBJECTIVES

The objectives of this project, as stated in the Project Statement of March, 1971, remain unchanged. After a year of operation of this Contract, it is evident that the original objectives were reasonable and are attainable in the three-year period.

As recognized at the initiation of this Contract, the funding and the 3-year period are not adequate to develop a final answer to the question, "Is genetic control of A. aegypti feasible?" However, we believe we are making marked progress in developing a baseline for a sound approach to this question.

D. ACCOMPLISHMENTS TO DATE

Description of the present facilities:

The MBU is established in an old 14 room mansion in Nyali, north of Mombasa. The house is rented jointly with W. Hausermann, the MBU Field Director, who inhabits four rooms of the upper floor. One large room on the same floor serves as library-office, another room with adjacent bathroom as a consultant's room. On the ground floor MBU occupies three rooms of the former kitchen tract as insectaries and one large room as general laboratory. The former garage serves as a workshop in which most of the laboratory furniture is made by one of the staff under the guidance of a research scientist. This workshop will later on serve as mass rearing insectary. Two other, smaller rooms are used as store and animal room. One other large room with adjacent bathroom is available as self contained guest laboratory to research scientists of ICIPE, the University of Nairobi and the Division of Vector Borne Diseases of the Kenya Ministry of Health. It is expected that one or two more research units of ICIPE may take up permanent residence in the MBU laboratory, The MBU would then be designated as the ICIPE Coastal Research Centre.

The staff of MBU consists currently of 3 research scientists, one of which is the field director, 4 laboratory technicians, 1 carpenter, 4 field assistants and various casual helpers for fieldwork as required. See detailed staff list in ANNEX 2.

Due to the move of the MBU laboratory to the new premises and also to the slow arrival of the two research scientists MBU had a comparatively slow start as regards research work (see ANNEX 3, December Report). However, the ancillary staff is now adequately trained for routine work and most of the equipment and supplies are assembled, so that now more time can be devoted to research than a few months ago.

For a geographic description of the coastal area with maps see the Report of December 1971 in ANNEX 3. Some evidence for the presence of two distinct A. aegypti populations - one indoors and one outdoors - was also given in this report.

Research Findings

Project 1: Routine monitoring of A. aegypti in Rabai location.
(responsibility: W. Hausermann)

A brief description of the routine work is given in ANNEX 4 which includes also a map of the area. Added as ANNEX 5 is a short description of the location Rabai. An interim report on the routine monitoring is enclosed as ANNEX 6 (see also ANNEX 3).

Findings:

Indoor population of A. aegypti:

The population is rather stable throughout the year. Wide fluctuations occur within single houses or in clusters of houses, due to human interference. Climatic changes seem to be of little importance.

Outdoor population of A. aegypti:

The only reasonable way to sample the outdoor population proved to be the ovitrap method. Adult catches, either by the landing-biting method or by handnet were not successful during most of the time. Sampling of natural larval habitats was equally unrewarding. The adult outdoor A. aegypti population was negligible during dry periods and building up about 14 days after substantial rains from eggs stored in dry larval habitats.

Seasonal fluctuations of A. aegypti populations have been described more recently by Trpis (WHO/VBC/71.291). The uniqueness of the situation in Rabai (and other places on the Kenya Coast like Likoni, Junju, Kaloleni, Chony etc.) consists in the fact that two populations or subpopulations occur side by side, one going through 12 or more generations a year, the other through only three or four.

Interpretation of data

The accumulated data will be statistically evaluated only, when the observation series is finished.

Research design:

Originally it was intended to monitor the A. aegypti populations of the study area with a minimum effort just sufficient to give background information for the planning and evaluation of specific experiments. During the visits of the consultants Drs. Trpis and Crovello the design was expanded to its present level (ANNEX 4) with the hope to gain additional data for the construction of lifetables. The experience gained up to today made it evident that it would be more practical to reduce the routine monitoring to determination of oviposition outdoors and container index indoors. All other information could be collected more efficiently in specific experiments. It is planned to continue routine monitoring at least until the end of this year at this reduced level.

Within the framework of the routine monitoring 10 villages in Rabai location have been designated as experimental villages. More villages can be selected in short time. A standard procedure to describe such villages on "mosquito terms" and permitting quantitative comparison is developed at present. Special experiments have been carried out only in three villages so far. Establishing this standard proceuure of description and comparison needs experiments which can now be carried out based on the experiences gained during the routine monitoring and the lifetable project.

Project 2: Preparation of lifetables for indoor and outdoor populations of A. aegypti (responsibility: W. Hausermann)

Preliminary results of a mark-release-recapture experiment with an indoor population are presented in ANNEX 7.

Findings:

1. The distribution of adult A. aegypti is positively correlated to the breeding level in different houses.
2. Males disperse more evenly than females, who accumulate in houses with good breeding.

3. Life expectancy of males is - as in the laboratory - much less than that of females.
4. Some females were observed to finish a gonotrophic cycle in the field within 3 days, while other females tried to get a bloodmeal for five and more days.
5. Females frequently use more than one bloodmeal to pass through one gonotrophic cycle and are landing on man (and take a bloodmeal) in any stage during their gonotrophic cycle.
6. The indoor population in the study village of 16 houses was estimated to be of the size of 300 - 800 individuals at any given day of the observation period.

Interpretation of data:

The above findings are based entirely on various arrangements of the raw data on capture and recapture of some 3,000 mosquitoes during a period of 40 days. A statistical analysis and the calculation of lifetable parameters will follow in cooperation with Dr. Crovello from UND. Special attempts will be made to compare the accuracy of various methods of data evaluation (Jolly, 1963, 1965; Leslie, 1951, 1952; Bailey, 1951, 1952; Fisher-Ford, 1947; Jackson, 1939).

Research design:

For the construction of a lifetable additional specific questions have to be answered in specific experiments. One such experiment will be to measure dispersal of males and females immediately after eclosion and before hosts become attractive and a landing-biting catch can be made. It must also be established when and where females get inseminated (see preliminary experiment in ANNEX 7).

Another series of observations has to be made in order to refine the recognition of the stage of the gonotrophic cycle without dissection.

A further experiment will be to measure oviposition in an experimental village by replacing water jars in regular intervals.

Projects 3 and 4: Environmental definition of indoor and outdoor habitats and ecological relationships between domiciliary ("indoor"), peridomiciliary ("outdoor") and extradomiciliary ("forest") habitats (responsibility D. Fanara).

Findings:

1. *Aedes aegypti* utilizes the largest variety of possible breeding compared with 6 other species of *Aedes* (*Stegomyia*) inhabiting the Bejuma forest (ANNEX 8).
2. Only the dark color form of *Aedes aegypti* was recovered from forest breeding sites (ANNEX 8).
3. Indoor landing-biting and rearing of larvae from water pots indicate that Momsabu village on the edge of Bejuma forest has only the intermediate color form of *Aedes aegypti* present (ANNEX 9).
4. Peak biting in huts at Momsabu occurred in the morning hours with biting recorded throughout the day (ANNEX 9).

5. Forest landing-biting at Bejuma resulted in a capture of Aedes aegypti in the following proportions: (ANNEX 9)

8.5 % intermediate color males
21.3 % intermediate color females
10.6 % dark color males
59.6 % dark color females

6. Sex ratios (male : female) for Aedes aegypti taken in landing-biting were:

3 : 6 for intermediate color forms taken indoors
3 : 7 for intermediate color forms taken in the forest
1 : 7 for dark color forms taken in the forest (ANNEX 9)

7. In a mark and recapture study it was determined that the indoor breeding, intermediate color forms of Aedes aegypti move throughout the village under study for a distance of about 90 meters (ANNEX 10).
8. It was also shown that the indoor breeding, intermediate form will travel up to 237 meters to visit the forest situation. Both a male and a female moved from the hut into the forest (ANNEX 10).

Relevance:

In terms of a control project based on a genetic translocation release the following points can be made concerning the 8 listings under accomplishments:

1. In the forest situation Aedes aegypti utilizes a vast number of breeding sites (by comparison with other species) and this may be an important issue in a control project.
2. Because only the dark color form of Aedes aegypti was observed in rearings of forest collected larvae it may be that the paler indoor breeding form does not enter the forest for ovipositing. This would be an important point, if universally true, in the planned genetic control of Aedes aegypti.
3. The presence of an intermediate rather than pale form indoors suggests some gene pool mixing with the dark forest variety, however, no dark forms were ever taken indoors.
4. An understanding of biting habits is important in the evaluation of the bionomics of a potential disease vector.
5. Although only the dark color form of Aedes aegypti was reared from forest breeding sites, it was shown that the intermediate color form was present in the forest in about 1/3 the numbers of the dark form. This was at least true for human biting, however, the dark form may not feed on man as readily as the intermediate form.
6. The difference in sex ratios from landing-biting studies indicate that the dark form males of Aedes aegypti do not readily come to man. This may indicate that man is not the common host of this variety. Points 5 and 6 are clues to understanding the bionomics of the forest breeding Aedes aegypti and the eventual determination of its relationship to the indoor breeding variety.

7,8. The degree of movement by the indoor form of Aedes aegypti and the forest forms is an important item for the logistics planning of a pilot genetic marker release or a genetic translocation release.

Project 5: Genetic relation between indoor and outdoor populations. (responsibility W. Hausermann)

The genetic studies are not advanced enough yet to write an interim report. Some time, however, was spent to describe the variability in

coloration of "indoor" and "outdoor" specimens in order to permit the clear assignment of field collected material to either one of the two populations present in the area. The characters on which the distinction is based are described in ANNEX 11.

Findings:

1. Indoor and outdoor A. aegypti can be distinguished on the basis of their blackness. The indoor form is pale or brownish; the form breeding commonly in outdoor habitats is coal black. The basis of distinction is the pigmentation of the cuticle of mesonotum, postnotum and pleurites, and the pigmentation of the black scales on mesonotum and abdomen. Also the whiteness of the white scales on the abdomen may differ, the "white" being more creamcolored in pale specimens. The black outdoor mosquito corresponds largely with the description of A. aegypti formosus (Mattingly, 1957) except that white scales are sometimes found on the first abdominal tergite.

   The paler indoor mosquito varies widely in its coloration between a dark A. aegypti aegypti and A. aegypti var. queenslandensis. Pale indoor specimens without white or pale scales on first abdominal tergite or the back of the head were also observed. Thus indoor and outdoor A. aegypti do not clearly fall into the categories of A. aegypti aegypti and A. aegypti formosus respectively.
2. Preliminary observations on the frequencies of common mutants like spot abdomen (s), gold, brown and pale mesonotum (G) and speck palp (Sp) indicate different gene frequencies for black and pale A. aegypti. G and s, at least during the dry season, were only found in domiciliary, indoor habitats. Sp was at the same time more common in samples from peridomiciliary, outdoor, habitats.
3. In their oviposition habits, pale domiciliary populations are quite distinct from black peridomiciliary populations. No oviposition of pale mosquitoes was observed in ovitraps placed outdoors although black mosquitoes oviposited regularly in ovitraps outside houses and pale mosquitoes move freely between houses and away from villages (see ANNEXES 6, 7, 9 and 10). This separation between the two populations on the basis of oviposition may not be maintained during periods of rain when air humidity is high, oviposition sites abundant and the cloud cover is dense.

Research designs:

Since the project is still at its beginning there are no reasons to modify the research design.

It is foreseen to follow up with a specific study on gene frequencies for up to 10 of the most common mutants in each of the two populations. This gene frequency analysis will be coordinated with the research projects of a ICIPE research scientist, who is coming out next year. This scientist will be specialized in population genetics and will determine the isozyme polymorphisms in populations by electrophoresis. Attempts will also be made to demonstrate gene flow between the two populations with the use of genetic markers.

Related to our studies on the genetic relation between domiciliary and peridomiciliary A. aegypti experiments on host preference will be made in the Vector Biology Laboratory of the University of Notre Dame. Contact has also been made with Dr. Mukwaya and Mr. Sempala of the E.A. Virus Research Institute in Entebbe, who are interested in the genetics of host preference of Stegomyia species.

Experiments on the vectorial capacity of A. aegypti for human filariasis have already been started by Dr. W. Kilama of the Medical School of the University of Dar es Salaam, who spent two weeks as MBU's consultant in Mombasa. Further cooperation is planned.

Studies on vectorial capacity of A. aegypti for saurian malaria have been started by a graduate student from Notre Dame, Mr. L. Munstermann, who is residence at MBU for the summer of 1972.

Projects 6 and 7: Production of translocation heterozygotes and selection of homozygotes and genetic engineering of release strains for translocations and sterile males (responsibility: P. McDonald)

At the same time that studies of the ecology of the target insect species are being undertaken, chromosomal translocations are being developed as genetic control mechanisms for future releases. Experience dictates that in addition to carrying the genetic mechanism the released individuals must be field adapted if they are to be effective. A report on the production of translocations and an inventory of the translocation stock developed so far is given in ANNEX 12.

Findings:

To date thirty-nine translocations have been induced and isolated by genetic methods. Thirty-one have been induced in an indoor strain. These translocations are each being maintained by repeated selection from genetically marked stocks. Seven translocations have been tested to isolate homozygotes which have greater potential for genetic control than the heterozygotes. None of the seven have given homozygotes to date.

The multiple-marker stocks used for maintainence of translocations are undergoing a process of genetic assimilation (Africanization) with both indoor and outdoor collected material. At present, three different marker strains with 75 percent local genome are being increased to 88 percent local genome for the indoor strains. Three different marker strains with 50 percent local genome are being increased to 75 percent local genome for the outdoor strains. These strains will allow for easy maintainence of translocations with an ever increasing genetic identity to the naturally occurring populations.

E. DISSEMINATION AND UTILIZATION OF RESEARCH RESULTS

1. Since this is the first year of the contract period, no research publications have yet appeared. Several projects are near completion and manuscripts are in preparation. One semi-popular article has appeared:

   Conner, W. 1972. MBU-N.D. and mosquitoes in Africa. Notre Dame Science Quarterly X(3):4-6.

2. The MBU has played an important role in the development of the International Centre for Insect Physiology and Ecology in Nairobi. Annex 14 shows the organization of ICIPE; initially, MBU was the largest single research component, albeit the ICIPE is now growing rapidly and this is no longer the case. Nevertheless, this A.I.D. contract came at an essential time in the gestation of ICIPE and the stimulatory role has been larger than the size of the contract.

   All three MBU Research Scientists have given ICIPE evening seminars in Nairobi, which were open to scientists from various research institutes and the University of Nairobi.

Speaker	Title	Date
W. Hausermann	Problems in genetic control of *A. aegypti*	5 May 71
T. Crovello (Assoc. Prof., U. Notre Dame)(MBU consultant)	Life tables and strain variation in *A. aegypti*	5 Jan. 72
P. T. McDonald	Chromosomal rearrangements and genetic control	2 Feb. 72
D. Fanara	Population dynamics of aquatic communities under biological and chemical control regimes	3 Mar. 72

Dr. Paul McDonald represented MBU and ICIPE at the Annual Scientific Conference of the East African Medical Research Council in January, 1972. His talk was entitled "Ecological studies on *Aedes aegypti* preliminary to genetic control".

In addition to visits by the Principal Investigators, Professors Craig and Rai, the MBU used the services of the following consultants:

October, 1971 - Dr. Eugene Gerberg, Director, Insect Control and Research, Inc., Baltimore, Md. Gave valuable advice on siting of the laboratory, laboratory design and mass production of mosquitoes. An exceptionally valuable consultant whose services will be requested again in the coming year.

January, 1972 - Professor Theodore Crovello, Univ. Notre Dame
Research Fellow Milan Trpis, Univ. Notre Dame
Dr. Crovello is an authority on biomathematics, population dynamics and experimental design. Dr. Trpis is an authority on mosquito ecology, in East Africa and elsewhere. The

working plan (Annex 1) was designed by the Research Scientists in collaboration with Crovello and Trpis. Both of these Consultants continue to conduct research at Notre Dame related to this contract. Both will return to Mombasa during the coming year.

April, 1972 - Mr. George Lawrence, Accounting Office, Univ. Notre Dame, visited both MBU and the home office of ICIPE in Nairobi. Mr. Lawrence advised on bookkeeping procedures and financial management. Since this contract has been one of the first to go through ICIPE, it has been the source of procedures for handling other contracts. Mr. Lawrence was particularly helpful in this respect. He worked effectively with all parties.

June, 1972 - Dr. Wenceslaus Kilama, Lecturer in Parasitology, Medical School, Univ. Dar-es-Salaam, served as Consultant on bionomics of Aedes simpsoni and on genetics of vectorial capacity. Laboratory research at Notre Dame has shown that, contrary to textbook information, certain East African strains of Aedes aegypti are highly susceptible to human filarial worms. We are undertaking a joint program with Dr. Kilama to determine field epidemiological significance of this observation.

3. The following are examples of relationships that have contributed to effective cooperation between MBU and various entities in East Africa:

   a. We have had splendid cooperation with the Division of Vector-Borne Diseases, Kenya Medical Department. The Director, Dr. J.D. Roberts has given us every support and encouragement. For the first six months of this contract, we used a house belonging to DVBD as our laboratory (see Annex 3). No rent was charged and we were made most welcome. All of our scientists have been trained in mosquito identification by Mrs. E.C.C. VanSomeren of DVBD. We hope to have Mrs. VanSomeren come to Mombasa as a Consultant during the present contract year, to advise us on developing a reference collection. Mr. George Oketch, Field Entomological Supervisor for the Coastal Region, DVBD, has given us frequent and valued advice on mosquitoes on the coast and has paved the way for our collectors to work in villages.

   b. Mr. Vijay Prashar of the DVBD trained at Notre Dame for one year, February 1971 to February 1972, working as a graduate student in the Vector Biology Laboratory under a WHO Fellowship. Since his return to Nairobi in February, Mr. Prashar has continued to cooperate with the MBU team, especially in developing a study on genetic variability in A. aegypti and A. simpsoni.

c. Dr. Louis Mukwaya and Mr. S. Sempala of the East African Virus Research Institute, Entebbe, Uganda, expect to visit MBU in connection with their own research on bionomics of Stegomyia mosquitoes. In 1971-72, G.B. Craig was Principal External Examiner for the Ph.D. thesis of Dr. Mukwaya at MaKerere University. The thesis was on genetics of zoophilic and anthropophilic behavior in Aedes simpsoni, a topic closely related to MBU interests.

d. Dr. Wenceslaus Kilama, Lecturer, Medical School, University of Dar-es-Salaam, has served as an MBU Consultant. In addition, we are developing a joint program with Dr. Kilama on strain differences in ability of Aedes aegypti to transmit filariasis.

e. Personnel from MBU have visited the East African Institute of Vector-Borne Disease, Amani, Tanzania, to consult with the Director, Philip Wegesa, and with Dr. John Reybold on filariasis. In addition, Director Wegesa and G.B. Craig are working as joint organizers on a symposium to be held in 1973 on "Variability in East African Bloodsucking Diptera as it Affects Disease Transmission". We hope that this symposium will be supported by ICIPE and that it can be held in conjunction with the East African Medical Research Council Annual Scientific Meeting, Kampala, January, 1973. This symposium should serve to draw together all of the medical entomologists in East Africa.

f. Personnel from MBU have visited the East African Tropical Pesticides Research Institute, Arusha, Tanzania, to consult with the Director, Dr. Materu, on construction of experimental huts and field population cages.

g. We are developing plans with Dr. Canute Khamala, Lecturer in Entomology, University of Nairobi, for a joint study on variation in morphology of Aedes simpsoni at different altitudes on Mount Kenya.

h. Dr. Reuben Olembo, an insect geneticist and Head, Botany Dept., University of Nairobi, has been invited to serve as Visiting Professor at Notre Dame during the spring of 1973.

i. In June, we were honored by a visit by Mr. Towett, Minister of Education for Kenya. Mr. Towett toured the laboratories and talked to our personnel in the afternoon, then spent a social evening with MBU scientists. We had valuable discussions on the training of Kenyans as technicians and research aides.

j. MBU has served as a base of operations for ICIPE scientists from Nairobi interested in insect populations on the coast. We provide laboratory space, transportation and, at times, living quarters for periods of up to ten days. A nominal fee is charged. Examples of recent visitors and their interests:

Prof. Thomas Odhiambo and Mr. Joel Ojal
ICIPE Administration
Prof. J. DeWilde and P. Scheltes
Diapause in coffee insects
Prof. Detrich Schneider
Olfaction in Lepidoptera
Prof. Carroll Williams
Hormones in tropical insects
Prof. David Smith
Sensory physiology in Lepidoptera
Dr. William Wood
Chemistry of tsetse flies and ticks
Prof. Koji Nakanishi
Insect chemistry
Prof. Jerrold Meinwald
Insect chemistry
Dr. Reinhard Leuthold
Termite caste determination

F. STATEMENT OF EXPENDITURES

The budget for the first year of operation is as follows:

Expenditure	3 year total	Year 1 (July 71 - June 72 Proposed	Actual	Remainder
AT NOTRE DAME				
1. Salaries & Benefits	18,960	31,385	25,071	6314
2. Supplies	9,405	3,000	1,114	1886
3. Bank Charges	1,500	500	19	481
4. Travel	20,600	6,867	9,073	-2,206
5. Indirect - UND	12,935	4,102	3,343	759
	143,400	45,854	38,620	7,234
IN KENYA				
6. Salaries & Benefits	21,000	7,000	4,605	2,395
7. Housing Allowance	10,800	3,600	3,380	220
8. Rental-Space	12,000	4,000	4,521	- 521
9. Supplies	7,695	2,500	5,399	-2,899
10. Travel	12,000	4,000	3,215	785
11. Capital	28,905	12,000	9,222	2,778
12. Indirect - ICIPE	23,100	8,275	8,226	49
	115,500	41,375	38,568	2,807
TOTAL:	258,900	87,229	77,188	10.041

There are several reasons for the budgetary surplus at the end of the first year. The most important of these is the diligent work by Dr. Hausermann to hold expenditures down; we were aware of the pressing need for a radiation source (see section F) and needed to save money for it. The savings in salary of scientists is due to the fact that Dr. McDonald joined in September and Dr. Fanara joined in December, thus saving 7 man-months. The deficit in travel is due to the large amount of travel required in year 1, to get the scientists to the duty station and to have enough consultants to get the project off to a proper start. The savings in Kenya salaries, item 6, was due to slow hiring and training of staff; we did not want to start with a full complement of inexperienced people.

There were no significant budgetary problems.

G. WORK PLAN AND BUDGET FORECAST

1. The work plan for the coming year 2 of the contract) is given in ANNEX 1. The following are anticipated accomplishments:

   Project 1. (Population fluctuation) The gathering of data

will be terminated by January, 1973. A final report on the bionomics of A. aegypti in the Rabai area, with cross references to A. aegypti elsewhere in Africa, should be ready by June, 1973. This information will provide a background for the results of coming field experiments on population manipulation. Note that monitoring of populations will continue throughout the year in the proposed experimental villages.

Project 2, 3, 4 (Life tables and bionomics). These projects will be completed by June, 1973, and final reports will be available by the end of 1973.

Project 5 (Feral vs. domestic populations) This project is not yet far advanced far enough to allow predictions about termination. However, significant statements about the amount of gene exchange between the two populations will be feasible by June of 1973.

Project 6 (Chromosome translocations) We hope to produce translocation homozygotes, with the Rabai genetic background, during the coming year. Chances of getting any given translocation in the homozygous condition are small; hence, the date of termination of this project is unpredictable. If we are unable to develop a homozygote in a local strain, we will use homozygotes maintained at Professor Rai's laboratory at Notre Dame for release.

Projects 7 and 8 (Genetic engineering and mass prodution) While these projects have barely started, we hope that improved and still improving efficiency of our laboratory technicians will allow us to carry out the necessary crosses for Africanization of our marker and translocation strains before the end of contract year 2. By June of 1973, initial problems of mass rearing should be solved and small releases will be made to test, improve and adapt the release strains for translocations, sterile males and markers. If the releases are successful, the translocations and sterile males will also be used to determine the reproductive potential of the indoor breeding population under genetic stress.

Project 9 (Release strategies) will only be started after June, 1973.

2. The procedures to be used and activities to be carried out in year 2 are similar to those of year 1.
There are three significant additions:

a. We propose to purchase a portable radiation source, Cesium-137, that will deliver 50,000 r of gamma per hour and weigh only 1200 pounds (J. L. Shepherd Co., Glendale, Calif., Model 109-CS-1; $8200, including shipping). This unit, which has a high safety factor, will be used both to produce radiation-sterilized males for release and to induce chromosome translocation. Initially, we had hoped to obtain this irradiator via the International Atomic Energy Agency. However, we have

found that IAEA deals only with governments and would not be able to furnish apparatus or funds directly to ICIPE or to this project. In the past, control experiments with radiation-sterilized mosquitoes have failed, in part because the radiation weakened the males and reduced vigor. However, very recent research at Notre Dame has shown that when pupae are irradiated under nitrogen, competitiveness of resulting males is much improved. We wish to use irradiation-induced sterility as a marker, to determine effectiveness of our released males. There is even a chance that they could achieve control. In our experiments on radiation-induced translocations, we now have to travel to Nairobi to use an X-ray source, irregularly available, at Kenyatta Hospital. Males often die in transit. Research would be greatly facilitated with a source at MBU. Following the termination of this contract, the radiation source will be given to ICIPE, Nairobi, for additional research on radiation effects on insects.

b. In February of 1972, we expect to have an additional scientist in MBU. This postdoctoral geneticist-biochemist would be a member of the MBU team but would be stationed at the ICIPE laboratories in Nairobi. He would conduct research on genetic variability in East African populations of Aedes mosquitoes as measured by electrophoretic determination of isozyme polymorphisms. As part of an application to the United Nations Development Program, ICIPE has requested $20,000 per year for four years (to February, 1977) for this project, under supervision of G. B. Craig. The proposed postdoctoral already has been selected and is in training at Notre Dame. He would serve as a valuable liaison between MBU and the ICIPE home laboratories. Although his research will contribute markedly to the ends of the present contract, especially in solving the indoor vs. outdoor population enigma, the electrophoresis project will not impose any additional cost on the AID contract.

c. The coastal region of Kenya is so rich in insect life and the laboratory facilities at MBU are so good that ICIPE has designated our center as its Coastal Research Centre, with Dr. Hausermann as Director. There is a good possibility that one or more additional ICIPE projects will be based with us. Certainly, there is ample space in our 14-room building. Such projects would share in rental payment and upkeep. One such project is under Professor Philip Corbet of Waterloo University, Ontario, and would be funded by the Canadian Government and the U.N.D.P. grant to ICIPE. The Corbet group would work on ecology of Aedes aegypti, especially in regard to possibilities for biological control by a predatory mosquito, Toxorhynchites brevipalpis. Dr. Corbet has had many years of experience with field research on East African mosquitoes. His interest in Toxorhynchites fits closely with our interests both at MBU and at Notre Dame. Addition of

1-2 postdoctoral scientists, plus staff, from the Corbet group would markedly strengthen the work under this contract. There is also a possibility that we may be joined by a Swiss research group, under Professor Martin Luscher, working on caste determination in termites. Both the Corbet and Luscher groups would begin work in February, 1973.

3. Several administrative matters have been identified as possibly impending accomplishments.

   a. We have had a problem in housing of Research Scientists. Our laboratory is located in Nyali, directly to the north of the island of Mombasa. The most reasonable housing is located at Likoni, south of Mombasa, and this is where Drs. McDonald and Fanara live. Travel to the MBU laboratory requires a ferry boat trip plus toll bridge plus travel through the Mombasa business district. It often requires 90 minutes or more. Housing is available in Nyali, near the laboratory, but rent is much higher. Working efficiency would be greatly improved if all Research Scientists lived within a few minutes of the laboratory, especially because much of this biological research must be done evenings and weekends (mosquito cultures need frequent tending). Therefore, all Research Scientists have been required to move to Nyali and their housing allowance has been raised in the budget for year 2 from $1200 per year to $1848 per year. This is a modest amount, well below the $2400 per year given to ICIPE Research Scientists in Nairobi. Note that typical rental (not including utilities) for a couple in Likoni is $2016 and in Nyali is $3360. Thus, these payments are far from generous.

   b. The administrative load on Field Director Hausermann is much heavier than was anticipated. His time as a scientist is eroded by responsibilities for bookkeeping, personnel management, building and vehicle maintenance, correspondence, typing, photocopying, etc. No provision for administrative and secretarial help was included in the initial budget. We now propose to add a half-time Bookkeeper-Managerial Aide, with salary of $2400 per year.

5. Anticipated budget - year 2 (1 July 1972 - 30 June 1973)

Expenditure	Total 3 years	Year 2 Initially proposed	Year 2 Currently proposed*
AT NOTRE DAME			
1. Salaries and Benefits	98,960	32,980	35,520**
2. Supplies	9,405	3,000	1,000
3. Bank Charges	1,500	500	500
4. Travel	20,600	6,867	6,867
5. Capital	-	-	8,201***
6. Indirect costs-UND	12,935	4,311	4,667
	143,400	47,658	56,755
IN KENYA			
7. Salaries and Benefits	21,000	7,000	7,000
8. Housing Allowance	10,800	3,600	5,544****
9. Rental-Space	12,000	4,000	4,000
10. Supplies	7,695	2,500	3,500
11. Travel	12,000	4,000	4,000
12. Capital	28,905	11,000	9,000*****
13. Indirect costs-ICIPE	23,100	8,025	8,025
	115,500	40,125	41,069
TOTAL	258,900	87,783	97,824

* Note carry-over of $10,041 that was not spent in year 1, making a total available in year 2 of 97,824.

** Salaries and benefits of 3 scientists plus 2540 for half-time services of bookkeeper-managerial aide.

*** Purchase and shipment to Mombasa of portable irradiation source, Cesium -137.

**** Raise in housing allowance of 3 scientists to allow them to live in Nyali, close to MBU laboratory.

***** For additional vehicle plus mass-rearing equipment plus additional microscopes and environmental control apparatus.

H. APPENDICES

1. MBU Work Plan
2. MBU Staff List
3. MBU Progress Report, July - December 1971
4. Population Monitoring Program, Rabai
5. Description of the Rabai study area
6. Interim Report, Project 1, Hausermann, Population Dynamics
7. Interim Report, Project 2, Hausermann, Life Tables
8, 9, 10. Interim Reports, Projects 3 and 4, Fanara, Feral Populations
11. Interim Report, Project 5, Hausermann, Genetics
12. Interim Report, Project 6, McDonald, Chromosome Translocations
13. Mosquito Strains at MBU
14. ICIPE Organization
15. Visit Report, K. S. Rai

Tentative assignment of projects and responsibilities - Mosquito Biology Unit, ICIPE

Projects	MBU Staff	Spin offs	Consultants	Supportive research at UND
1. Description of population fluctuations in main study area - designation of 10 - 20 villages for experiments	Hausermann * + Fanara + McDonald + 2 field assistants full time + villagers	Qualitative sampling techniques Basic data for releases	Trpis Crovello Van Someren	Crovello : Storage of data + processing by computer Rai : Karyotype analysis of species collected
2. Lifetables in indoor and outdoor niches -special weight on fertility and fecundity	Fanara (larvae) Hausermann (adults) 2 assistants part time + villagers	Quantitative sampling technique Basic data for releases Get lifetable data also thru releases (e.g. sterile males, -sea horse key exp.)	Trpis Crovello	Crovello : data processing Crovello : lifetables in the laboratory
3. Environmental definition of indoor and outdoor habitats	Fanara *	Benefits to lifetable + genetic study + indoor/outdoor problem		Crovello : data processing
4. Ecological relations between indoor and outdoor niche -manipulation of population by source reduction and insecticides	Fanara * Hausermann + 2 field assistants	Release strategy (lifetables) Model of movements		Crovello : data processing

Projects	MBU Staff	Spin offs	Consultants	Supportive research at UND
5. Genetic relation between indoor and outdoor populations	Hausermann * 1 field assistant part time 1 lab. assistant part time	New mutants Release strategy	Trpis Crovello Craig McDonald Rai Kilama	Trpis : Host preference Crovello : laboratory life tables Crovello : speed of development under light regimes Kilama (Univ. Dar es Salaam) : Vectorial capacity for saurian and avian malaria and filariasis
6. Finding translocations + production of homozygotes	McDonald * + 2 lab. assistants		Crovello Rai Craig	Rai and associates
Genetic engineering of release strains for translocations and sterile males	McDonald * + 2 lab. assistants part time	Field tests on small scale thru releases	Craig Rai	Rai and associates
8. Mass rearing -qualitative control + fitness evaluation of translocations and sterile males	McDonald Hausermann	Field tests on small scale thru releases	Gerberg LaBrecque Craig Rai	Craig and associates
9. Develop release strategies -combination of releases with source reduction + insecticides for village isolation	McDonald * Hausermann Fanara		Rai Craig	Crovello : data processing comparing to status of the system before release and control villages

*=primarily responsible

MBU Staff List

June 1972

Name and position	Field work at present stage	Special assignments
Dr. Walter Hausermann Project Leader	Population dynamics, dispersal, distribution of A. aegypti in Rabai location. Sampling techniques, marking techniques. Description of pale and dark form of A. aegypti and genetic analysis.	Design and fabrication of lab. furniture, cages and collecting kits
Dr. Paul T. McDonald Cytogeneticist	Construction of translocations and genetic engineering of stocks for releases.	Design of lab. furniture and cages
Dr. Dean M. Fanara Ecologist	Manipulation of A. aegypti population by management of larval habitats. Habitat descriptions, life table studies in natural environment.	Design and fabrication of collecting kits and field laboratory
Mr. Joseph D. Arende Lab. technician for project	Rearing of field collected material. In future also mass production.	Draftsman
Miss Rosbella Chesang Lab. technician for project	Maintenance of field collected strains. Care for genetic crosses.	Care for mice and other host animals jointly with Mr. Maina
Mr. David Maina Lab. technician assigned to Dr. McDonald	Maintenance of genetic marker strains and translocation stocks. Care for genetic crosses.	Care for mice and other host animals jointly with Miss Chesang
Mr. George Tsuna Lab. technician	Maintenance of genetic marker strains, jointly with Mr. Maina	Preparation of mosquito cages
Mr. Steve Wanjohi Caretaker	Care of house and garden. Assistance in workshop.	
Mr. Julius Mwandandu Field assistant -supervisor of casual helpers	Routine monitoring of A. aegypti in Rabai location. (relative density measurements) Release experiments.	Driver Climatic measurements in Simakeni
Mr. James Rimba Field assistant	Routine monitoring of A. aegypti. (stand in for Mwandandu or Kengo)	Carpenting

MBU Staff list (contd.)

Name and position	Field of work at present stage	Special assignments
Mr. Douglas Kengo Field assistant	Routine monitoring of *A. aegypti*. Release experiments.	
Mr. Samuel Mburu Field assistant	Life table studies. Release experiments.	
Mr. Livingstone Ndenge Field assistant	Life table studies. Release experiments. Manipulation of larval habitats.	Climatic measurements in Mbaa (near Mazeras)
Mr. Robert Chai Casual helper in Shauri Moyo 2-3 days/week	Mosquito catcher for mark-release-recapture experiments.	
Mr. Stanley Karisa Casual helper in Shauri Moyo 2-3 days/week	Mosquito catcher for mark-release-recapture experiments.	Climatic observations in Shauri Moyo, 5 min./day
Various casual helpers	2 mosquito catchers for each of the four sublocations Simakeni, Ganga, Rabai Mwele, each pair working one morning per week under supervision of J. Mwandandu. Landing biting catches and handnet catches for routine monitoring of *Stegomyia* in Rabai location.	

ANNEX 3

The Mosquito Biology Unit (MBU) of the International Centre of Insect Physiology and Ecology (ICIPE)

Progress Report No. 1 July - December 1971

Research Staff in Mombasa :

Dr. Walter Hausermann : Project leader
In Mombasa since July 16.

Dr. Paul T. McDonald : Geneticist - Cytologist
In Mombasa since September 15.

Dr. Dean Fanara : Ecologist
Expected on December 27.

Consultants :

Dr. Eugene J. Gerberg : Advisor on organization, equipment and mass production of mosquitoes.
On visit from October 31 to November 10.

Objectives for the first six months

1. Installation of laboratory and insectaries. Procurement of equipment.
2. Employment and training of ancillary staff.
3. Description of study area. Selection and mapping of field sites.
4. Description of local *Aedes aegypti* populations. Colonization of feral and domestic strains for genetic analysis.
5. Incorporation of translocations into genetic background of local populations.
6. Develop schedule for routine monitoring of population fluctuations by relative methods.
7. Mark-release-recapture experiments, absolute population estimates and dispersal studies.

1. Installation of laboratory and insectaries, procurement of equipment

Initially it was planned to set up a laboratory in a residential house provided by the Division of Insect Borne Diseases (DIBD) of the Kenya Ministry of Health. This building was located close to the Mombasa Airport and just at the time MBU moved in, Kenya decided to expand its Mombasa Airport. Unfortunately it was only November when the final plans for the airport extensions were worked out and it became obvious, that the prospective laboratory would be cut off from any access by road.

Since then alternative quarters have been found for MBU. A lease is at present worked out and it is expected that by January 1st MBU can move into its final quarters.

Due to the uncertain future of the DIBD house very little had been invested in fixed installations. The major efforts so far have been made in assembling the necessary lab furniture and insectary equipment.

At present MBU is sufficiently equipped to maintain 20-30 different mosquito strains on a routine basis. Simultaneously we can also cope with 50-60 larval or egg collections from the field. For genetic crosses there are also ample cages (pint and gallon size) available.

A first shelf for mass production of mosquitoes (120,000 at a time) is at present being constructed according to the instructions left by Dr. E. Gerberg. The necessary egg masses for mass production can be produced any time without interrupting the routine operations.

In expensive equipment MBU possesses at present 1 long wheel base Landrover. We will purchase a second one in the near future as soon as ICIPE obtains customs clearance. Further, MBU possesses one high powered microscope equipped for microphotography and one dissecting microscope. It is planned to purchase a second dissecting microscope for MBU soon. An additional dissecting microscope is available for use but the private property of the project leader.

2. Employment and training of ancillary staff

In view of the large number of unemployed youths with school leaving certificates it was decided to recruit our field assistants among them. Employment followed in about monthly intervals. This permitted to introduce the newcomers to their tasks with appropriate personal attendance. According to ICIPE regulations ancillary staff is employed for a probationary period of two months after which an assistant is confirmed or dismissed, according to his performance. At present four people are working for MBU under annual contracts with ICIPE. One person so far had to be dismissed and one other person is still on probation. As soon as Dr. Dean Fanara arrives it is planned to employ one more person, bringing MBU's ancillary staff to a total of six.

For field experiments as well as for the routine field observations various numbers of local helpers are employed on a daily basis. These people will serve as mosquito catchers and also act as mediators with the local population, without whose cooperation our work would be impossible. At present MBU has 6 youths working between 4-6 days per month against a daily salary. In addition two youths are contracted to record the daily rainfall and the daily maximum and minimum temperatures at their villages (=selected field sites). The number of such people and employment days will increase once the field experiments start on a larger scale.

3. Description of the study area, selection and mapping of field sites

The coastal belt of Kenya is rather uniform in its ecological potential. It runs parallel to the Indian Ocean and is some 20-30 km wide. Its western boundary is more or less defined by the 50 cm isohyet. The climate is described as subhumid to semiarid. The year starts hot and dry. The change from N.E. monsoon to S.E. monsoon brings heavy rains in April and May. June to September are the coolest months of the year. In October with the change to the N.E. monsoon the heat starts building up again. Occasional showers occur from June to December. The climax vegetation is wooded grassland with patches of forest. Adjacent to the west lies the arid Nyika or Duruma low belt with thorn bush and grassland. *Aedes aegypti* is common in the coastal belt only and rarely found in the scarce settlements of the Nyika.

The coastal belt is divided into two distinct subunits:

a. The coastal plains are characterized by vast sisal, coconut and sugar cane plantations and cattle ranges, leaving little space for subsistence farming. Numerous hotels and housing estates along the beaches, various industries and the plantations provide employment for a large part of the population.

b. The foot plateau, a gently undulating country with rivers and gullies, is rather loosely populated by subsistence farmers. It is characterized by large irregular stands of coconut, mango and cashewnut. Some hill crests and gullies are still covered with patches of original forest. Towards the coast as well as towards the arid Nyika the coconuts give way to grassland. Cattle and goats are common everywhere. The staple crop is maize. Bananas and pineapples are unexpectedly rare.

The A. aegypti populations of the Kenya Coast are rather unique among those of East Africa, in that the species is commonly found breeding indoors in containers for water storage (Teesdale, 1955; Van Someren et al., 1955).

Our surveys to various parts of the coast have established that the frequency of indoor breeding varies with the locations. So was no indoor breeding observed among the newly settled Akamba people in the Shimba Hills or the Digo people in Kwale. Only occasional indoor breeding was observed in Duruma homes at the border between the coastal belt and the Nyika. Indoor breeding, however, is extremely common among the Rabai, Giriama and Swahili in the road triangle Mombasa - Mariakani - Kilifi (Figure 1). The distribution of indoor breeding seems to be related to particular water storage habits. Easy access to fresh water does not eliminate indoor breeding, but rather enhances it, for then water is stored in several containers, each of which serves a different purpose. Consequently each container is depleted less frequently. In the arid Nyika where water is rare, one water container serves often for the inhabitants of several houses and is depleted and refilled almost daily. In the water rich Rabai area three containers per house are quite common. One is for drinking water, one for cooking and one for personal hygiene or as reserve. As a result the indoor breeding density is very high (Breteau indices of 100 or more).

That indoor breeding can easily be eliminated by a more frequent water exchange is demonstrated by the low incidence of indoor breeding around Mombasa Airport, where the people are instructed regularly by staff of the Mombasa Municipality and fined if they are found with mosquitoes breeding indoors.

The main study area so far selected lies in the coastal foot plateau along the road from Mazeras to Rabai (Figure 2) over a stretch of about 5 km. The area is quite densely populated (200-300 inhabitants per km^2). The houses are arranged in clusters of 5 to 15. Along the road these clusters are dense, 100-200 m apart; farther from the road they are scattered more widely, 500 m and more apart. Even more isolated clusters of houses are found along the Mariakani - Kaloleni road to the west.

The arrangement of houses into clusters seems to be ideal for all sorts of ecological and genetical studies. Depending on the requirements particular

experiments can be carried out across several adjacent clusters or in a single more isolated one.

Similar conditions can also be found in the coastal plain and foot plateau north of Mombasa. So far, however, no particular sites have been investigated there.

Different conditions are found closer to Mombasa in the area around the airport. There the only breeding occurs outdoors; indoor breeding being prevented by regular source reduction.

4. Description of local A. aegypti populations. Colonization of domestic and feral strains.

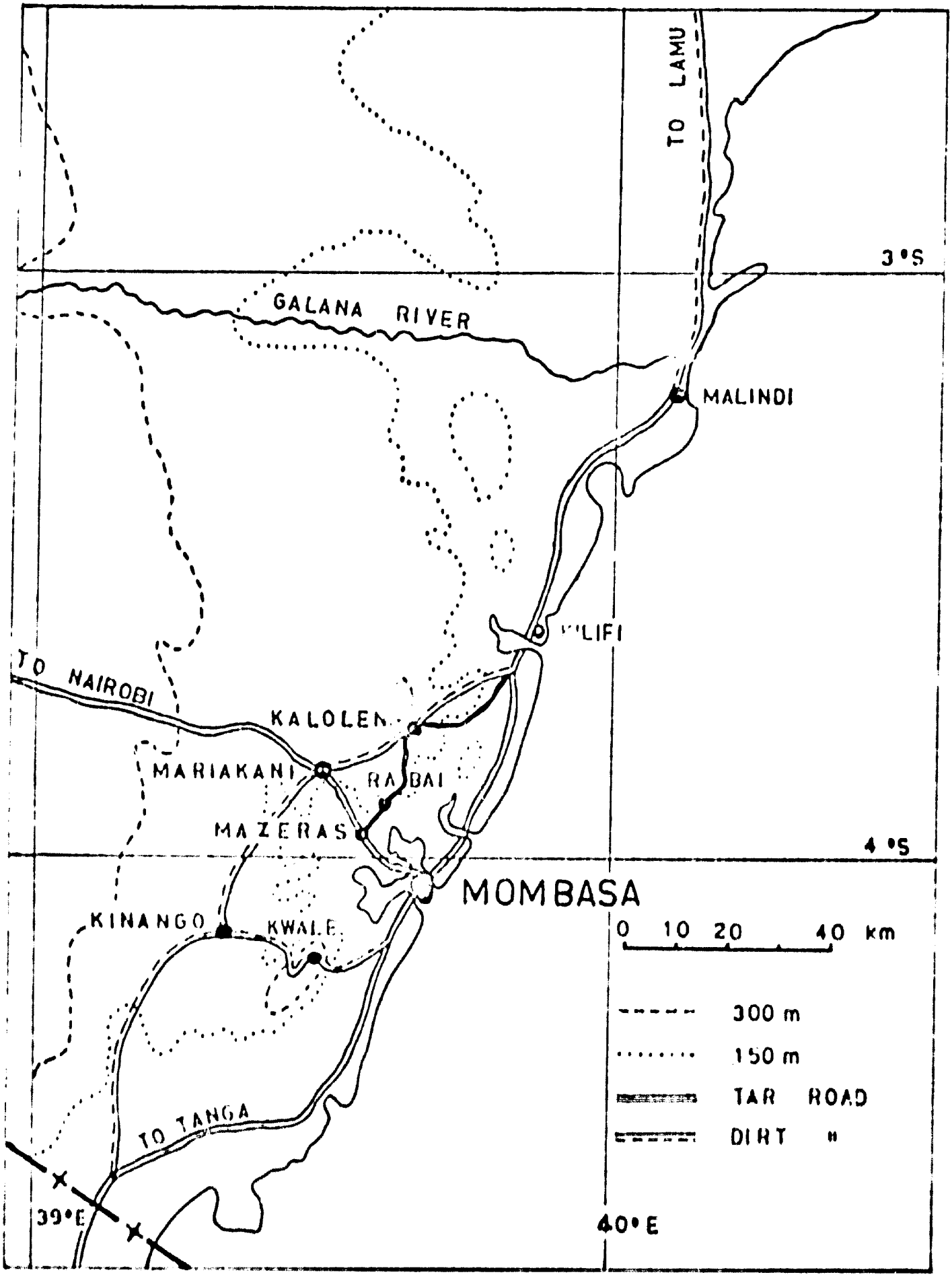
The A. aegypti of the Kenya Coast display a great deal of colour variation (Van Someren et al., 1955). Two forms are distinguished, "dark" and "pale", which differ also in their behaviour. The dark forms occur essentially outdoors while the pale forms are breeding and biting indoors. Despite their different haunts, however, the populations are not completely distinct. Even the two colour extremes can mate with each other and are fully interfertile. Moreover a whole range of intermediate colour forms can be obtained by appropriate selection and crossing of strains collected indoors (McClelland, 1960). In the present investigation several of McClelland's intermediates have been found at varying frequencies among larval samples from indoor as well as outdoor habitats.

The extreme dark form, A. aegypti ssp. formosus (Mattingly, 1957), recognized by its first abdominal tergite being entirely black, has so far been collected from outdoor habitats only. These habitats included steps in coconut trees - probably the most common habitat for Aedes on the Kenya Coast - treeholes and seed pods, but also tires, tins and earthenware jars. The latter are occasionally installed at the foot of coconut palms to collect rainwater. Outdoor larvae may thus be transferred to indoor water containers after each rain.

Associated with the larvae of A. aegypti ssp. formosus were usually specimens of the type form, predominantly dark, but occasionally displaying paleness on thorax and abdomen, indicating the presence of the mutant alleles gold and spot.

In larval samples from indoors, pale specimens displaying the various alleles of spot abdomen and pale abdomen were dominant and included specimens of a paleness which was never encountered in outdoor habitats. Type form specimens including some rather dark ones, however, were obtained as well.

During the cool dry season of the last months up to November, outdoor breeding was rather rare. The sporadic showers of the period did as a rule not permit the larvae to develop but were detrimental to the quiescent eggs of large numbers of larval habitats by inducing hatching. Miraculously these habitats received again eggs after drying out, i.e. outdoor oviposition was still going on, although no outdoor biting was recorded. Indoor breeding on the other hand continued all the time, unaffected by the occasional but limited catastrophes caused by cleaning housewives alerted after our visits.


TO LAMU
3°S
GALANA RIVER
MALINDI
VILIFI
TO NAIROBI
KALOLENI
MARIAKANI
RABAI
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MOMBASA
KINANGO
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150 m
TAR ROAD
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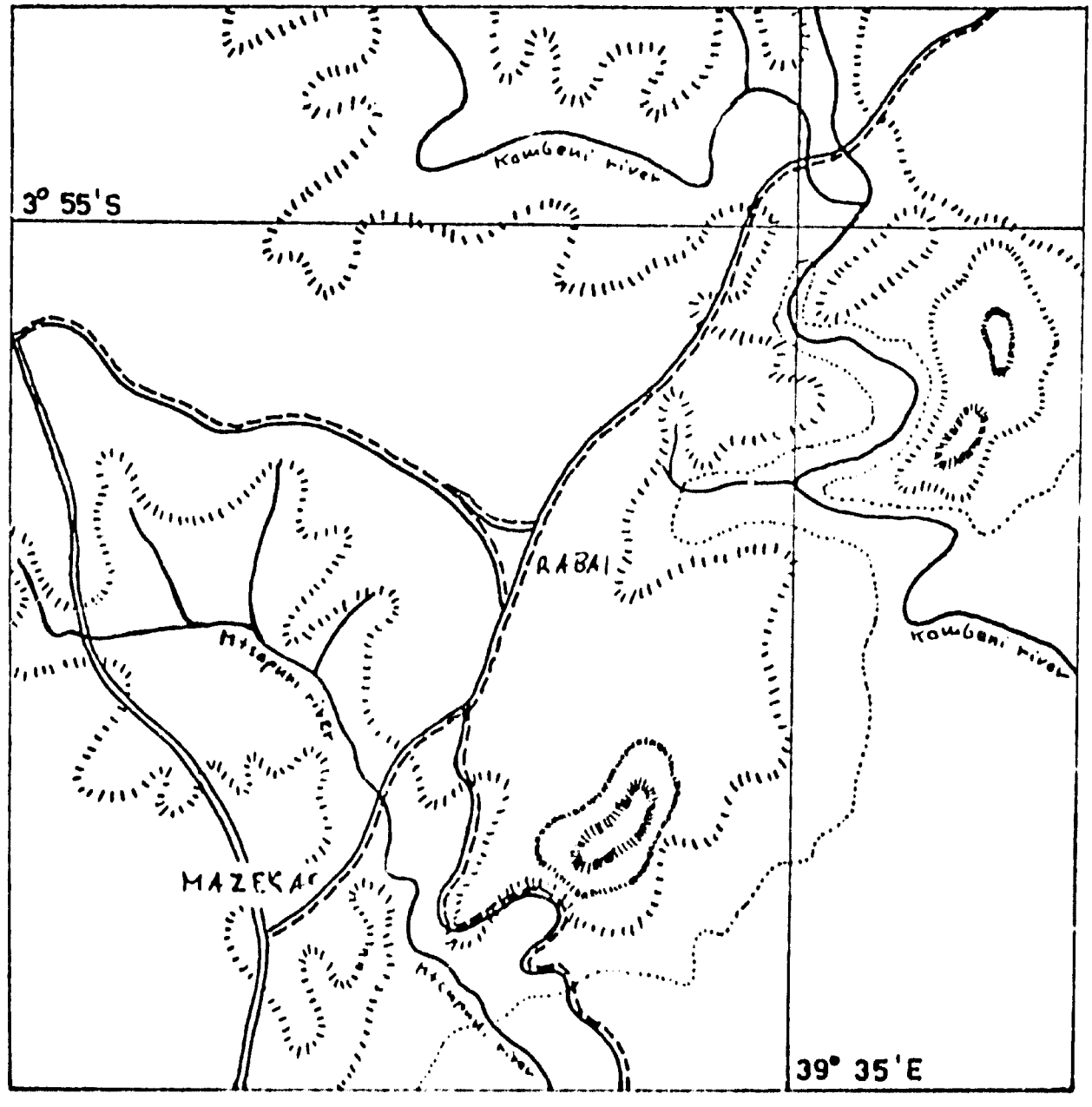
TO TANGA
39°E
40°E

Location Rabai - Main Study Area



For experiments in genetic control a population breeding entirely indoors would be ideal, i.e. isolated and extremely vulnerable. Unfortunately the real indoor breeding populations seem to have still considerable genetic exchange with outdoor breeding populations despite their distinguished paleness. Although it is conceivable that the extremely pale components of the indoor populations are confined entirely to indoor breeding and that the dark A. aegypti ssp. formosus does not enter houses voluntarily, it is very likely that some intermediate forms (the majority?) exploit both ecological niches to various extents and channel the gene flow between the populations.

For the introduction of genetic mechanisms it seems therefore important to know the selective pressures and behavioural traits governing the separation of pale and dark. Genetic analysis of field collected strains and release experiments with genetically and paint marked specimens will thus be the next steps. Various indoor and outdoor strains from different villages have been colonized and are available for genetic analysis and the introduction of genetic mechanisms.

5. Incorporation of translocations into genetic background of local populations

Newly established strains of MKWAJA (collected in outside waterdrums in Tanzania) and SHAURI MOYO (collected indoors in water containers in field study area) provided young adult males for irradiation. The males were irradiated to 2500 rads or 3500 rads and progeny from these were tested for the presence of translocations with a RED (re/re ; s/s ; blt/blt) multiple chromosome marker strain. Six translocations were selected for maintenance against the background of the RED marker strain. All were produced from 2500 rad treatment.

Lines of five translocations are being maintained and crosses have been made to produce homozygotes where possible. The results to date indicate that one of the lines, AT(1:3)c, has produced viable homozygotes. (Table 1)

Young males of the MAZERAS strain (collected indoors in water containers in field study area) have been irradiated with 2000 rads, 2500 rads and 3500 rads and their progeny will be screened for the presence of translocations against a multiple chromosome marker stock, ru/ru ; Si s/Si s ; blt/blt. The emphasis is to be placed on selecting large numbers of easily maintained translocation heterozygotes with subsequent continuous searches for homozygotes.

Markers are currently introduced into a MAZERAS strain to produce an essentially African strain with suitable markers for selecting and maintaining translocations. In the F_2 generation are re/re ; s/s ; blt/blt and wh/wh ; Si s/Si s ; blt/blt. The stocks of wh/wh ; Si s/Si s ; min/min and ru/ru ; Si s/Si s ; min/min are being enlarged in attempts to make them suitable for marker strains as well.

6. Routine monitoring of population fluctuations by relative methods

For field experiments planned in the course of this investigation it seemed important to have sufficient information on the population fluctuations of the natural populations prior to any interference.

Table 1 : Translocations available

Translocation	Origin	Crossover units for pseudolinkage	Test for homozygosity
AT (1:2) a	MKWAJA	$M - s^+$ (4.5)	in progress
AT (1:2) b	MKWAJA	$re^+ - s^+$ (2.1)	none in 5th generation
AT (1:3) a	SHAURI MOYO	$M - blt^+$ (9.8)	not tested
AT (1:3) b	SHAURI MOYO	$re^+ - blt^+$ (0.7)	none in 5th generation
AT (1:3) c	MKWAJA	$re^+ - blt^+$ (3.1)	homozygotes in 4th generation

It was therefore decided to monitor A. aegypti for the study area on a routine schedule. The methods applied are classic:

1. Relative density of biting and landing adults
2. Frequency of breeding
3. Frequency of oviposition

The procedure, somewhat complicated by the presence of two more or less separate populations, includes the following monthly censuses:

Indoor populations

1. House index and Breteau index for 100 houses from at least 25 different clusters of houses between Kombeni river and Mazeras.

2. Overall indoor landing biting rate from some 25 houses with indoor breeding in at least 10 different clusters.

   Each house visited by 3-4 field scouts for one hour

Outdoor populations

1. Landing biting rates measured on ideally four different sites of the study area in 13 h catches (6 a.m. - 7 p.m.).

   These outdoor catches are not yet fully established because there is practically no outdoor biting at present. It is planned to limit these outdoor catches later on to the peak biting hours of the day.

2. Frequency of breeding

If available 50 larval habitats of any kind containing water are sampled for mosquito larvae once per month. From each habitat up to 20 larvae or, if less, all larvae are collected and raised to adults for identification. The number of habitats positive for A. aegypti and the ratio of A. aegypti to other Aedes are then determined.

3. Frequency of oviposition

The frequency of oviposition is determined by ovitraps set for 2 days in 5 different areas. However, December is the first month that this has been carried out and the results are not yet available. So far each month 50 larval habitats have been sampled for Aedes eggs by scratching out the dirt and flooding it. Besides A. (Stegomyia) aegypti this sampling produced also larvae of A. (S.) simpsoni, A. (S.) metallicus, A. (S.) heischi, A. (S.) vittatus, A. (S.) calceatus, A. (Aedimorphus) haworthi, A. (Diceromyia) adersi and A. (Finlaya) fulgens.

The results of the routine monitoring are presented in Table 2.

7. Mark-release-recapture experiments, absolute population estimates and dispersal studies

No mark-release-recapture experiments have been made so far.

References

McClelland, G.A.H. (1960), A preliminary study of the genetics of abdominal colour variations in Aedes aegypti (L.)(Diptera - Culicidae). Ann. Trop. Med. Parasit., 54:304-320.

Mattingly, P.F. (1957), Genetic aspects of the Aedes aegypti problem. I. Taxonomy and bionomics. Ann. Trop. Med. Parasit., 51:392-408.

Teesdale, C. (1955), Studies on the bionomics of Aedes aegypti (L.) in its natural habitats in a coastal region of Kenya. Bull. Ent. Res., 46:711-743.

Van Someren, E.C.C., Teesdale, C. and Furlong, M. (1955), The mosquitoes of the Kenya coast; records of occurrence, behaviour and habitat. Bull. Ent. Res., 46:463-493.

Table 2 : Results of routine monitoring

	July	August	September	October	November	December
Climate :						
Mean max. temperature	26.9	26.7	28.2	29.3	31.9	/
Mean min. temperature	19.5	18.7	18.6	21.0	21.4	/
Rainfall in mm	62.7	12.9	25.9	7.6	21.6	/
Indoor populations :						
House index *	63	64	78	77	76	93
Breteau index **	71	89	117	112	117	144
Landing ♀♀ per manhour	/	/	1.41	1.25	1.53	1.41
Outdoor populations :						
Breeding (% pos. habitats)						
Larvae	/	51	/	/	/	46
Quiescent eggs	/	/	16	42	26	/
% *A. aegypti* of all *Aedes*	/	59	32	85	56	52
Landing ♀♀ per manhour	/	/	/	/	0.01	0.01

* House index - No. houses with indoor breeding among 100 houses surveyed
** Breteau index - No. indoor water containers with larvae per 100 houses surveyed

Routine monitoring of A. aegypti in Rabai location

Purpose : Description of fluctuations of indoor and outdoor A. aegypti populations in relation to environmental factors like rain, temperature and humidity and in relation to other mosquitoes.

Methods : Relative density measurements based on adults, pupae, larvae and oviposition.

Area under study: The area under study is depicted on map 1. It is divided into four sublocations for sampling. The sublocations are called Simakeni, Ganga, Rabai and Mwele.

Each sublocation contains ten sampling sites, houses for indoor A. aegypti and sampling routes for outdoor A. aegypti. At each sublocation the sampling sites are designated individually considering the spread and size of clusters of houses and the topographical structure as well as the vegetation cover.

Procedures: 1. Sampling adult mosquitoes

1.1. Indoor captures of adults are made as landing-biting-catches in 10 houses per sublocation. The houses in each sublocation are numbered from 1 to 10. Two men start their indoor capture round at 7 30 h and terminate it at 11 30 h. They spend 20 minutes in each house and have 4 minutes to get from one house to the next. The captures are made in single tubes and the tubes put into a bag marked with the number of the respective house.

1.2. Outdoor captures are made with the handnet along a route crisscrossing through the sublocation and visiting the four sites with the densest vegetation cover and the heaviest shade. Two men start the outdoor capture round at 7 30 h and terminate it at 11 30 h. The mosquitoes caught within each hour of the morning are put into separate cups labelled 1 to 4.

2. Sampling larvae and pupae

2.1. Indoor sampling is carried out in the same ten houses per sublocation as the indoor landing-biting catch. So far A. aegypti has been the only mosquito found to breed regularly in the indoor water pots. No attempts are therefore made to verify this for each container found positive with larvae in the course of this routine monitoring.

After the landing-biting-catch of the morning one man visits all ten houses of the sublocation again in the afternoon and records the number of water pots with water and the number of pots with larvae per house. He then proceeds to catch with a handnet and count all 4th instar larvae and pupae in one designated container of each house. The figures are then recorded and the larvae and pupae replaced again. This afternoon inspection is carried out by one man and takes about 3 hours.

2.2. Outdoor sampling is made simultaneously with the indoor sampling by a second man. He visits 20 designated and numbered outdoor larval habitats. The habitats were selected with consideration of site and kind. The following kinds of habitats are distinguished: CT = coconut step, TH = tree hole, TR = tire, DC = habitat of domestic origin, like pot, pan, tin etc. No short l sting habitats, i.e. habitats which are easily removed, disturbed or destroyed or habitats of small capacity were designated for routine inspection. (Possibly during rainy season a number of additional habitats of this kind may be added to the routine inspection).

The field scout inspecting the outdoor habitats numbered from 1 to 20 records for each habitat, whether it contains water and larvae. If it is positive for larvae, he tries to count the number of pupae present. Samples of up to 20 pupae or larvae are then collected from up to 8 positive larval habitats to establish the species composition in the laboratory.

3. Oviposition

Oviposition is sampled by means of ovitraps of the "Fay" design. After traps placed inside houses failed for three months to yield eggs, it was decided to put these traps only outdoors. Twenty traps are set in each of the four sublocations in preselected and marked sites: 10 under peridomiciliary and 10 under extradomiciliary conditions. The traps are left in the field for 48 hours and then collected. The oviposition strips with eggs are conditioned and the eggs hatched and reared in the laboratory one week after collection.

Equipment: Transport: 2 bicycles, 2 baskets

Recording: 2 clipboards, 2 ballpens, recording forms, individual notebooks

Sampling of adult mosquitoes: 2 handnets, 2 aspirators, 40 clothbags, 16 single-pair cages, about 200 glasstubes + cotton wool, 2 flashlights

Sampling of larvae: 2 pipettes, 2 handnets for larvae, 2 plastic basins, 4 plastic cups, 32 styrene tubes for larval collections, 2 flashlights

Oviposition traps: 80 oviposition traps, 1 water container a 10 lt, oviposition strips, 2 plastic food containers, paper towels

Staff : Permanent staff: Mr. Julius Mwandandu
Mr. Douglas Kengo

Casual helpers : 2 in each sublocation, selected by Mr. Mwandandu and working under the supervision of Mr. Mwandandu and Mr. Kengo in the morning

Stand-in for permanent staff: Mr. James Rimba

Rearing and Identification of field collected material in the laboratory Mr. J. Arende under supervision of W. Hausermann.

Time Schedule

Day	Location	Time	Activity	Staff involved
Monday	all four sublocations	800 - 1200	collection of ovitraps	J. Mwandandu D. Kongo
		1330 - 1630	special assignments	J. Mwandandu D. Kongo
Tuesday	Simakoni	730 - 1130	indoor captures landing-biting-search of adults	J. Mwandandu + 1 man
			outdoor captures handnet	D. Kongo + 1 man
		1300 - 1600	indoor sampling of larvae and pupae	D. Kongo
			outdoor sampling of larvae and pupae	J. Mwandandu
Wednesday	Canga	730 - 1130	indoor captures	D. Kongo + 1 man
			outdoor captures	J. Mwandandu + 1 man
		1300 - 1600	indoor sampling	J. Mwandandu
			outdoor sampling	D. Kongo
Thursday	Mabai	.....	AS TUESDAY	.....
Friday	Mvolo	.....	AS WEDNESDAY	.....
Saturday	all four sublocations	800 - 1200	setting of ovitraps	J. Mwandandu D. Kongo

Description of the Location Rabai

The Location Rabai is an administrative unti of the Kilifi district headed by a chief. It extends along the Mazeras-Kaloleni road and is bordered by steep slopes in SE and the Mariakani-Kaloleni road in the NW. The NE border is the Kombeni river and the SW border the Mombasa-Nairobi road. The area of approximately 35 km^2 is inhabited by about 5000 persons, half of them children under 15 years. Most of the people are subsistence farmers with coconuts as an additional cash crop.

Buildings are mostly rectangular with mudwalls and thatched roofs. Often the mudwalls are plastered with cement. A second type of house, called traditional, is made entirely of thatchwork, either with palm leaves or grass. The houses are usually assembed in clusters of 5-15 and belong to people of related families. Often there is piped water to such a village with tap or two, from which everybody collects the water for storage in the house. The distance between clusters of houses varies. Along the road to Kaloleni these clusters are close, farther from the road the villages are more scattered.

The vegetation cover of the area reflects the transition of the semiarid coastal belt to the arid Nyika or thornbush-grassland. The steep slopes and hills to the coast are partially covered with primary forest. The gently undulating country to the west of the coastal slope is covered with grassland and native palms as well as planted coconut palms. The palm groves are thinning out to the west and are finally limited to a few depressions, which hold the water. The palm groves with mango trees are densest towards the forest area near Buni (see map).

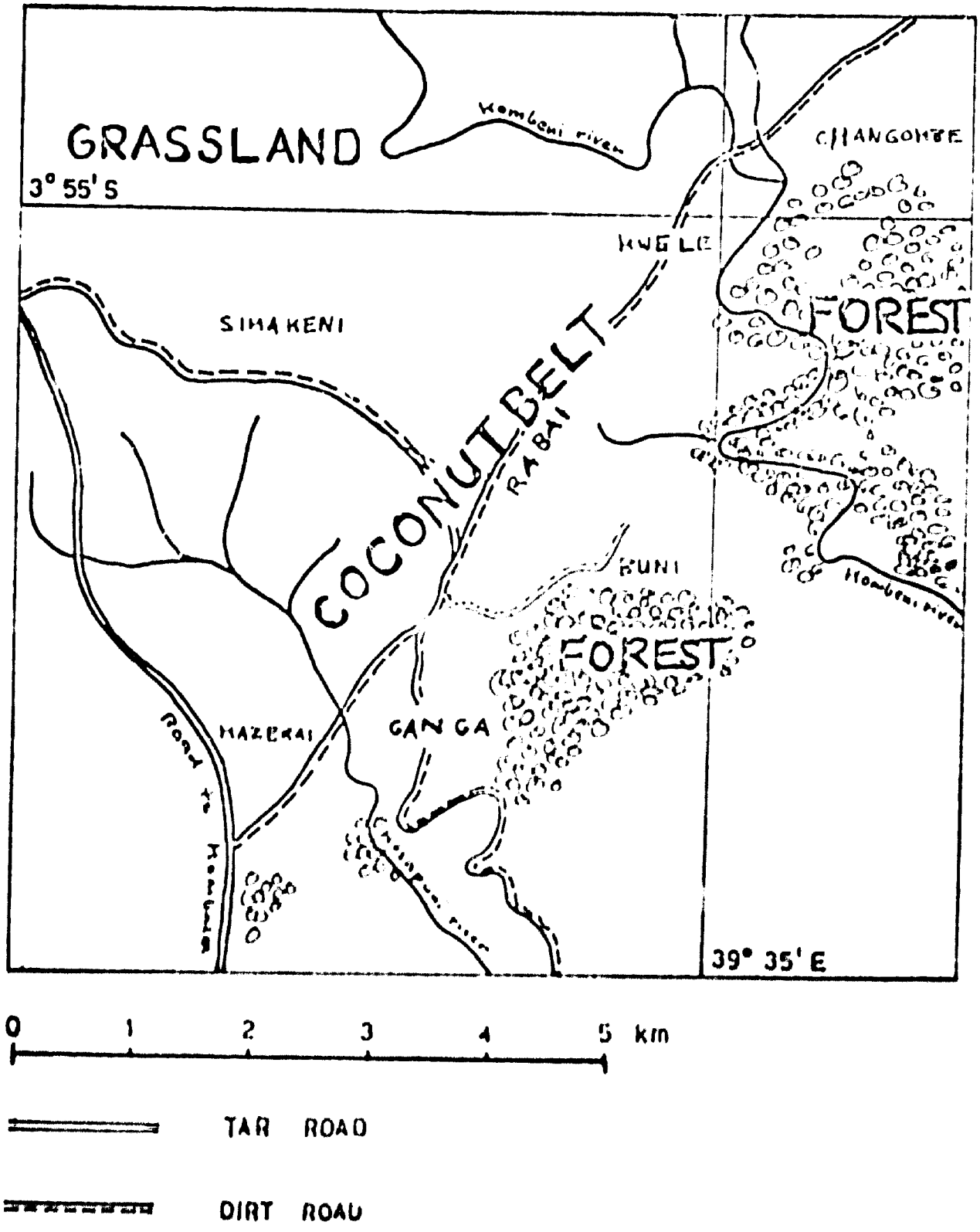
The understorey vegetation in the coconut groves in grass, often replaced by the weed Lantana due to overgrazing by cattle and goats. During the short rains from November to early January and in the long rains from May to September the peasants plant the staple crop maize. Cassava is also quite common. Rice is planted only exceptionally during the long rains. Banana, pineapple and pawpaw are rare. Bananas are almost entirely limited to the narrow gullies on the steeper slopes towards the coast.

Whether the scarcity of banana and pineapple - and with it a scarcity of larval habitats for Aedes (Stegomyia) simpsoni - reflects a drier climated compare to places, where these plants are common, or simply a lack of interest of the peasants is not clear. The question will be discussed with members of the WHO Aedes Research Unit in Dar es Salaam, where banana and pineapple are a common habitat for A. simpsoni under similar climatic conditions.

The density of the human population with about 140 persons per km^2 is quite high. Th s almost the whole vegetation cover and fauna of the location are under intensive influence of man. However, two areas of primary forest are still left as Forest Reserves at the SE border in the coastal foothills, one on Benygundo hill called Buni or Kisimani Forest and one along Kombeni river and on the hills to the NE. Primary grassland with thornbush (Acacia spec.) and palms of the genera Borassus and Hyphaene is found to the western border of the location.

To describe the habitat relationships of mosquitoes the terms domiciliary, peridomiciliary and extradomiciliary are used. Domiciliary is a mosquito which breeds and bites indoors. Peridomiciliary are the mosquitoes breeding and biting in areas, where the vegetation cover has been altered through cultivation or

grazing. Extradomiciliary are mosquitoes breeding in the few areas, where the vegetation cover has not been altered to a great extent by man.


GRASSLAND
Kombeni river
3° 55' S
CHANGOMBE
KWELE
SIMAKENI
FOREST
COCONUT BELT
RABAI
BUNI
Kombeni river
FOREST
MAZERAI
GANGA
Road to Kilifi
Chamgeni river
39° 35' E
0 1 2 3 4 5 km
TAR ROAD
DIRT ROAD

Interim Report to Project 1

Title: Routine monitoring of A. aegypti in Rabai location

Responsibility: W. Hausermann

Purpose and methods applied

The purpose of the project and the methods applied are outlined in a brief manual handed out to MBU staff involved. A copy is attached (ANNEX 4 of ANNUAL RESEARCH REPORT).

Course of project and general comments

The project was started in July 1971 with one man sampling larval habitats for indoor and outdoor populations. From August onwards landing-biting catches were added and in December ovitrap collections were introduced. Until then the project was a two men one week per month affair. All observations were of a qualitative nature rather than quantitative and various sampling methods were in trial (Progress Report No. 1, ANNEX 3).

In January the project was expanded, for it was thought that quantitative data on lifetable parameters could be gathered simultaneously. By the end of January the project was established at its present level and by April the personnel involved was reasonably proficient in its tasks.

The onset of the rainy season in May brought substantial increase in the work load of the field staff and also of the laboratory, where the field collected samples had to be handled. Unfortunately it also became obvious that the field staff could not maintain the level of efficiency and accuracy required. On the laboratory end the amount of work increased correspondingly such that it interfered with other projects. I decided to reduce the project to the level it was originally meant to be.

All routine sampling in this project was carried out in domiciliary and peridomiciliary conditions only. No censuses were taken in the forests or the grassland to the west (extradomiciliary conditions).

Results

The information collected on fluctuations of A. aegypti in domiciliary and peridomiciliary conditions are presented in table 1 a + b and figures 1 and 2.

Domiciliary populations:

According to container and Breteau index the overall population density of the domiciliary A. aegypti seems fairly stable throughout the year. On the other hand week by week observations in single houses demonstrated that these A. aegypti subpopulations fluctuate widely, mainly due to interference with the larval habitats by man. The landing-biting catches show some fluctuations from week to week. These fluctuations can again be partially attributed to human activities. For instance all women of a village may be cooking in their

houses all day during a celebration and thus alter the normal landing-biting rate through smoke. Other fluctuations, however, reflect changes in the natality rate. A sudden increase in the percentage of males in the landing-biting catch from one week to another, e.g. weeks 5 - 6 and 15 - 16 (figure 1), seems to indicate the emergence of males. Whether or how these rapid and irregular fluctuations relate to climatic factors is not understood.

Peridomiciliary populations:

Contrary to the domiciliary population the peridomiciliary population shows a clear cut dependance on the rainfall pattern. After rainfalls in late December and early January, high level of oviposition activity was noted in late January and February. The absence of rains brought a decrease in oviposition activity for March and April. A small rise of oviposition in weeks 13 and 14 of April is clearly the result of a rain in late March (week 12). After heavy rains in early May (weeks 18 and 19) a rise in oviposition has taken place after week 21. The evaluation of these ovitrap collections is still in progress. The exact percentage of ovitraps positive for A. aegypti is not available yet but it will be close to or above 50%.

The observations on outdoor larval habitats (table 1) correspond with the ovitrap collections. However, the sampling of larval habitats is more laborious and the results are less clear cut, because the number of water filled habitats is changing constantly. As an addition to the results from ovitraps it must, however, be noted that the peridomiciliary habitats dry very quickly and that a very small percentage of larvae makes it through to the pupal stage, if the habitats are not refilled by rains in intervals of less than 3 to 4 days. At one time, after a rainfall, larval development had started in 30 habitats under observation. An estimated 1500 - 2000 larvae were developing, but only 2 pupae were produced. The tire habitat is the habitat lasting longest into dry spells. However, their capacity to carry larvae is reduced after some time without rain and Culex spec., mainly Culex pipiens fatigans become the most common species.

The experiences made by sampling the adult populations are shown in table 2. Day long landing-biting catches on place and walking slowly through the vegetation yielded practically no mosquitoes during long dry spells and a very low percentage of Stegomyia during the recent rains. In February we switched over to hand net catches, walking through the vegetation, shaking grasses and bushes with a stick and catching the mosquitoes flying off with a handnet. Although these catches yielded mosquitoes more reliably the percentage of Stegomyia and even more of A. aegypti was very low. Nicely, however, the handnet catches demonstrated an increase of the overall mosquito populations after the offset of the severe dry spell lasting from February to April.

A breakdown into species and genera collected during this project is presented in table 2.

Conclusions

The presented findings on seasonal fluctuations of A. aegypti are not quite consistent with the results of Teesdale (1955) working near Mombasa. He noted a clear decrease in population density for larvae and adults indoors during the dry months from January to April. The difference to our observation

of an almost stable indoor population may be explained by the fact that Teesdale saw no need to distinguish between a pale indoor and a dark outdoor form. It was only towards the end of his investigations that these colour differences and associated differences in habitat preference were noted (Van Someren et al. 1955). Also Teesdale worked in an area closer to the coast than Rabai. This area is more urbanized and in surveys we have found that A. aegypti of the type form breeds also in water drums and small rain water cysterns in the backyards of houses. Thus care has to be taken not to generalize from the conditions prevailing in Rabai for the whole of the Kenya coast. Outdoor breeding type form populations exist along the Tanzania coast and may well exist along the Kenya coast as well. Population fluctuations of the type described by Teesdale (1955) have recently been described by Trpis (1971) for the Buguruni population in Dar es Salaam. And there as described by Teesdale (1955) A. aegypti makes use of indoor larval habitats without being totally adapted to indoor life as the populations in the Rabai area.

That an A. aegypti population totally adapted to man's permanent water containers is almost stable throughout the year has also been described recently by Sheppard et al. (1969).

The sampling methods used for indoor populations, counting containers positive for breeding and landing-biting catches gave satisfactory results and shall be continued for another 7 months. Measuring the pupal output in selected houses on the other hand gave unsatisfactory results and will not be continued within the frame of project 1. Special observation series of this kind, however, will be carried out under close supervision within project 2.

The sampling methods for outdoor populations gave mixed results. Oviposition traps, a modification of the CDC trap (Fay, 1968) made of a tin can painted black, proved to be a very sensitive and precise tool for oviposition sampling. The method is simple and its use will continue until January 1973. Observation and sampling of natural habitats is less accurate than the use of oviposition traps. To work out the species composition of a large number of positive habitats is extremely laborious and subject to bias and will not be continued. Counting the pupal output of selected larval habitats proved to be too difficult for the field assistants and will also be discontinued.

Handnet catches yielded very low numbers of A. aegypti and Stegomyia in general. Based on the results of Teesdale (1958), however, the results are not that unexpected, especially if the time spent on catching is considered. Handnet catches outdoors will continue on a month to month basis dependant on the efforts to be spent on other projects.

References

Fay, R.W. & Eliason, D.A., 1966. A preferred oviposition site as a surveillance method for Aedes aegypti.
Mosq. News, 26: 531 - 535.

Sheppared, P.M., Macdonald, W.W., Tonn, R.J. & Grab, B., 1969. The dynamics of an adult population of Aedes aegypti in relation to Dengue haemorrhagic fever in Bangkok.
J. Anim. Ecol., 38: 661 - 702

Teesdale, C., 1955. Studies on the bionomics of Aedes aegypti (L.) in its natural habitats in a coastal region of Kenya.
Bull. ent. Res., 46: 711 - 743.

----- 1959. Observations on the mosquito fauna of Mombasa.
Bull. ent. Res., 50: 191 - 208.

Trpis, M., 1971. Seasonal variation in adult populations of Aedes aegypti in the Dar es Salaam area, Tanzania.
WHO/VBC/71.291.

Van Someren, E.C.C., Teesdale, C. & Furlong, M., 1955. The mosquitos of the Kenya coast; records of occurrence, behaviour and habitat.
Bull. ent. Res., 46: 469 - 493.

Table 1 a : Summary of the results of routine monitoring of *A. gambiae* in 4 sublocations of Rabai.

Month	January				February				March	
Week No.	1	2	3	4	5	6	7	8	9	10
Climate										
Mean max. temp.	29.7	31.2	31.9	32.2	32.5	31.6	32.7	32.2	32.7	32.6
Mean min. temp.	21.9	21.9	22.5	21.9	23.3	23.3	21.6	21.6	21.8	22.6
Rainfall	87.8	12.0	0.0	0.0	3.0	8.3	0.0	0.0	0.0	0.0
Indoor population										
Larvae :										
Protozoa index	143	—	—	—	140	131	145	150	158	140
Container index	87	—	—	—	98	90	98	88	92	92
Adults :										
Landing-biting-rate per man hour										
♀♀	1.2	—	—	—	2.1	2.8	1.6	1.9	2.7	2.0
♂♂	0.6	—	—	—	1.8	2.8	1.8	0.8	1.3	1.3
Outdoor population										
Larvae :										
% habitats with larvae	98	—	—	—	10	25	20	5	2	5
% *A. gambiae* in larval samples	64	—	—	—	38	82	18	0	0	73
No. *A. gambiae*	—	—	—	—	17	22	13	0	0	8
Adults :										
Handnet catch/ man hour (all species) ♀♀	—	—	—	—	0	1.0	0.3	0.3	0.4	0.6
♂♂	—	—	—	—	0	0.7	0.6	0.3	0.9	0.5
No. *A. gambiae*	—	—	—	—	0	0	0	0	0	0
Oviposition :										
% traps positive	—	—	—	40	28	32	22	12	12	5
% *A. gambiae*	—	—	—	54	43	64	41	100	35	50
No. *A. gambiae*	—	—	—	273	89	223	24	61	23	20

Table 1 b : Summary of the results of routine monitoring of *A. gambiae* in 4 sublocations of Kabai.

Month	March				April				May	
Week No.	11	12	13	14	15	16	17	18	19	20
Climate										
Mean max. temp.	33.6	31.9	32.2	32.1	32.3	32.1	31.7	30.4	27.1	27.1
Mean min. temp.	22.4	21.9	23.6	23.1	23.5	22.3	22.8	22.6	21.9	21.9
Rainfall	17.5	11.5	0.0	9.7	0.7	8.0	5.6	17.1	218.8	53.0
Indoor population										
Larvae :										
Breteau index	112	140	132	110	133	130	128	142	131	113
Container index	82	92	85	90	89	92	85	79	83	83
Adults :										
Landing-biting-rate per man hour										
♀♀	2.0	2.0	1.6	2.8	2.0	2.3	1.9	2.7	0.8	2.4
♂♂	0.8	1.2	1.2	1.2	0.8	2.9	1.2	1.6	0.8	1.3
Outdoor population										
Larvae :										
% habitats with larvae	0	10	5	0	18	20	5	14	26	40
% *A. gambiae* in larval samples	0	—	—	0	—	78	—	86	—	41
No. *A. gambiae*	0	—	—	0	—	94	—	61	—	23
Adults :										
Handnet catch/ man hour (all species) ♀♀	0	0.7	1.2	2.8	4.7	1.9	3.9	3.8	1.6	4.6
♂♂	0	0.2	0.3	0.5	0.3	1.2	0.5	0.8	0.7	2.0
No. *A. gambiae*	0	0	0(5)*	0(1)*	0	4	2(2)*	0	0	0
Oviposition :										
% traps positive	0	2	8	12	2	5	5	4	5	9
% *A. gambiae*	0	100	74	49	88	63	60	72	94	69
No. *A. gambiae*	0	26	53	168	29	86	46	50	34	104

* other *Stomoxyin*

Table 2 : List of species and genera encountered in different sampling techniques.

Method of sampling	Species collected	Frequency of appearance	Time of peak
Ovitraps outdoors	Aedes (Stegomyia) aegypti	++++	after substantial rain
	A. (S.) simpsoni	++	"
	A. (S.) metallicus	+++	"
	A. (S.) calceatus	+	"
	A. (S.) hoischi	+	"
	A. (S.) vittatus	++	"
Larval collections from outdoor habitats			
a) Treeholes, Coconut steps	A. (S.) aegypti	+++	"
	A. (S.) simpsoni	++	"
	A. (S.) metallicus	++	"
	A. (S.) calceatus	+++	"
	A. (S.) hoischi	+++	"
	A. (S.) vittatus	++	"
	A. (Finlaya) fulgens	+	"
	A. (Aedimorphus) [illegible]	(+)	"
	A. (A.) haworthi	(+)	"
	A. (Diceromyia) aderni	+	"
	A. (Danius) michaelikati	once	"
	Toxorhynchites brevipalpis (predatory)	+	"
	Culex (Lutzia) tigripes	+	"
	C. (Culex) pipiens fatigans	++	"
	Culex several species	++	"
b) tires, discarded domestic containers	Same species as above, except :		
	A. (S.) hoischi		
	A. (Aedimorphus) spec.		
	A. (D.) michaelikati		

Method of sampling	Species collected	Frequency of appearance	Time of peak
Mosquito catch outdoors	All Stegomyia listed above	(+)	2-4 weeks after substantial rains
	A. (S.) vittatus	+	1 – 2 weeks after light rains
	A. (E.) fulgens	(+)	
	A. (A.) hispanicus	(+)	"
	A. (A.) ngoye	(+)	"
	A. (A.) argenteopunctatus	(+)	"
	A. (Kerteszia) albotaeniatus	+++	at peak of dry season, reduced in numbers during and after rains
	Eretmapodites two species	(+)	2-4 weeks after substantial rains
	Culex several species	+++	1-5 weeks after rains
	Culex (L.) tigripes	++	2 – 4 weeks after substantial rains
	Mansonia uniformis + africanus	++	4-8 weeks after rains
	Anopheles several species	(+)	"
Human-biting catch outdoors	A. (S.) aegypti	(+)	2-4 weeks after rains
	A. (S.) simpsoni	(+)	"
	A. (S.) metallicus	+	"
	A. (S.) calceatus	(+)	"
	A. (S.) vittatus	+	"
	A. (E.) fulgens	+	"
	A. (E.) albothorax	(+)	dry season and during rains
	C. (L.) tigripes	++	2-4 weeks after rains
	C. (C.) pipiens fatigans	++	"
	Culex several species	++	"
	Anopheles (M.) gambiae	(+)	after rains
	Eretmapodites quinquevittatus	(+)	"
Larval collections			
Indoors	A. (S.) aegypti	++++	all year round
	C. (C.) p. fatigans	(+)	occasionally introduced from water cisterns
	C. (L.) tigripes	(+)	"

[illegible]	Species collected	Frequency of appearance	Time of peak
Landing-biting catch indoors	A. (S.) aegypti	++++	all year round
	A. (B.) albothorax	(+)	dry season
	Anopheles several species	+	"
	Mansonia spec.	+	4-8 weeks after rains
	Culex (C.) p. fatigans	++	3-5 weeks after rains

Note 1 : Aedes (S.) soleatus has been collected regularly by D. Fanara in the Bojuna forest, a habitat which can be described as extradomiciliary. The species has, however, not been encountered yet during the monitoring routines carried out exclusively in domiciliary and peridomiciliary habitats.

Note 2 : The identifications of the species listed have not been verified in all cases yet.

MEAN MAXIMUM AND MINIMUM TEMPERTURES

FIGURE 1

29.7, 31.2, 31.9, 32.2, 32.5, 31.6, 32.7, 32.2, 32.7, 32.6, 33.6, 31.9, 32.2, 32.1, 32.3, 32.1, 31.7, 30.4, 27.1, 27.1,

21.9, 21.9, 22.5, 21.9, 23.3, 23.3, 21.6, 21.6, 21.8, 22.6, 22.4, 21.9, 23.6, 23.1, 23.5, 22.3, 22.8, 22.6, 21.9, 21.9,

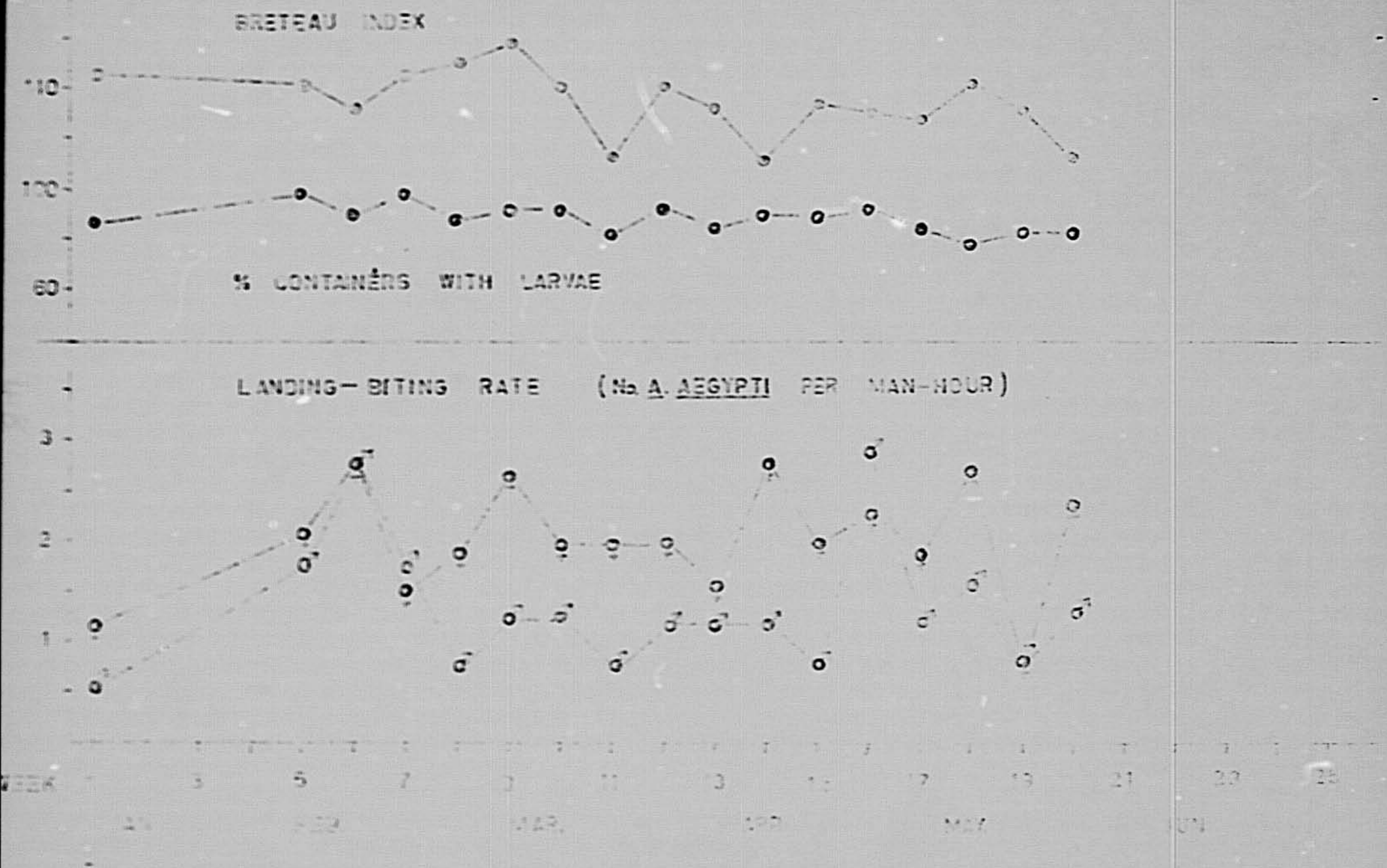
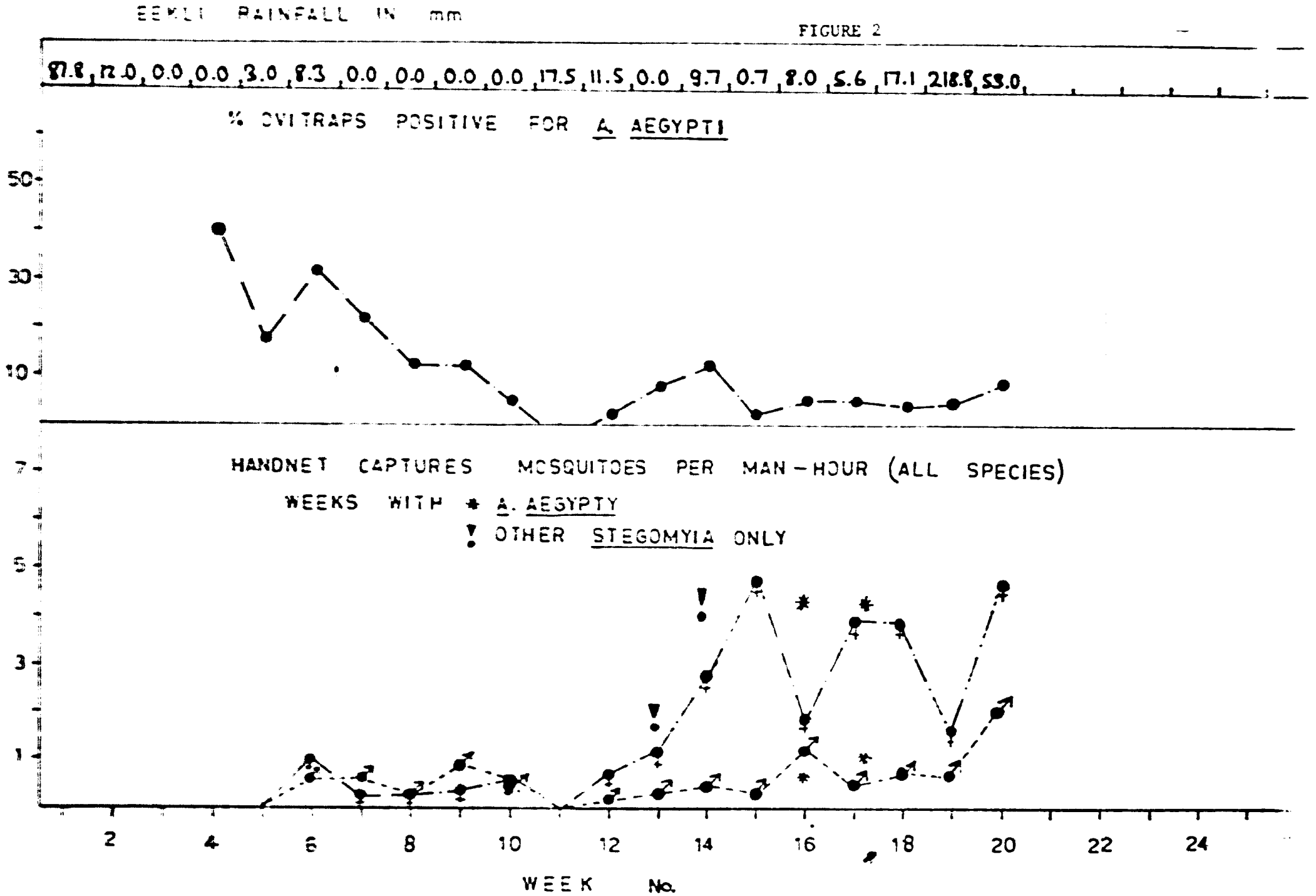


FIGURE 2


WEEKLY RAINFALL IN mm
87.8, 12.0, 0.0, 0.0, 3.0, 8.3, 0.0, 0.0, 0.0, 0.0, 17.5, 11.5, 0.0, 9.7, 0.7, 8.0, 5.6, 17.1, 218.8, 53.0
% OVITRAPS POSITIVE FOR A. AEGYPTI
50
30
10
HANDNET CAPTURES MOSQUITOES PER MAN-HOUR (ALL SPECIES)
WEEKS WITH * A. AEGYPTY
! OTHER STEGOMYIA ONLY
7
5
3
1
2
4
6
8
10
12
14
16
18
20
22
24
WEEK No.

Interim Report to Project 2

Title: Construction of lifetables for indoor and outdoor populations of A. aegypti.

Responsibility: W. Hausermann

Introduction

The construction of a life- or life-fertility-table for a field population of A. aegypti is not a one experiment affair, but a way to correlate diverse experiments and to present quantitative information on natality, mortality, fertility, dispersal, life expectancy and key environmental factors producing population change in a synoptic form.

This report presents the results of a recapture experiment carried out in an experimental village of the Rabai area, called Shauri Moyo.

Purpose:

a. It is hoped, that the experiment will provide quantitative information on adult natality, mortality, dispersal and life expectancy of the Shauri Moyo A. aegypti population during the last part of the dry season, April 17 to May 26, 1972.

b. At the same time it was also attempted to develop a fast and reasonably accurate method to estimate the total size of adult populations of A. aegypti in experimental villages prior to a release or a control experiment.

Course of the experiment and methods employed

The experiments reported below were carried out during the dry season (March) to the beginning of the rainy season (May) 1972. Experience with marking methods was obtained in two pilot mark-realease experiments.

First two fluorescent dusts were used for marking and a battery operated UV lamp for recognition of the recaptured individuals.

Afterwards Humbrol spraypaint was used for marking the captured mosquitoes individually on thorax or wings. The paint was applied first with a fine brush and later with a minuten pin at the tip of a small rod. The tip of the minuten pin was twisted into a small loop out of which the exact size of drop could be placed on thorax or wing. Most critical for this procedure was to thin the Humbrol spraypaint to the right consistence. Ether was used as a thinner. For marking, the mosquitoes were handled as described by McClelland and Conway (1971).

In the pilot experiment marks were applied as one or two dots on thorax or wings. The recovery of wing-marked specimens was significantly less than thorax-marked ones. Three versus 15 of 40 individuals marked each way. Thus for the large scale marking experiment only thorax-marking was used.

Mosquitoes were collected in landing-biting catches in all houses of the village. The mosquitoes caught were kept separately for each house, marked during the day and released between 1600 and 1700 in the house, where they had been caught. A mosquito was considered released, when it managed to escape from the vertically held testtube, in which it was caught. Mosquitoes which could not escape were treated as handling casualties and killed.

The same effort was made to catch mosquitoes from each house. In the beginning 6 casual helpers were employed to catch between 800 and 1300 h. The total man hours summed up to 30 h per day. The second experiment showed that two men catching for half an hour each in each of the 16 houses collected as many mosquitoes as before in only 16 man hours. This observation indicates that the numbers of mosquitoes in a village are fairly small.

For the pilot experiments the releases and recaptures were made according to the pattern prescribed for Bailey's triple catch method (Bailey, 1951/52). The estimates of the number of mosquitoes - males and females - present in the village during these two pilot experiments were 429 and 578 respectively.

After the experience gained in these pilot mark-release-recapture experiments a multiple recapture experiment lasting for 40 days was finally started. The captured mosquitoes were numbered individually. By placing small dots of paint of two colours on predetermined positions on the scutum 255 numbers could be applied with one colour pair. A total of 920 individuals, male and female, were numbered in a 15 days period. Follow up recaptures were carried out daily up to 40 days after marking had started. The total number of individuals recaptured at least once in this period is 276 for females and 62 for males. Additional catches made during the routine monitoring produced the recaptures of a female 43 days after it had been marked.

Results

The results presented here rely on a first hand extraction of the capture and recapture data and are only preliminary. Listings of the available life history information for all marked mosquitoes will be sent to Dr. Crovello at UND for data processing and statistical analysis.

The results as far as obvious already now are:

1. Distribution of the adult mosquitoes within the village as assessed by landing-biting catches is closely correlated to the distribution of larval habitats. Houses with good breeding were also houses with good catching (table 1, figures 1 and 2). The distribution of unmarked individuals, represented by first captures is almost the same as for recaptured individuals. However, the males seem to be more evenly distributed than the females, for in houses with low landing-biting activity the proportion of landing males was higher than in houses of high landing-biting activity. This trend is illustrated in figure 3, where the percentage of males is plotted as function of the total landing-biting catch, and would imply that males disperse more randomly than females.

2. Dispersal activity of marked individuals is presented in table 2 and figure 4. Of the total of 689 observations of marked mosquitoes subsequent to the release 599 were made on females and 90 on males. 42% of the observations on females and 39% of the observations on males indicated a move

from one house to the other between subsequent observations (table 2). Thus females and males moved rather freely between the houses. (Note that no oviposition of indoor females was recorded in ovitraps placed outdoors). Almost all houses of the village received immigrants from more than half of the other houses of the village and vice versa. This is demonstrated in figure 4 for house 15 from which comparatively few connections were made with other houses.

Marked males and females were approximately equal in their dispersal activity. The more even distribution of males through the village which was indicated in the distribution of captures of unmarked individuals could be explained, however, by a more random dispersal in the period between emergence and sexual maturity, before the male is attracted by man and caught in a landing-biting catch.

No recaptures were made in house 16, a tiny bachelor's hut without water, but the first captures of mosquitoes were only made after the marking period was over.

3. Life expectancy, mortality, natality

No attempts were made yet to use the recapture data for estimates of these parameters. As a start of an analysis the survivorship curves of the observed (=recaptured) males and females are presented in figure 5. The upper curve represents the number of individuals, males and females separately, seen last at any given day after marking. The lower curve represents the total number of survivors among the observed individuals for any given day after marking. The real survivorship curve must be higher than the curve drawn, for it must be considered that not all individuals were of the same age when marked and that a number of individuals survived without being recaptured.

Formulas have been developed to estimate the real number of individuals surviving to any given day (Jolly, 1965), but the laborious calculations have not yet been started.

There is a notable bend in the observed survivorship curves of males and females on day one after marking. The numbers surviving to one day after release are lower than would be expected, i.e. lower than if the curve were smooth. This bend may be due to emigration or mortality caused by the marking and will be investigated in a further marking experiment with a different marking method.

4. Population fluctuations, population density

The total number of captures per day are presented in figure 6 for males and females separately. For space reasons the graph shows the captures only up to day 37 (May 22) instead of day 45 (June 7), when the last marked female was caught accidentally. The daily capture rates fluctuated widely and it is difficult to distinguish real population trends from fluctuations caused by daily events. Counts of the pupal output in the 23 habitats available (21 positive at the beginning) indicated that the rise in total captures from April 23 to 26 and the rise of male captures from May 18 to 21 was a real population increase following a peak of pupal output 4 - 5 days earlier. Unfortunately the pupal counts carried out by a field

assistant turned out to be unreliable at the last inspection and no quantitative correlation is possible. That the observed increase in captures between April 23 and 26 represents a real population increase is also indicated by the proportional decrease in recaptures for the same period.

The decline in male captures from April 26 to May 5 also must indicate a real population trend, for, disturbed by the assistant counting pupae every day and stirring up the sediments in the water pots, 4 housewives started cleaning their pots on April 21 and 22, thus reducing the pupal output. No steps were undertaken to stop this and luckily enough the largeness and fragility of many of the water pots prevented too rigorous a control to endanger the experiment.

It seems that the males are better indicators for population trends due to their shorter lifetime and because their coming to man is not influenced by bloodmeals taken and by gonotrophic cycles.

No attempts were made yet to estimate the total population size. The formulas developed for estimates based on multiple recapture need very laborious calculations (Jolly, 1965; Fisher-Ford, 1947), which will be carried out on a computer by Dr. Crovello in UND. The high percentage of recaptures by the end of the marking experiment indicates a small population.

5. Bloodfeeding and duration of gonotrophic cycle

During the experiment, a note was made on blood content and ovarian development, when the female was marked and each time it was recaptured.

Judging from the 139 females which were recaptured more than once there is little correlation between feeding pattern and ovarian development between subsequent oviposition. Even females which had started but not terminated oviposition were caught taking bloodmeals. Several females were seen to go through up to three and four gonotrophic cycles during 20 to 30 days of observation. The reliability of our judging the stage of ovarian development will be tested in a special experiment. If the classification is reliable it would be possible to estimate the average duration of the gonotrophic cycle in nature. At present there are data on females which terminated a gonotrophic cycle in three days, while other females needed 2 and 3 bloodmeals until oviposition and other females even came 4 and 5 times to the mosquito catchers until they managed to get a bloodmeal. This corresponds with the findings of Sheppard et al. (1969) and McClelland & Conway (1971).

6. Dispersal of males and females between emergence and sexual maturity and insemination of females

The importance of information on this interim period between emergence and the onset of sexual and reproductive activity is self evident and has been pointed out in the above paragraphs on dispersal and survivorship.

A pilot experiment has been made with mosquitoes marked with fluorescent dust 1 - 22 hours after emergence. Of the females recaptured 18% were inseminated in less than 48 hours, 65% in less than 72 hours and 82% in less than 96 hours. All females caught 96 hours after emergence were inseminated. The total number of females recaptured was 69. A more extensive experiment of this kind is planned.

Conclusions

Conclusions made at the present stage of evaluation can only be fragmentary and indicate the direction of further experiments. Three of these have already been suggested above.

References

Bailey, N. T. J., 1951. On estimating the size of mobile populations from recapture data. Biometrika, 38: 293-306.

_____ 1952. Improvements in the interpretation or recapture data. J. Anim. Ecol., 21: 120-127.

Fisher, K. R. A., & Ford, E. B., 1942. The spread of a gene in natural conditions in a colony of the moth Panaxia dominula L. Heredity, 1: 143-174.

Jackson, C. H. N., 1939. The analysis of an animal population. J. Anim. Ecol., 8: 238-246.

Jolly, G. M., 1965. Explicit estimates from capture-recapture data with both death and immigration-stochastic mode. Biometrika, 52: 225-247.

McClelland, G. A. H., & Conway, G. R., 1971. Blood-feeding and gonotrophic cycle of the mosquito Aedes aegypti assessed by mark-release-recapture in a suburban habitat in Tanzania. WHO/VBC/71.272.

Sheppard, P. M., Macdonald, W. W., Tonn, R. J., & Grab, B., 1969. The dynamics of an adult population of Aedes aegypti in relation to Dengue haemorrhagic fever in Bangkok. J. Anim. Ecol., 38: 661-702.

Table 1 : Distribution of *Aedes aegypti* during mark-release-recapture experiment in Shauri Moyo, April 17 – May 26, 1972.

House No.	No. Inhabitants per house	Breeding level No. pupae per day	Total landing-biting-catch for whole period ♀	♂	No. of unmarked mosquitoes = first captures per house ♀	♂
2	9	no water	32	24	28	24
9	0	"	39	29	27	27
16	2	"	15	14	15	14
5	2	0 – 1	58	26	40	25
7	0	"	44	30	42	30
13	5	"	74	68	43	50
15	2	"	45	26	41	25
1	0	2 – 4	181	33	87	29
4	6	"	143	60	87	52
12	4	"	104	49	81	44
8	3	5 – 10	97	70	84	63
10	3	"	202	75	136	66
3	3	11 – 50	429	167	233	148
6	8	"	389	96	218	80
11	8	"	316	131	187	117
14	3	"	90	41	50	36
Total	58		2258	944	1399	830

Table 2 : Dispersal of marked *Aedes aegypti* during mark-release-recapture experiment in Shauri Moyo, April 17 - May 26, 1972.

House No.	No. released with individual mark in first 15 days per house		Dispersal activity of recaptured individuals between subsequent captures					
			Entering house n_i		Leaving house n_i		Balance for particular house	
	♀	♂	♀	♂	♀	♂	♀	♂
2	9	6	4	0	4	2	0	-2
9	7	6	9	0	9	1	0	-1
16	0	0	0	0	0	0	0	0
5	15	4	11	1	6	0	+5	+1
7	6	4	1	0	4	0	-3	0
13	12	10	17	8	6	0	+11	+8
15	22	9	8	1	10	2	-2	-1
1	26	13	30	3	21	3	+9	0
4	44	11	16	3	17	4	-1	-1
12	29	16	10	2	18	3	-8	-1
8	39	12	10	1	10	2	0	-1
10	59	17	24	4	28	1	-4	+3
3	102	75	27	1	45	7	-18	-6
6	124	31	33	4	42	6	-9	-2
11	54	31	34	4	23	4	+11	0
14	25	19	16	3	7	0	+9	+3
Total	573	264	250	35	250	35	0	0

FIGUR

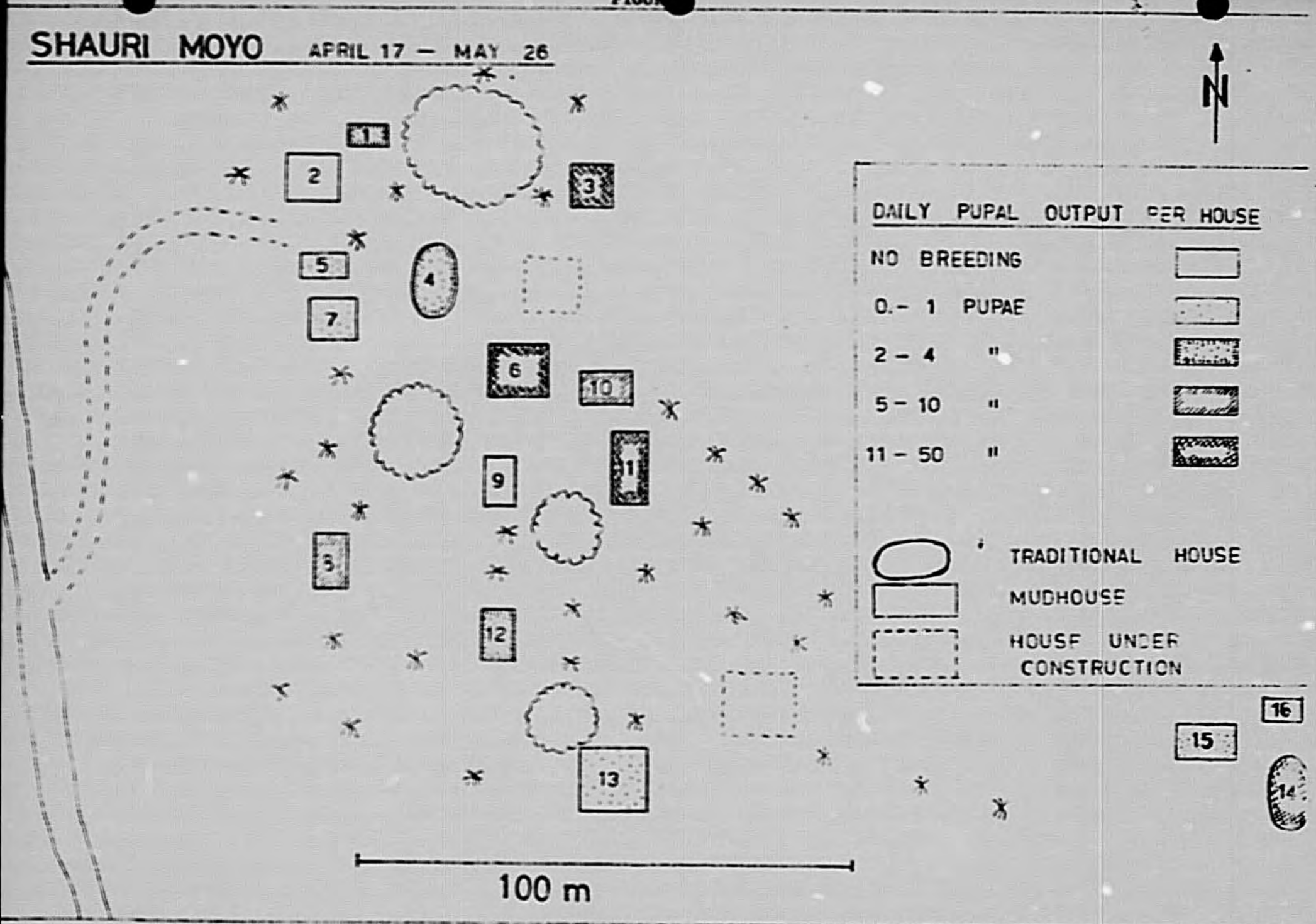

SHAURI MOYO APRIL 17 – MAY 26
N
DAILY PUPAL OUTPUT PER HOUSE
NO BREEDING
0.– 1 PUPAE
2 – 4 "
5 – 10 "
11 – 50 "
TRADITIONAL HOUSE
MUDHOUSE
HOUSE UNDER CONSTRUCTION
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
100 m

FIGURE 2

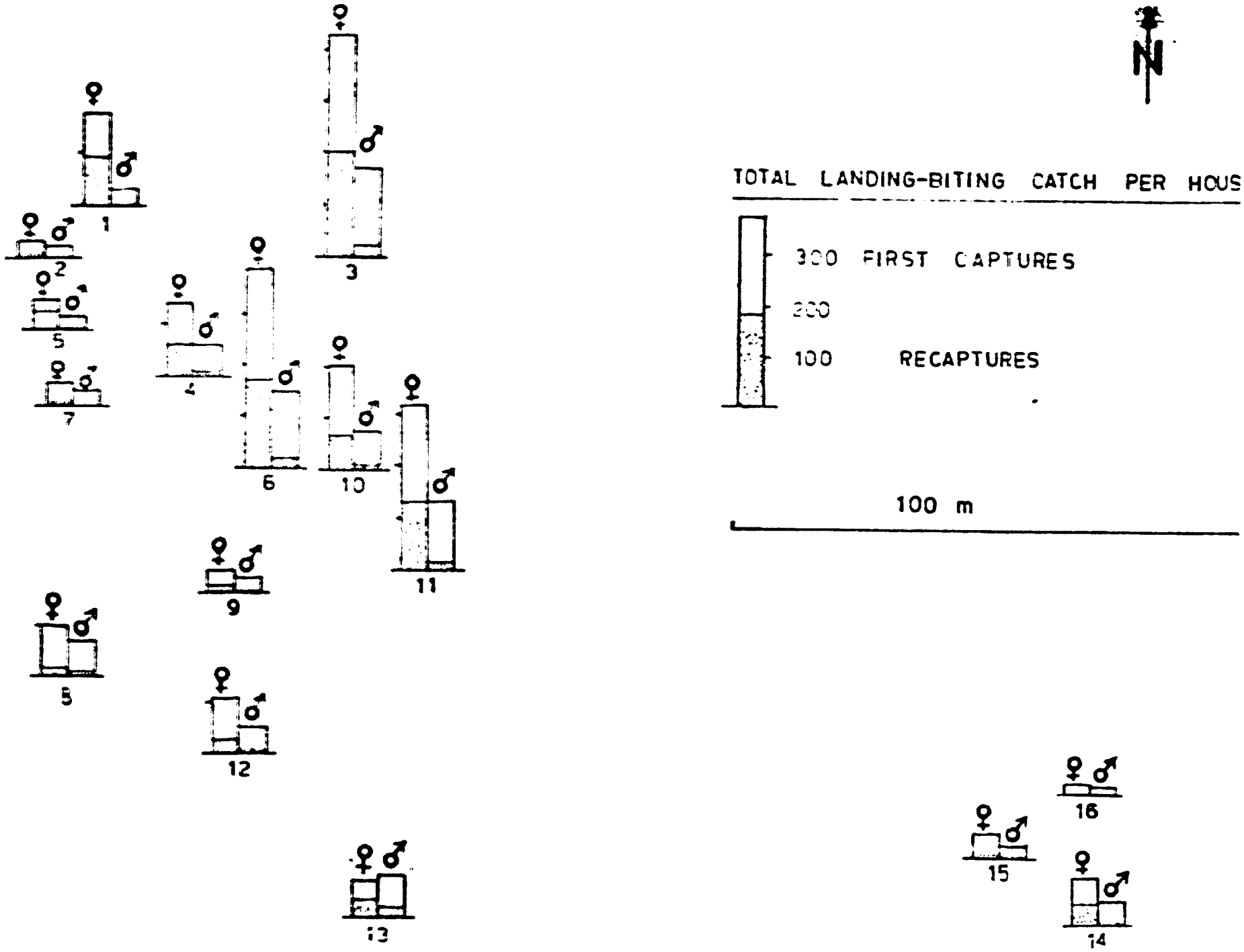

TOTAL LANDING-BITING CATCH PER HOUS
300 FIRST CAPTURES
200
100 RECAPTURES
100 m
N
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16

FIGURE 3

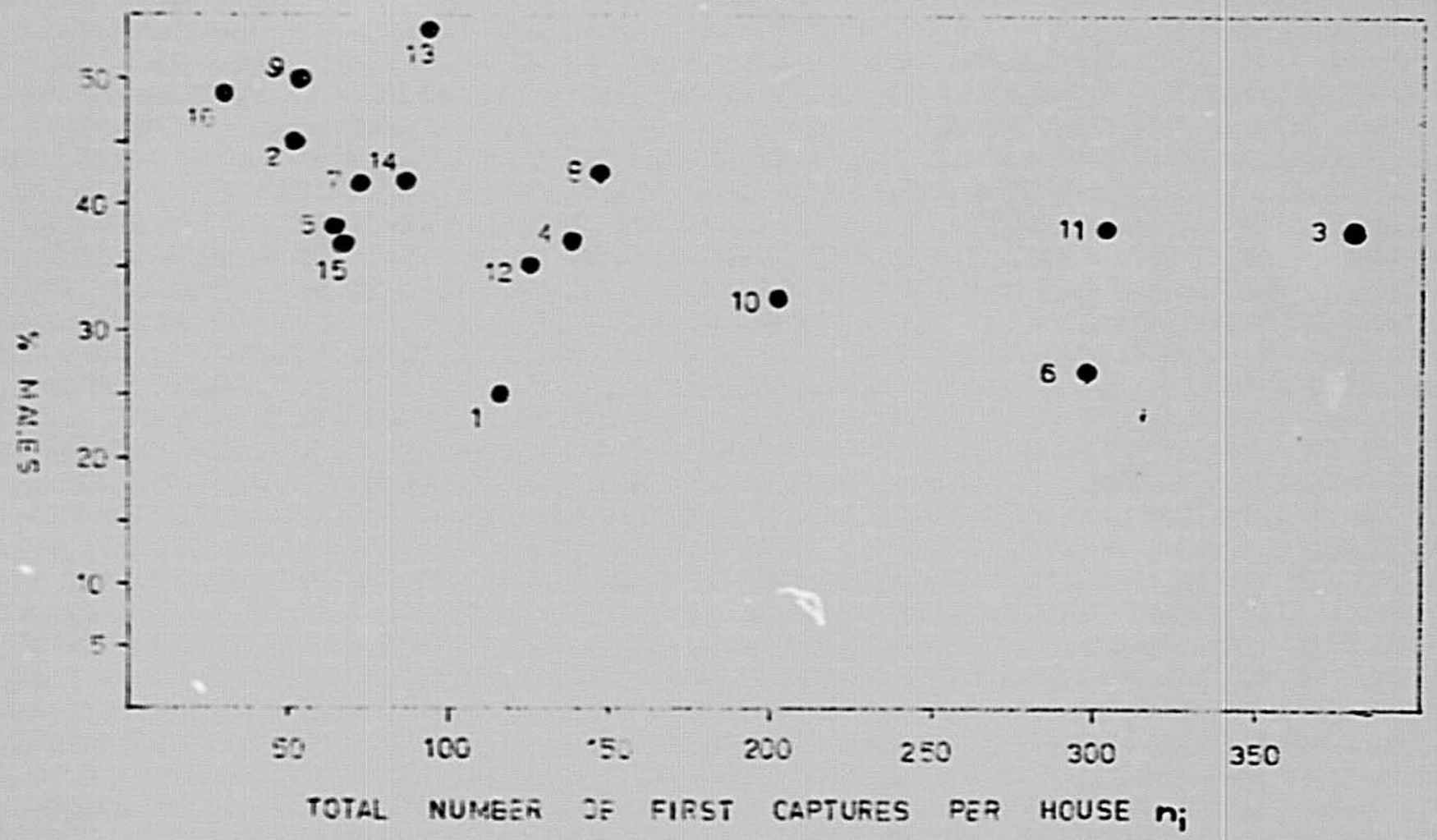

% MALES
50
40
30
20
10
5
50
100
150
200
250
300
350
TOTAL NUMBER OF FIRST CAPTURES PER HOUSE n_i
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16

FIGURE 4

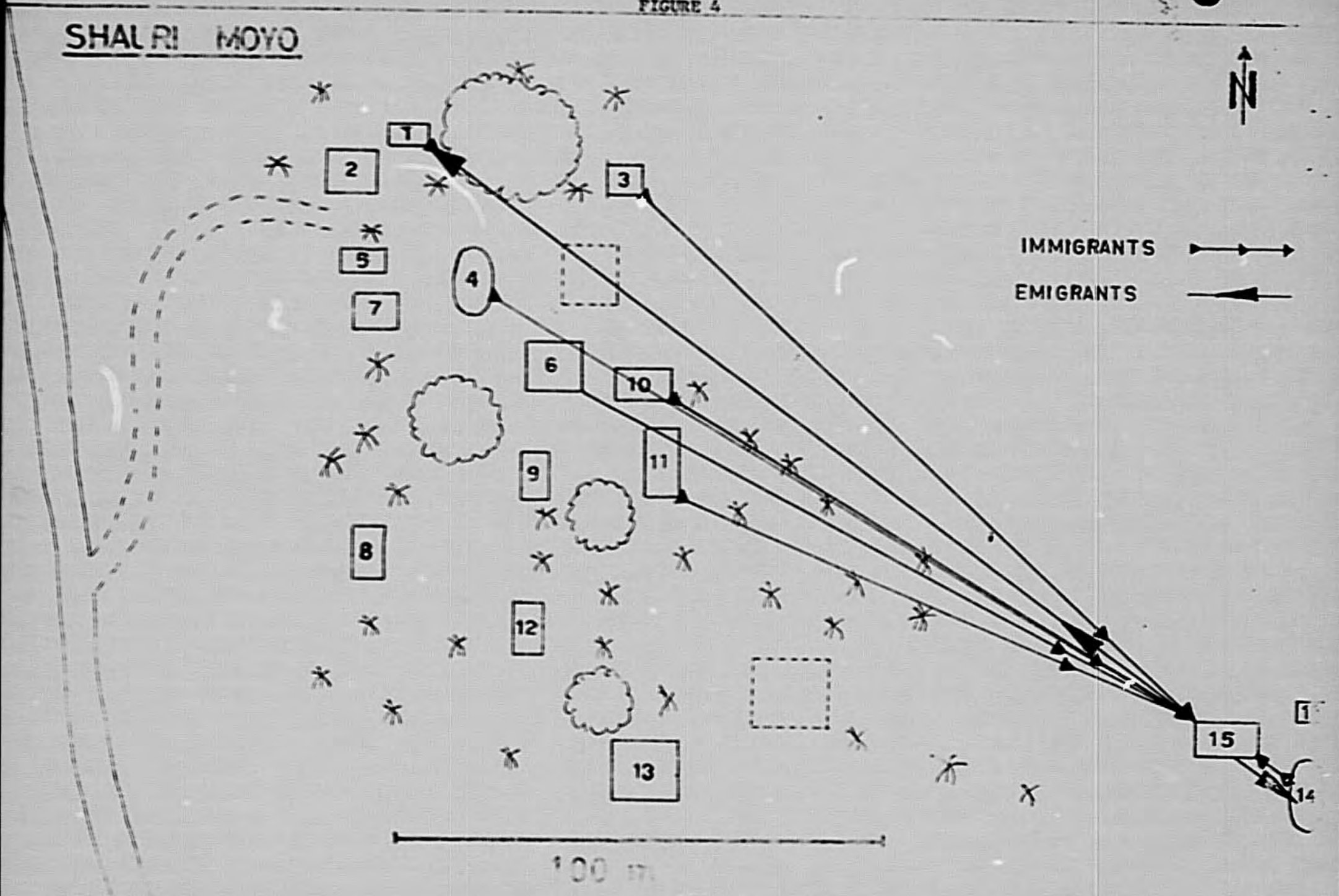
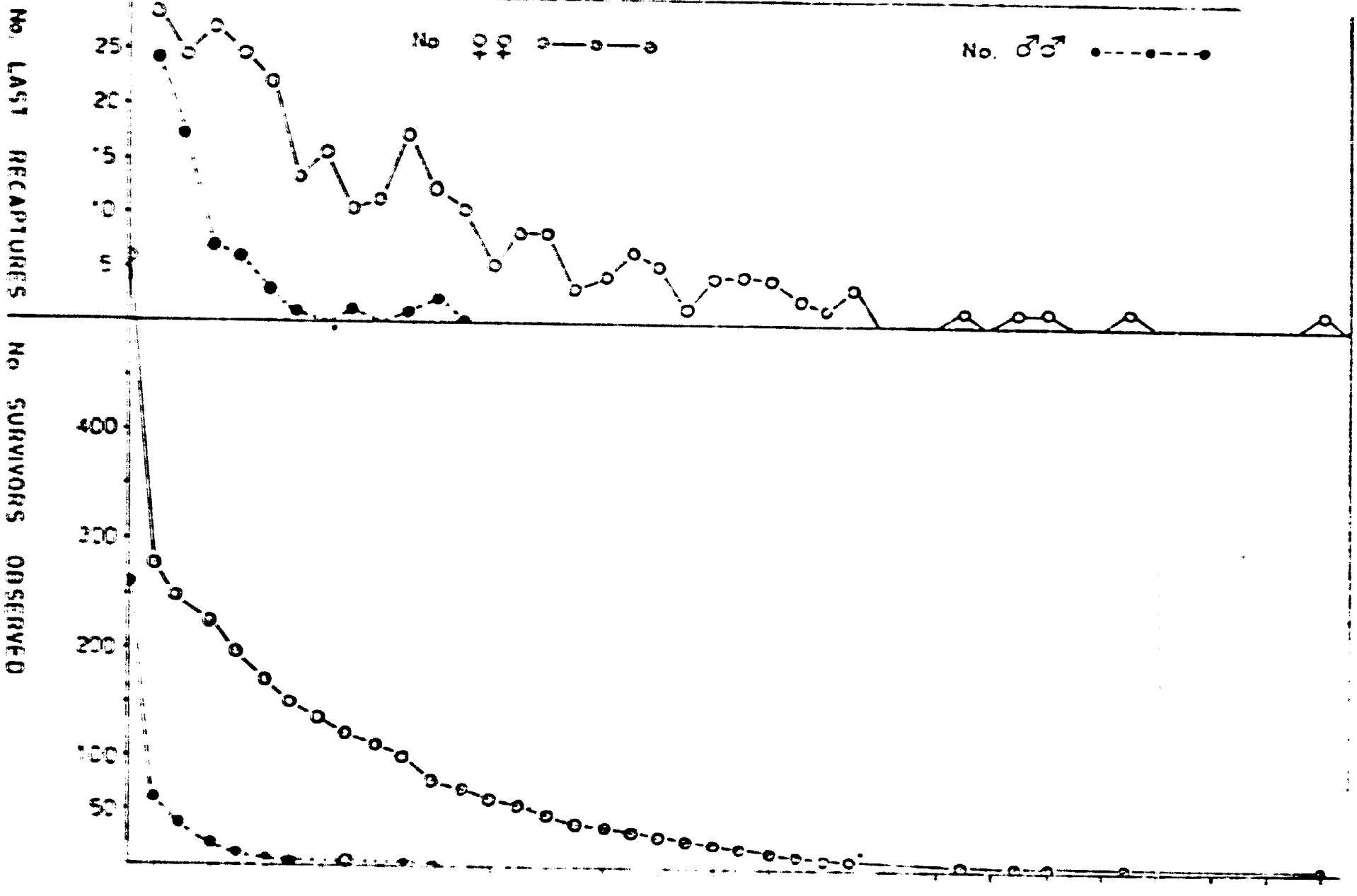
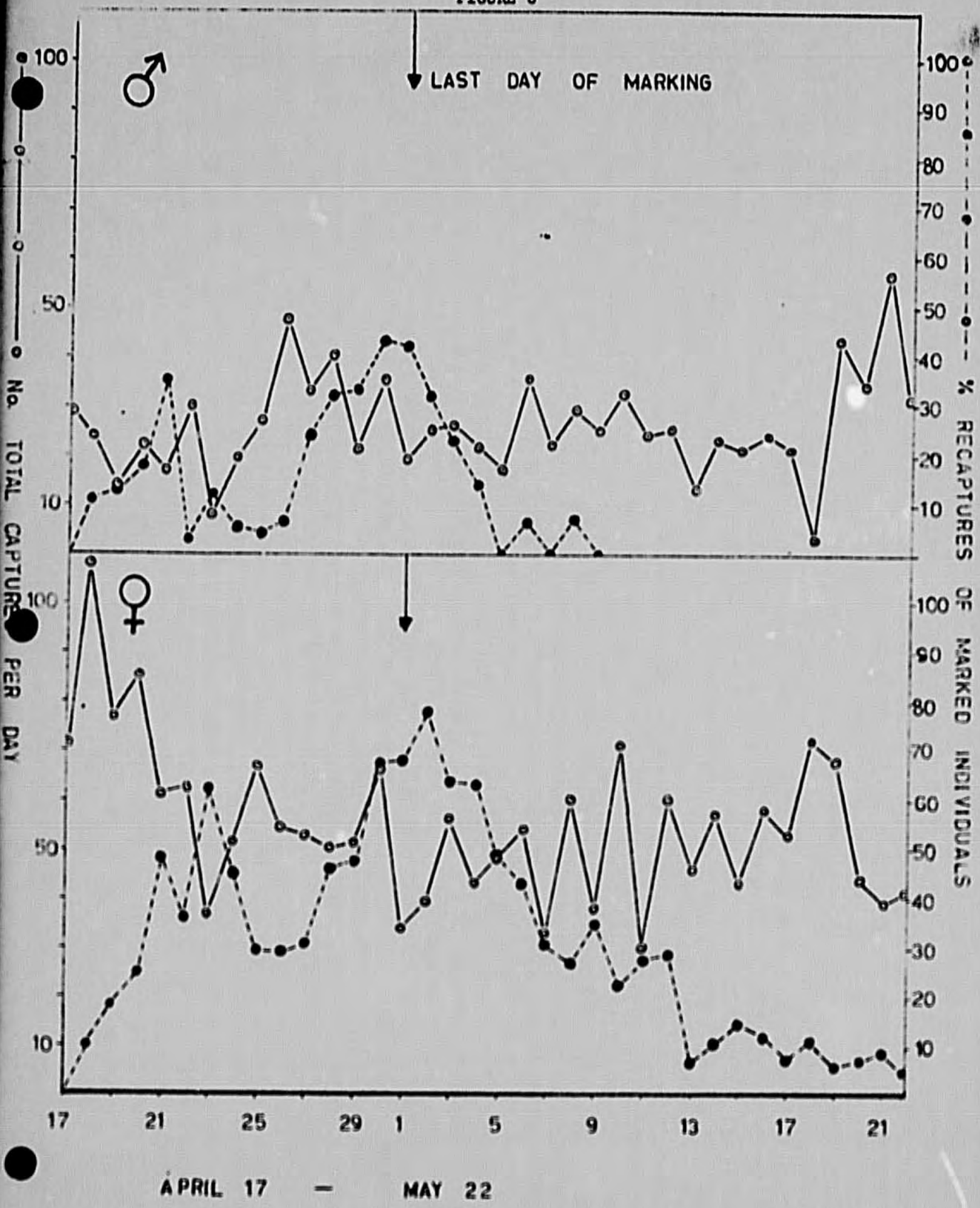

SHALRI MOYO
N
IMMIGRANTS
EMIGRANTS
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
100 m

FIGURE 5


No. LAST RECAPTURES
No SURVIVORS OBSERVED
No ♀♀
No. ♂♂
25
20
15
10
5
400
300
200
100
50


LAST DAY OF MARKING
♂
♀
No. TOTAL CAPTURES PER DAY
% RECAPTURES OF MARKED INDIVIDUALS
100
50
10
90
80
70
60
40
30
20
17
21
25
29
1
5
9
13
17
21
APRIL 17 — MAY 22

BREEDING SITES UTILIZED BY AEDES (STEGOMYIA) SPECIES IN THE BEJUMA STREAMBED FOREST (KENYA)

Dean M. Fanara
Report No. 1

INTRODUCTION

As a preliminary step in the investigation of gene flow between dark colored, forest breeding Aedes (Stegomyia) aegypti (Linnaeus) and the paler colored, indoor breeding form, it was useful to define the forest breeding habitat of the study area.

The Bejuma streambed begins about 70 meters from Momsabu Village in the coastal hills area of Changome near Mombasa, Kenya. This seasonal streambed runs for about 400 meters before emptying into the usually flowing Kombeni river. It is joined by one small side branch. The actual streambed area, including a nearby parallel ridge, is heavily forested with a moderate canopy and understory. The ridge is bisected by a canopy covered path used by the women of the village for obtaining their domestic water supplies and by groups of bathers.

This study was conducted during the dry season months of February and March 1972.

MATERIALS AND METHODS

A path was cut through the streambed proper. The researcher and his two assistants concentrated on locating all possible Aedes breeding sites in the swath of forest accompanying the Bejuma streambed, including the ridge with its path. All sites were flooded with water and the immediate hatching of Stegomyia species identified the site as positive. Each positive site was numbered and figured on a map.

A sample of larvae was removed from each numbered site and reared in the MBU/ICIPE Laboratory at Mombasa. The emerging adults were sexed and keyed to species employing the keys of Edwards (1941) and Gerberg and van Someren (WHO/VBC/70.236).

The physical parameters of each breeding site were measured.

Sixteen water filled bamboo stems were spaced on a transect running from Momsabu village to the Kombeni river. These ovipositional sites were allowed to dry and then were reflooded. Then a sample was removed from each stem and laboratory reared for identication.

RESULTS

The number of sites identified as positive for Aedes (Stegomyia) was 45. In Table I it can be noted that the larvae of Aedes heischi van Someren were present in 32 sites, Ae. aegypti in 29 sites, Ae. calceatus Edwards and Ae. vittatus (Bigot) in 8 sites, Ae. metallicus (Edwards) and Ae. soleatus Edwards in 7 sites, while Ae. simpsoni (Theobald) was in 3 sites. Aedes of other subgenera were present, but are not covered in this report. They occurred in 57% of the sites that were positive for the Stegomyia.

RESULTS (continued)

Ae. aegypti shared 22 or its 29 positive breeding sites with Ae. heischi, but only 4 out of 8 (50%) with Ae. vittatus. By contrast only once did Ae. heischi share with Ae. vittatus. In 8 sites Ae. vittatus occurred in rock holes while Ae. heischi occurred in rock holes only in 2 out of 32 sites. Ae. aegypti utilized 7 of the 11 rockholes, 20 of the 32 tree holes and of the 16 bamboo stems Ae. heischi was the only other Stegomyia utilizing the bamboo stems (4 out of 12 with only 2 overlapping with Ae. aegypti).

DISCUSSION AND CONCLUSIONS

Ae. aegypti utilized the largest variety of possible breeding sites in the Bejuma streambed forest. This ability to utilize such a wide array of breeding sites certainly gives a clue as to the way in which it became associated with man.

Ae. heischi was more selective in its choice of breeding sites, but did appear to utilize every site that fitted the description of a shaded tree hole.

Ae. vittatus was more ubiquitous in this forest than the larval record would indicate. It confined its egg laying mainly to the large isolated pot holes in the Kombeni river and in the streambed proper of the side branch of the Bejuma. The side branch is more level and contains large potholes which fill with water during brief showers.

The other Stegomyia were found to be breeding in tree holes but appeared in limited numbers as compared with the aforementioned species.

REFERENCES

Edwards, F. W. 1941. Mosquitoes of the Ethiopian Region III, London, Brit. Mus. Natur. Hist., 499 p.

Table I

ADULTS OF *AEDES* (*STEGOMYIA*) REARED FROM BEJUMA FOREST SITES

Site	Type*	Height (m.)	Vol. (c.c.)	*aegypti*	*calceatus*	*heischi*	*metallicus*	*simpsoni*	*soleatus*	*vittatus*
1	R	0.0								14
2	R	0.0								1
3	R	0.0		1						23
4	T	0.2		10		5	1			
5	T	0.8								
6	C	1.0		10		1				
7	T	1.0		6		1				
8	R	1.0	740	20	1	1				
9	T	0.8		9		4				
10	R	0.0		4						7
11	R	0.0		4						1
12	R	0.0		20						2
13	R	0.0								11
14	R	0.0	320	17		5				
15	T	5.0			2	6			2	

Table I cont.

Site	Type*	Height (m.)	Vol. (c.c.)	aegypti	calceatus	heisoni	metallicus	simpsoni	scleatus	vittatus
16	R	0.0		21						
17	T	0.2		5	1	18				
18	R	0.0		39						
19	T	1.2	580	10		1			1	
20	T	2.4				7				
21	T	0.8		4		13				
22	T	1.5		2	2	16			5	
23	T	0.3		12		1				
24	T	0.5				7				
25	T	1.0				10				
26	T	0.3		1		13				
27	T	3.0		2		19		1	1	
28	R	0.0		1						
29	T	0.2		13		1		1		
30	T	0.7					1			
31	T	2.0				2			7	
32	T	0.2				7				

Table I cont.

Site	Type*	Height (m.)	Vol. (c.c.)	aegypti	calceatus	heischi	metallicus	simpsoni	soleatus	vittatus
33	T	2.0	90	1	4	18				1
34	T	1.7		6		1	1			
35	T	0.4	115	11	2	12		1		
36	T	1.8	460	2	2	21	2			
37	T	0.5	190			8			1	
38	T	0.5	70			11	2			
39	T	0.7	55			13	1			
40	T	1.3	330	10			1			
41	T	2.2	3,500	7		3				
42	T	0.6	160	18		4				
43	T	1.6	80	1		11				
44	T	0.0		10	2	2			1	
45	T	0.3	30			12				
			TOTAL	276	16	273	9	3	18	60
			**PERCENT	42.2	2.4	41.7	1.4	0.5	2.7	9.2

* B = baobab seedpod, C = coconut shell, T = treehole, R = rock hole
** Totals 100.1% due to rounding off.

INDOOR AND FOREST BITING BY AEDES (STEGOMYIA) AT CHANGÓMBE NEAR MOMBASA, KENYA

Dean M. Fanara

Report No. 2

INTRODUCTION

It was necessary to understand some of the dynamics of indoor and forest biting by Aedes aegypti (L.) in preparation for a mark, release, and recapture experiment. This study was conducted in the area of Changombe concentrating on Momsabu village hut clusters on a hill overlooking the Kombeni river. Linking Momsabu village to the forested Kombeni river is the seasonal, forested streambed known as Bejuma. This study was conducted mostly during April 1972, a dry spell.

More extensive studies have been conducted in coastal areas of East Africa by other researchers such as Trpis (Who/VBC/71.291) in Dar es Salaam, Tanzania and Teesdale (1955) in the Mombasa, Kenya area.

MATERIALS AND METHODS

The indoor landing biting catch was performed by having one field assistant enter one hut and seat himself by the water storage pot. He caught, with an aspirator, all mosquitos visiting his legs and placed them in one special vial per hour. He was replaced by the man conducting the forest biting study after 4 hours to prevent fatigue. A flashlight was used as an aid to capture.

The vial was prepared from a plastic bottle in which a nylon mesh was fitted to a portion of the cap center which was cut out. A hole just larger than the aspirator tube was burned into the side and a specially cut piece of bicycle innertube was fitted over the hole. It could be moved aside with the thumb of the hand holding the vial. All vials were stored in a plastic refrigerator dish with damp cotton to maintain the mosquitos alive.

The outdoor landing-biting catch was performed by the field assistant starting at one of 7 sites, on a rotational basis, and moving each hour to a new site. The sites were spaced as to evenly cover the 400 odd meters of the Bejuma streambed forest. The same capture technique was employed as used in the hut except that instead of a waterpot, a tree hole or rock hole positive for Aedes (Stegomyia) was utilized.

RESULTS

The indoor biting by Aedes (Stegomyia) at Momsabu village was confined solely to an intermediate color form of Ae. aegypti. This form is intermediate between the pale form and the dark form depicted by Edwards (1941).

The results of the indoor landing-biting catch as shown in Table I indicate that a biting peak was reached and maintained throughout the morning. Teesdale (1955) reported a morning peak and evening peak in a study conducted only a few miles away. The initial pilot study conducted by this researcher included captures from dawn until dusk and indicated dawn and dusk biting. These results were not significant enough to report on, and such early and late captures were very unpopular with the women of Momsabu village.

The forest landing-biting by Ae. aegypti at Bejuma forest was composed of 8.5% intermediate color males, 21.3% intermediate color females, 10.6% dark form males and 59.6% dark form females. This was during the dry period of April 1972 when all the Ae. aegypti reared from tree holes were exclusively the dark form.

The results of the forest landing-biting by Ae. aegypti are depicted in Table II. Peak biting occurred between 0830 and 1130 with a short burst of activity at dusk. Male Ae. aegypti visited the human bait early in the morning, but not at dusk.

The other Aedes (Stegomyia) which was biting in large numbers was Ae. vittatus (Bigot). As seen in Table II it was actively biting during the daylight hours until about 1730. The males of Ae. vittatus often visited the host as did those of Ae. aegypti.

Ae. calceatus Edwards and Ae. heischi van Someren each visited the human bait 3 times during the total of 127 man hours of capture.

Mansonia spp. entered the forest situation in rates up to 14 bites/man-hour at the end of April. By April 21 the indoor biting was suddenly taken over by Mansonia spp. in rates up to 7 bites/man-hour. At this point the experiments were terminated.

DISCUSSION AND CONCLUSION

No real discussion will be made regarding the indoor biting periodicity because of the small sample size and limited duration of sampling. It is interesting to note that about 1/3 of the visits are made by males. Laboratory rearings of indoor water pot samples indicate 57:43 male to female sex ratio.

The entrance of Mansonia spp. into the huts was also marked by an abrupt reduction of Ae. aegypti indoors. Possibly this was due to competition or due to the deligent pot cleaning brought on because of the attention paid to the pot contents by the mosquito captures. At the termination of the experiment only 2 houses had mosquito larvae in their water pots, while at the beginning 9 out of 10 were positive.

The appearance of Mansonia spp. did not decrease the biting rate of Ae. aegypti in the forest situation. The Mansonia spp. were passing their larval stages in the dense mats of Pistia sp. (water lettuce? growing in the still pools of the Kombeni river.

The large numbers of Ae. vittatus biting in the Bejuma streambed were breeding in the pot holes of the Kombeni river. Ae. heischi, which was reared in large numbers from tree hole samples, apparently did not favor man as a host.

The average daily biting rate, during daylight hours (630-1830), was 0.37 bites per man-hour for Ae. aegypti and 0.54 bites per-hour for Ae. vittatus. But note that Ae. aegypti males visited man at the rate of 0.06 per hour while Ae. vittatus visited at the rate of 0.46 per hour. The males of Ae. vittatus visit man more than the males of the forest variety of Ae. aegypti. The indoor variety of Ae. aegypti had a ratio of 0.3 males to 0.7 females visiting the host per hour. This would indicate that either indoor males have a greater preference to visit the host or that the conditions are simply more favorable.

The fact that 29.8% of the Ae. aegypti caught in the forest situation were of the type seen 100% of the time in the hut situation indicates that some gene pool mixing may occur. Yet, all larvae reared from the forest situation were observed to be of the dark form of Ae. aegypti. A more indepth study is indicated. Also note that the large amount of man activity including flooding of breeding sites, may make the reported results rather artificial. The same experiment will be conducted in the future with less environmental manipulations.

REFERENCES

Teesdale, C. 1955. Studies on the bionomics of Aedes aegypti (L.) in its natural habitats in a coastal region in Kenya. Bull. Entomol. Res. 46: 711-743.

Table I

MEAN NUMBERS OF *AEDES AEGYPTI* LANDING-BITING INDOORS AT MOMBASA VILLAGE IN A TOTAL OF TEN HUTS DURING APRIL 1972

Time Period	Mean Number/Man-Hour Males	Mean Number/Man-Hour Females	Total Hours
0800-0900	0.3	0.6	9
0900-1000	0.5	1.1	10
1000-1100	0.0	0.7	10
1100-1200	0.6	0.9	8
1200-1300	0.3	1.0	6
1300-1400	0.1	0.6	7
1400-1500	0.4	0.0	7

Table II

MEAN NUMBERS OF AEDES (STEGOMYIA) LANDING-BITING IN THE BEJUMA FOREST DURING APRIL 1972

Time Period	*Aedes aegypti* Mean Number/Man-Hour Males	*Aedes aegypti* Mean Number/Man-Hour Females	Total Hours	*Aedes vittatus* Mean Number/Man-Hour Males	*Aedes vittatus* Mean Number/Man-Hour Females
0630-0730	0.11	0.30	13.5	0.22	0.89
0730-0830	0.06	0.25	16.0	0.38	0.44
0830-0930	0.03	0.50	16.0	0.19	0.44
0930-1030	0.13	0.68	15.5	0.52	0.52
1030-1130	0.00	0.69	8.0	0.14	0.14
1130-1230	0.00	0.24	8.5	0.82	0.24
1230-1330	0.14	0.14	7.0	0.00	0.85
1330-1430	0.00	0.40	5.0	0.12	1.00
1430-1530	0.12	0.31	8.0	1.38	0.63
1530-1630	0.10	0.19	10.5	1.05	0.48
1630-1730	0.00	0.30	10.0	0.90	0.80
1730-1830	0.00	0.47	7.5	0.00	0.00
1830-1930	0.00	0.00	1.5	0.00	0.00

A PILOT MARK, RELEASE AND RECAPTURE STUDY UTILIZING AEDES (STEGOMYIA) IN THE CHANGOMBE AREA OF COASTAL KENYA

Dean M. Fanara

Report No. 3

INTRODUCTION

In order to study the dynamics of gene flow (if any) between the hut-breeding Aedes aegypti (L.) and the forest breeding, darker form, it is important to understand the flight activity of the two forms.

The area of Changombe with its village of Momsabu, which is in close proximity to the forested streambed of Bejuma, was considered ideal for this study. The landing-biting rates for forest and hut breeding species, the breeding sites, the larval ratios and several other factors were already known (See Reports No. 1a and 2 of this series). Also a small field station had been constructed at Momsabu village and was useful for the temporary internment of the mosquitos to be marked.

The concept of this experiment was to mark the hut breeding mosquitos with one color (red) and the forest breeding mosquitos with another (green) and release them in their approximate site of origin. Then utilizing landing-biting catches the recovered mosquitos would yield information as to flight range and degree of spatial contact between forms of Ae. aegypti.

It was also considered that such an experiment would serve as a training exercise for a future release of genetically marked Ae. aegypti.

MATERIALS AND METHODS

Indoor. Larvae of Ae. aegypti were collected from the water storage pots of Momsabu and placed in plastic cups. The larvae were fed dried liver powder. The cups were placed in sleeve cages in a dark, cool, damp field station in Momsabu.

The newly emerged adults were aspirated into dusting chambers. Each dusting chamber was a one pint plastic refrigerator storage container fitted with screening at one end and an aspirator-size hole covered with rubber at the other end. The fine dust (1-2 micron particle size) of red fluorescent dye was blown through the chamber with an atomizer. All mosquitos were lightly dusted and would fluoresce brightly under the ultra-violet light.

The mosquitos were released at dusk in hut no. 5 which was about midway between the 10 huts used in this study (95 m. from no. 1 and 80 m. from no. 10). Table I illustrates the dates and numbers released. The sex ratios were estimates derived from a laboratory rearing of a sample of larvae collected from 9 of the 10 huts at Momsabu (see Report No. 1 of this series).

Forest. Larvae of several species of Aedes (Stegomyia) were collected from treeholes and rockholes in Bejuma forest. The larvae were reared and the adults dusted with fluorescent dye powder (green) in the same manner as reported for Ae. aegypti from huts of Momsabu.

The species composition and sex ratios were estimated from a laboratory (MBU/ICIPE at Mombasa) rearing of material from this forest. Report No. 1 of this series details the results of the rearings. However, the ratios used to calculate the numbers reported in Table II are slightly different since not all 45 breeding sites contributed larvae to this study. Table II indicates the numbers released and the site of release. Site no. 35 was approximately half way up the Bejuma streambed. It is a heavily forested spot with a thick canopy. The landing-biting rates were found to be high for this site. All releases were made at dusk.

Landing-biting catches were utilized as a means of checking dispersal. See Report No. 2 of this series for a study of the results of landing-biting catches at Momsabu and Bejuma during the period of this study. All captured material was analyzed at the field station with an ultra violet lamp.

RESULTS

Indoor. Table III illustrates the results of the indoor capture of marked mosquitos. This is the result of daily landing-biting catches made from 6 April to 19 April 1972. By 21 April 1972 the biting was completely taken over by Mansonia spp. which invaded the huts. The abundance of Pistia sp. (water lettuce) in the Kombeni river gave rise to this large population of Mansonia spp.

All the Ae. aegypti taken indoors were of the intermediate form. During this experiment the indoor male landing ratio was 0.36 landings per man-hour while the indoor female biting ratio was 0.54 (total of 54 man-hours of landing-biting capture). The male to female sex ratio was 3:6.

As shown in Table III, five marked females and one marked male were recovered. One female flew 90 meters while one male flew 95 meters. This was about the maximum range of the capturing stations (huts no. 1 to 10).

Forest. Table IV illustrates the results of the forest capture of marked mosquitos. This is the result of daily landing-biting catches made from 6 April to 21 April 1972. By 21 April numbers of Mansonia spp. as high as 14 per man-hour were captured in Bejuma forest. A total of 96 man-hours were employed.

Throughout this experiment the male landing rate for the intermediate form of Ae. aegypti was 0.04 per man-hour. The female biting ratio for this same form was 0.10 per man-hour. The sex-ratio was 3:7 males to females. The males comprised 2.6% of the forest capture and the females were 6.5%.

The male landing rate for the dark form of Ae. aegypti was 0.05 per man-hour The female biting rate for this form was 0.34 per man-hour. The sex ratio was 1:7 males to females. The males comprised 3.3% of the total forest capture and the females were 21.6%.

The male landing rate for Ae. vittatus was 0.47 per man-hour. The female biting rate for Ae. vittatus was 0.54 per man-hour. The sex ratio was nearly 1:1 The males comprised 29.4% of the total forest capture and the females were 34.0%.

The biting by females of Ae. calceatus, Ae. heischi, and Ae. metallicus comprised only 2.6% of the total and will not be considered.

As shown in Table IV two of the female Ae. vittatus which were marked and released in the forest were recaptured. The furthest distance traveled was 30 meters. The dark form of Ae. aegypti yielded only two females recaptured. Both were caught at the site of release. The most interesting point is the indoor marked (red) intermediate form of male and female Ae. aegypti that were captured in the Bejuma forest. The female was captured 90 meters from the site of release and the male was captured 237 meters from the site of release.

DISCUSSION AND CONCLUSIONS

Indoor. The continued release of small amounts of marked mosquitos was dictated by the small amounts emerging daily during this experiment. Because of this type of mark and release no attempt at applying a Lincoln Index or other population size estimate will be made. The important note is that the indoor populations do move about the hut cluster of Momsabu. They move up to some 90 meters at least.

No forest marked (green) Aedes spp. were captured indoors during this experiment. At no time during the dry season was a truly dark form of Ae. aegypti taken indoors.

Forest. Because of the type of release used no population estimates will be attempted. The Ae. vittatus did not appear to move any great distance. However, they were marked and released at about ½ the amount compared with the dark form of Ae. aegypti and so they must be adept at finding man. The two dark forms of female Ae. aegypti that were recovered were in the release site. The dark form of Ae. aegypti would appear not to readily seek man as a host. The hut recovery rate for the intermediate form of Ae. aegypti (female) was 0.09 per man-made hour, while the forest recovery rate for the dark form Ae. aegypti (female) was 0.02 per man-hour. The numbers released were comparable (see Table I and Table II).

The appearance in the forest of a hut marked, intermediate form, male Ae. aegypti at 237 meters from release site was most interesting. If the male would travel that distance and visit a host (perhaps seeking a mate?), would it not also copulate with a forest Ae. aegypti? And would it then contribute to the forest Ae. aegypti gene pool?

The appearance of a similarly marked female some 90 meters from a hut (and breeding site) also poses some questions. Where does she oviposit? Did she enter the forest as a virgin?

The sex ratios are interesting. The intermediate form of Ae. aegypti landing-biting in the hut was in a ratio of 3:6 (male:female). The intermediate form of Ae. aegypti landing-biting in the forest was in a ratio of 3:7. But the ratio of the dark form of Ae. aegypti in the forest was 1:7. It would appear that the males of the dark form of Ae. aegypti are not as attracted to humans as are the intermediate form males.

Table I

NUMBERS RELEASED FROM LABORATORY REARED HUT POPULATIONS OF *AEDES AEGYPTI* MARKED RED

Release Date	Release Site	Total	*Aedes aegypti* *Male	*Female
5/4	H5	100	57	43
6/4	H5	100	57	43
7/4	H5	100	57	43
8/4	H5	100	57	43
10/4	H5	25	14	11
13/4	H5	75	43	32
	TOTAL:	500	285	215

* Estimated on the basis of laboratory rearings of samples from sites used as breeding source for mosquitos for marked releases.

Table II

MOSQUITOS RELEASED FROM LABORATORY REARED FOREST POPULATIONS OF *AEDES* (*STEGOMYIA*) MARKED GREEN

Release Date	Release Site	Total No.	*Ae. aegypti* Male	*Ae. aegypti* Female	*Ae. vittatus* Male	*Ae. vittatus* Female
5/1	35	100	29	26	1	5
6/1	35	200	58	52	8	10
7/1	35	150	43	39	6	15
8/1	35	100	29	26	4	5
10/1	35	75	22	19	3	4
13/1	35	150	43	39	6	15
	TOTAL:	775	224	201	31	54

* Estimated on the basis of laboratory rearings of samples from sites used as breeding source for mosquitos for marked releases.

Table III

INDOOR CAPTURES OF MARKED MOSQUITOS

Date	Time	Site	Sex	Species	Dye Color	Distance from Release(m.)
11/4	08-0900	H8	f	*Ae. aegypti*	Red	73
11/4	08-0900	H8	f	*Ae. aegypti*	Red	73
11/4	09-1000	H8	f	*Ae. aegypti*	Red	73
11/4	09-1000	H8	f	*Ae. aegypti*	Red	73
12/4	09-1000	H1	f	*Ae. aegypti*	Red	95
18/4	13-1130	H2	f	*Ae. aegypti*	Red	90

Table IV

FOREST CAPTURES OF MARKED MOSQUITOS

Date	Time	Site	Sex	Species	Variety	Dye Color	Distance from Release(m.)
6/4	11-1200	35	f	Ae. vittatus		Green	0
10/4	09-1030	35	f	Ae. aegypti	Dark	Green	0
10/4	09-1030	35	m	Ae. aegypti	Inter	Red	237
10/4	15-1600	36	f	Ae. vittatus		Green	0
10/4	09-1030	35	f	Ae. aegypti	Dark	Green	0
10/4	06-0730	44	f	Ae. Aegypti	Inter	Red	90

Description of indoor and outdoor populations of Aedes aegypti labelled dark and pale. W. Hausermann, interim report, April, 1972.

Introduction

The variability in colour variation of A. aegypti has led to the description of three colour forms or variations: A. aegypti formosus, A. aegypti aegypti and A. aegypti var. queenslandensis (Mattingly, 1957). A. aegypti formosus is the blackest and A. aegypti var queenslandensis the palest extreme of the species. In the area of Rabai numerous colourvariations between dark black and pale are found side by side. Pale and black individuals, however, exhibit different habitat preferences and seem to live separate from each other as distinct populations. At a first glance pale to rather dark specimens (A. aegypti var. queenslandensis to dark A. aegypti aegypti are found together biting inside inhabited houses and breeding in man's water containers. Dark to pale (A. aegypti aegypti to A. aegypti var. queenslandensis) are found biting outdoors and under shelters and breed in outdoor habitats like treeholes, discarded tires etc.

Purpose

The questions asked are: what degree of isolation exists between these indoor and outdoor populations; what kind of isolating mechanism is operating and is there some genetice exchange between these two populations?

A first step to investigate this problem was to define members of these two populations by a few (as few as possible) distinct characters. Assuming, that during dry season the isolation between indoor and outdoor populations is the most rigid, 10 trial characters were selected from about 100 individuals from each population, collected as larvae from indoor and flooded outdoor habitats. The characters were scored on a 0 or 1 basis and are listed in table 1. After the first 200 individuals were scored the number of characters decisive for the distinction was narrowed down to the first 6 characters of table 6. The scaling pattern used by McClelland (1971) to describe population differences could not be used to distinguish between indoor and outdoor populations.

The validity of the established scoring system was tested on adults reared from ovitrap collections outdoors and additional larval collections from indoors made during dry season. The scoring along the guidelines in Table 1 permitted over time the correct assignment of over 100 specimens to their habitat without knowing their origin beforehand. Whether the same will be possible in rainy season remains to be tested.

Table 1 : The coloration characters by which pale and dark *A. aegypti* of the Rabai area can be distinguished.

Character description	Scoring 0	Scoring 1
1. Cuticle of scutum at the base prescutellar spot	black	non black
2. Cuticle of postnotum	black	non black
3. Sternopleuron and mesepimeron	black, dark grey or dark brown	red brown or pale
4. Dark scales of scutum	black	brown, gold or pale
5. Black scales of abdomen	shiny coal black	dull black
6. White scales on vertex and first abdominal tergite	white	cream coloured to yellowish
7. Erect scales of occiput	black	brown or yellow
Scale patterns		
8. First abdominal tergite	no white scales	white or pale scales
9. Vertex, decumbent white scales arranged	narrow, defined pattern	broad pattern, scattered
10. Pattern of abdominal tergites	minimum of white scales, basal white band on tergites only	white scales at posterior fringe of tergite and extending at posterior angles

Project 6: Dr. Paul T. McDonald
Interim Report

Production of translocation heterozygotes and selection of translocation homozygotes.

The induction of translocations has been achieved by the -ray irradiation of newly emerged males. Samples have been irradiated to 2000, 2500, 3000, 3500, and 4000 rads using a ^{60}Co Gammatron Unit at Nairobi. The irradiated mosquitoes were from colonies of field collected Aedes aegypti, either from an indoor or an outdoor population. The irradiated mosquitoes have been mated with a multiple-marker stock: rust eye on linkage group one, Silver mesonotum and spot abdomen on linkage group two and black-tarsi on linkage group three. Except for the semidominant Silver, all markers are recessive mutants. Females from the F_1 were single-pair crossed to the multiple-marker stock and selected progeny scored for the segregation of the markers.

Control crosses and low fertility experimental crosses have been analyzed for the segregation of markers of the three linkage groups of Aedes aegypti. Pseudolinkage between markers of different linkage groups indicates that a translocation had occurred involving break points near the markers. Only pseudolinkages of 0 to 20 cross over units have been saved, as these are most easily maintained. Other marker stocks will be used to isolate and maintain translocations with break points such that they would be difficult to maintain with this marker stock.

Thirty-nine translocations have been isolated from progeny of F_1 daughters of radiation-treated males. These translocations with their fertility and recombination data are listed in table 1. They include 11 involving linkage groups one and two, 8 involving linkage groups one and three, 18 involving linkage groups two and three, and two multiple translocations.

Translocations were isolated with treatments of all dosages of radiation but not with equal frequencies. Preliminary evidence (table 2) from experiments II and III indicates that a greater percentage of egg papers from F_1 females had low (less than 70 percent) hatches with higher dosages than with lower dosages. The recovery of detected translocations follows the same pattern, with the higher dosages accounting for higher percentage recovery.

Translocations have been maintained in the multiple-marker stock by selecting out individuals with wild type genes for the markers involved in the pseudolinkage. Each translocation is maintained, characterized by a given phenotype and generally a particular degree of sterility.

In Aedes aegypti, sex is determined by a single gene, with Mm being the male genotype and mm, the female genotype. As all non-autosomal translocations are linked to the m gene it is possible to test all translocations for homozygosis by crossing two heterozygotes together as soon as there are sufficient numbers of heterozygotes. This can occur when the translocation is first isolated. In the case of all translocations involving pseudolinkage of

linkage group two markers, the translocation is also marked with the wild allele for Silver mesonotum. Only individuals genetically detected as homozygous for the wild allele of Silver need be tested as possible homozygotes for the translocation, as the Silver locus maps quite close (less than 10 cross over units) to the spot locus and translocation break point, in all these translocations. In the case of translocations not involving linkage group two, the translocation homozygote and heterozygote would have similar phenotypes, and several must be tested in hopes that some would reveal themselves as homozygotes.

Homozygotes would be detected as they produce only translocation heterozygotes when mated with the normal multiple-marker stock. Seven translocation stocks have been studied for the production and isolation of homozygotes. Only three produced possible homozygotes. When analysis was made for homozygosis, none were apparent (table 3).

Table 1

Translocation	Hatch %	Linkage (cross over units)			No. progeny examined
		$ru^+(I)-s^+(II)$	$ru^+(I)-blc^+(III)$	$s^+(II)-blc^+(III)$	
Control	98	50	45	45	55
Control	99	53	47	51	49
Control	93	45	43	43	52
1 : 2 o	47	0	63	63	15
1 : 2 f	37	0	63	63	19
1 : 2 j	43	0	62	62	8
1 : 2 n	58	0	62	62	13
1 : 2 o	27	0	64	64	11
1 : 2 g	43	3	52	55	31
1 : 2 m	27	8	42	25	12
1 : 2 a	45	15	46	54	23
1 : 2 p	58	18	50	62	8
1 : 2 k	47	19	43	57	21
1 : 2 i	46	20	60	40	5
1 : 3 h	52	50	0	50	6
1 : 3 i	64	50	0	50	4
1 : 3 k	65	33	0	33	12
1 : 3 d	70	56	1	55	85
1 : 3 e	54	32	4	28	53
1 : 3 f	67	58	6	62	16
1 : 3 g	48	33	7	27	15
1 : 3 i	46	60	12	64	25
2 : 3 a	64	42	42	0	53
2 : 3 d	64	63	63	0	27
2 : 3 e	50	46	46	0	43
2 : 3 m	53	40	40	0	5
2 : 3 q	23	53	53	0	10
2 : 3 r	46	50	50	0	24
2 : 3 t	43	30	30	0	10
2 : 3 g	59	40	40	2	62
2 : 3 j	57	56	62	6	16
2 : 3 b	51	57	61	11	23
2 : 3 l	61	44	33	11	9
2 : 3 u	27	44	33	11	9
2 : 3 v	56	45	39	13	31
2 : 3 p	58	45	45	14	29
2 : 3 n	69	45	47	16	31
2 : 3 o	49	44	43	18	95
2 : 3 o	48	53	49	18	57
2 : 3 f	29	30	30	20	30
M a	35	0	0	0	16
M b	29	35	18	18	17

Table 2

Dosage	Egg Papers (E.P.)	Low Hatch (70 %) (L.H.)	Translocations recovered (T.R.)	L.H./E.P.	T.R.
I					
Control	9	0	0		
2000 r	27	3	2		
2500 r	28	6	3		
3500 r	28	8	3		
II					
Control	36	1	0		
2000 r	164	26	6		
3000 r	137	33	9		
4000 r	98	24	8		
III					
Control	8	1	0		
2000 r	56	12	2		
3000 r	39	8	3		
4000 r	45	10	3		
II + III					
Control	44	2	0	.05	
2000 r	220	38	8	.17	
3000 r	176	41	12	.23	
4000 r	143	34	11	.24	

Table 3

F_1 of Heterozygote x Heterozygote

Translocation	Heterozygote Phenotype in F_1	Homozygote Phenotype in F_1	Number F_1 tested	Number of tested without progeny	Number of tested with all heterozygote progeny	Number of tested with heterozygote and normal progeny
1 : 2 c	43	0	0			
1 : 3 d	—	—	10	7	0	3
1 : 3 e	—	—	9	7	0	2
2 : 3 a	31	0	0			
2 : 3 c	22	0	0			
2 : 3 e	22	1	1	1	0	0
4 a	5	0	0			

Mosquito strains maintained at the Mosquito Biology Unit of ICIPE, P.O. Box 80804, M o m b a s a / Kenya

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Aedes aegypti "dark" form:

Strain : MNAZI
Origin : Eggs collected with dirt samples from steps in coconut palms (MNAZI) in Rabai Location in August 1971.

Strain : TREEHOLE
Origin : Eggs collected with dirt sample from large treehole in mango tree in Mazeras (Rabai Location) in August 1971.

Aedes aegypti "pale" form:

Strain : GANGA
Origin : Adults caught in landing-biting catch in various houses of the village Ganga (indoor-landing-biting catch) in October 1971. Rabai Location.

Strain : XIPEVU
Origin : Adults collected in landing-biting catch inside various houses of the village Kipevu (next to Ganga) in October 1971. Rabai Location.

Strain : KIKOMANI
Origin : Collected larvae from water container in house of Josef Kai Hangari near Kaloleni on 5 Noverber 1971.

Strain : MITANGONI
Origin : Larvae collected from water container in house at Mitangoni (Road from Kaloleni to Milifi, clost to tar road). 5 November 1971.

Strain : MBALAMWENI
Origin : Collected as larvae from water containers in houses at Mbalamweni on 22 November 1971; Mbalamweni is along the road from Mariakani to Kaloleni at western border of coastal belt.

Strain : MIHI NGONI
Origin : About 30 larvae collected from indoor containers in 3 houses near Kaloleni on 31 March 1972, before rainy season.

Strain : GALANEMA
Origin : Collected in indoor water containers in 2 houses on 31 March 1972, before long rains. About 30 larvae.

Strain : PUNGU ZIWANI
Origin : Larvae collected in 6 houses in domestic containers before long rains. 20 April 1972.

Strain : MAZERAS
Origin : Larvae collected from water container in the house of James Rimba's uncle in Mazeras, August 1971.

Aedes aegypti "Type" strains

Strain : KIBAONI
Origin : Larvae collected from rainwater drum outside a house in Kibaoni, next to tar road Kilifi-Mombasa, near Vipingo. 5 November 1971.

Strain : MKWAJA
Origin : Larvae collected from rainwater drums in Mkwaja village (Tanzania coast) in backyards of several houses. 22 April 1971.

Strain : MIKOCHENI
Origin : Larvae collected from large pot with rainwater in roofless house being under construction. Mikocheni village is about 2 miles north of Mkwaja. 22 April 1971.

Strain : MATARAVE
Origin : Larvae collected from various natural breeding places at Matarave.

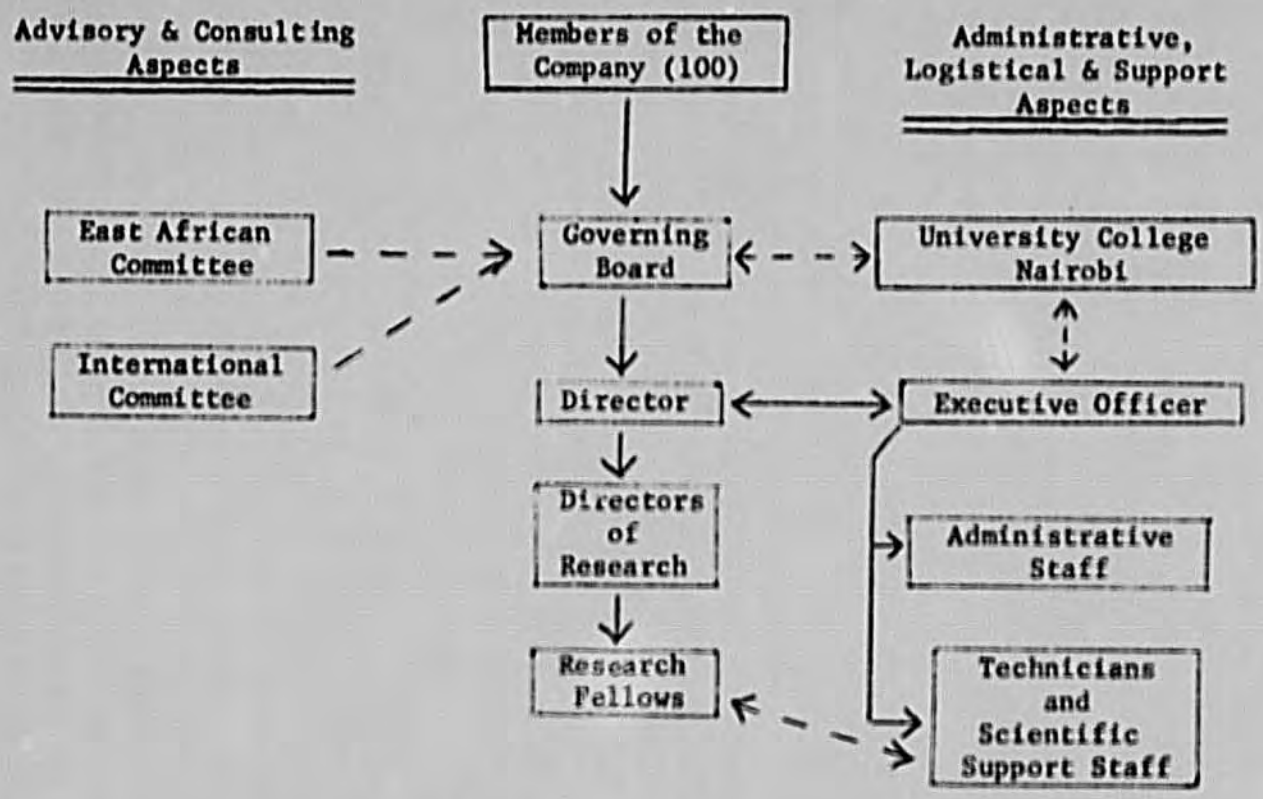
Aedes vittatus

Strain : KOMBENI
Origin : Larvae collected from rockpools along Kombeni river from October 1971 - March 1972.

Toxorhynchites brevipalpis

Strain : BEJUMA
Origin : Larvae collected in Bejuma forest from treeholes between March - April 1972.

ORGANIZATION OF ICIPE, 1972



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D. Wasawo, U. College, Nairobi, Kenya
T. Odhiambo, Director, ICIPE, Kenya

U.S. Joint Committee (NAS & AAAS):

Carroll Wilson, MIT, Chairman
H. Brown, Foreign Sec., NAS
G. Craig, Notre Dame
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D. Farner, U. Washington
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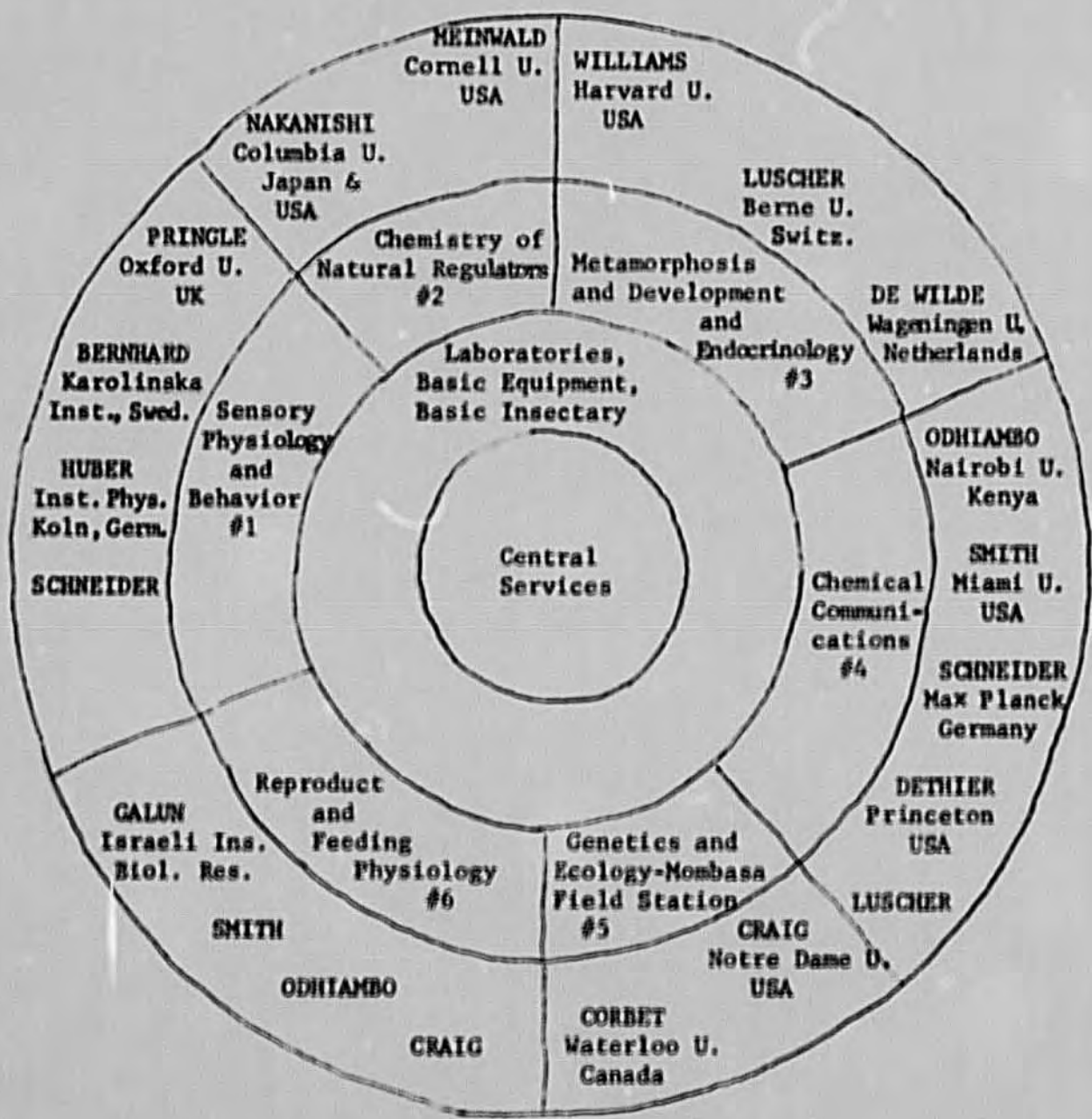
Roger Revelle, Harvard U.
Max Tischler, Wesleyan U.
Murray Todd, NAS
John Voss, Amer. Acad. Arts & Sciences
Carroll Williams, Harvard
Ruth Adams, U. Chicago & AAAS

ICIPE - 1972
International Centre for Insect Physiology and Ecology
RESEARCH PROGRAMS

Research Directors indicated in outside circle.



Major Emphasis:
Tsetse Fly, Glossina
Ticks, Ornithodoros
Army Worm, Noctuidae
Termites, Isoptera
Mosquito, Aedes

Additional Research Directors that do not yet have active programs:
BROWNING - Adelaide U., Australia
HELLE - U. Amsterdam, Netherlands
KULLENBERG - U. Upsala, Sweden
BARBIER - Nat. Prod. Inst., France
WEIS-FOGH - Cambridge U., England

ICIPE, 1972

Organizations Making a Financial Contribution

1. Alfred P. Sloan Foundation, USA
2. Research Corporation of New York, USA
3. National Academy of Sciences, USA
4. American Academy of Arts and Sciences, USA
5. Little Family Foundation, USA
6. National Science Foundation, USA
7. Allen Foundation, USA
8. Commonwealth Fund, UK
9. John Muir Institute of Environmental Studies, USA
10. United States Agency for International Development
11. University of Wageningen, Netherlands
12. University of Amsterdam, Netherlands
13. Royal Swedish Academy of Sciences
14. The Rockefeller Foundation, USA
15. Max Planck Society, Fed. Rep. Germany
16. The Volkswagen Foundation, Fed. Rep. Germany
17. The United Nations Development Programme
18. Swedish International Development Agency
19. The Ford Foundation, USA

Report on: Trip to University of Notre DameMosquito Biology Unit, Mombasa, Kenya

From: K. S. Rai, Principal Investigator Contract No. AID/csd - 3159

1. The trip was made during March 25-April 11, 1972. The purpose of the trip was to supervise the work of our Mombasa field unit, to make observations on the Aedes aegypti breeding characteristics in Kenya villages and on the extent of ecological isolation of breeding populations visavis the suitability for field releases of candidate genetic mechanisms and to hold discussions with our Unit personnel on our project needs and priorities.

2. I spent two days at the International Centre of Insect Physiology and Ecology in Nairobi discussing various scientific and administrative aspects of our project with Prof. Thomas Odhiambo and Mr. Ojal, the Director and Administrative Chief of ICIPE, respectively. Both were most cooperative.

3. In Mombasa, I spent considerable time making preliminary observations on several selected study sites, visiting some new ones and discussing various projects underway with the scientists in charge. All in all the personnel stationed in Mombasa - Drs. Hausermann, McDonald and Panara, are doing an excellent job. The Unit is off the ground and definitely running. The laboratory facilities for rearing and maintenance of various local strains and marker stocks etc. have been completed and the insectary is operational. Several field locations have been selected following preliminary screening by project personnel. In general, the breeding units of A. aegypti in Kenya are the small villages with no more than 10-15 houses in most cases. Water containers in the houses serve as A. aegypti breeding receptacles. These villages are quite small and appear to be fairly well isolated. Through the use of adult collections and analysis of oviposition jars placed in and around each selected village, the project staff are already accumulating considerable data on genetic composition of the A. aegypti populations breeding inside houses as well as those breeding outside in various types of bush habitats. The differences among these populations are both morphological and behavioral and well marked.

4. Dr. P. McDonald has put various genetic markers in the local Kenya strains, has used these African marker stocks to isolate 18 chromosomal translocations, and is attempting to get homozygotes for the same. He hopes to induce 200-300 translocations in the selected African stocks, isolate 6-10 homozygotes from among these translocations, evaluate their behavior in field population cages and finally make releases of various combinations of homozygotes in some selected villages.

In summary, all three scientists are working with dedication and gusto and they are already generating very useful information which should prove of great value not only for our future field releases but also provide much needed information on various aspects of field ecology and population dynamics of natural populations of A. aegypti.

5. Extensive discussions were also held with the three scientists on all aspects of the laboratory and field studies both underway and planned. Considerable emphasis was placed in these discussions on ways of optimising data output. A few new approaches were suggested to them, particularly on the use of mutagen-induced sterility as a tag for obtaining information on reproductive potential of a population subjected to various levels of sterility and/or control.

6. The need for a portable irradiation source in Mombasa is crucial. Currently, Dr. McDonald has to travel more than 500 miles to Nairobi (return trip) to irradiate mosquitoes for his translocations. Difficulties of not being able to get ready access to the source when he needs it were most obvious. The long trips result in high mosquito mortality. Besides, in order to undertake releases of radiation-sterilized mosquitoes for several studies, ready availability of a radiation source at our Unit will be indispensible. Following discussions with Prof. Odhiambo, I instructed the project leader to contact the International Atomic Energy Agency in Vienna for the loan or gift of a radiation source. Since formal requests to IAEA have to go through national governments, Prof. Odhiambo indicated his willingness to submit a formal request to IAEA if the preliminary enquiry revealed that IAEA could provide a source. If the preliminary enquiry from IAEA yields negative results, we must buy a portable source from the AID Contract funds.

7. Several ways of making reprints of current literature available to our staff in Mombasa were discussed.

8. Following intensive discussion with the staff in Mombasa, the following recommendations are made:

a. It was obvious that our rental subsidies to them are relatively low and not in tune with what housing actually costs in Mombasa. Two of the scientists live in Likoni where the housing is considerably cheaper than what it is on the island where the Unit is located. They are forced to take a ferry from and to work. This takes a minimum of one hour each way. They are paying $150.00 per month in Likoni where as a similar house on the island where the Research Unit is located cannot be rented for anything less than $200-250. I recommend that we increase their housing allowance to at least $150 per month (now it is $100) and ask them to live on the island. This move will improve both their productivity and their morale. They would be able to return to the lab in the evenings and the weekends. They have expressed a desire to be near their place of work and I feel we must increase their rent by at least $50 per month in order to partially meet the increased housing cost that this move will entail.

b. We should develop a system whereby any scientist that we may be interested in rehiring for an additional period beyond the initial two year appointment, gets a reasonable 1-2 month home leave with transportation paid for. Employees of international agencies such as the World Health Organization, International Atomic Energy Agency and of the U.S. Government abroad have that privilege. Again, this way we would be able to retain bright individuals or hire top quality personnel with this inducement.

Conclusions:

From personal observations and detailed discussions, it was obvious that we may have an ideal opportunity to study several ecological and genetic parameters of small, isolated, natural populations of a very important disease vector, *Aedes aegypti*, and to apply this knowledge to determine feasibility of genetic control of this species. Certain recommendations are made to enhance the scientific productivity and morale of our Unit in Mombasa. We have an excellent team of scientists - Hausermann, McDonald and Fanara, who are and will continue to make important contributions to the project. Such short term trips by outside consultants are vital in terms of stimulating our scientific staff in Mombasa, exchanging information with them and to occasionally review the progress made and delineate the prospects.