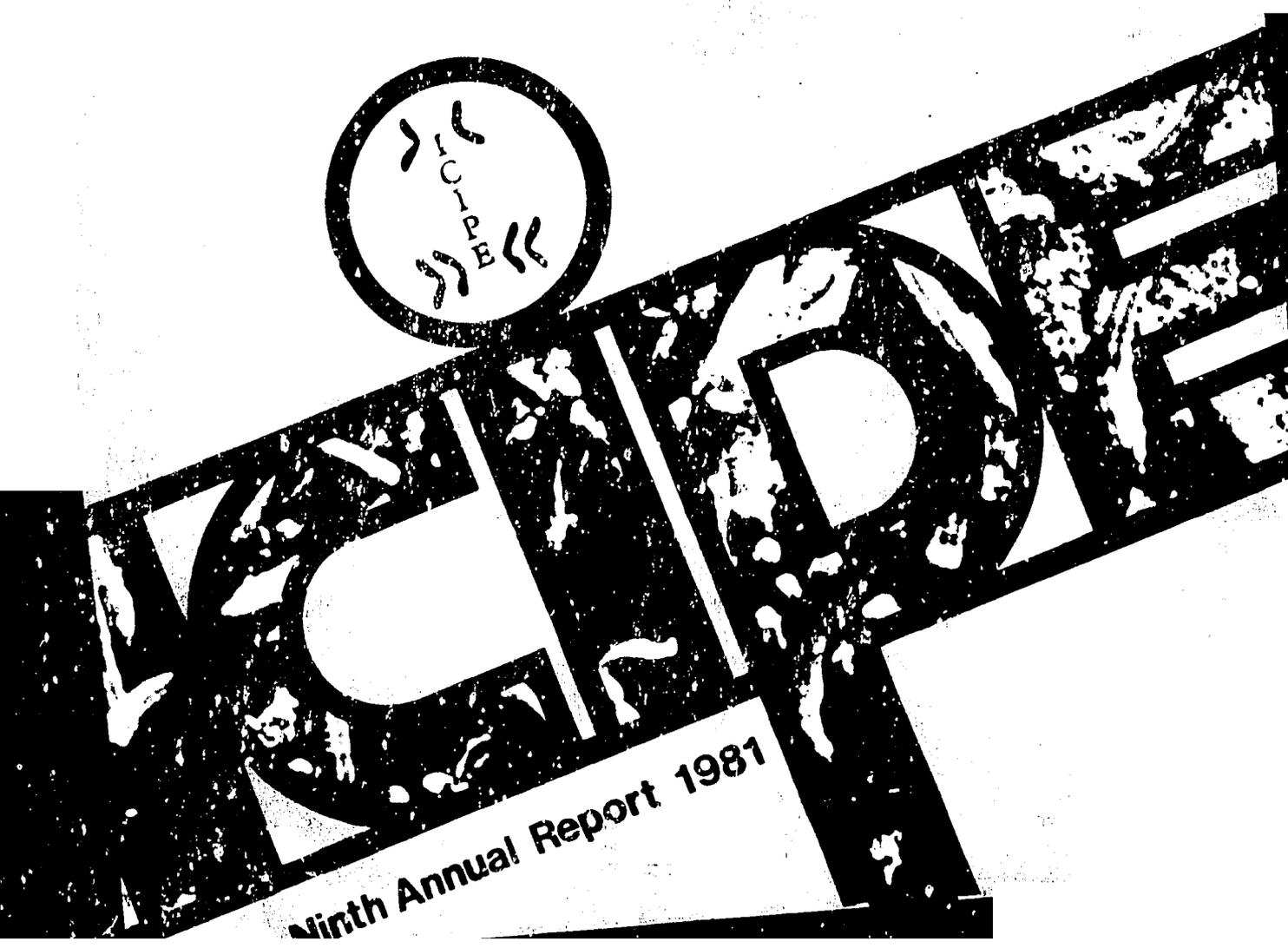
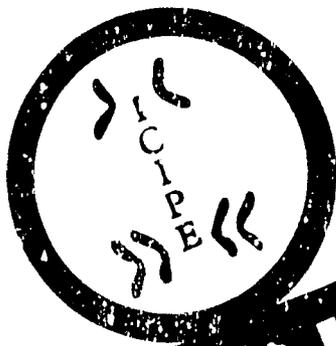


PN-ABB-632

THE INTERNATIONAL CENTRE OF  
INSECT PHYSIOLOGY AND ECOLOGY



Ninth Annual Report 1981

## **About ICIPE**

The International Centre of Insect Physiology and Ecology (ICIPE) was formally established in April 1970 in Nairobi, Kenya. The mandate of the ICIPE is to contribute to increased food production by undertaking mission-oriented research on pests of major crops, vectors of important livestock diseases, and insect carriers of human diseases critical to tropical rural health; and secondly, to increase the capacity of developing countries in pest management research and the application of the results of this research by training selected scientists and technologists in these fields. The Centre has been functional for ten years and is now an established institution contributing vital inputs into the global strategy to increase food production.

**THE INTERNATIONAL CENTRE OF  
INSECT PHYSIOLOGY AND ECOLOGY**

**NINTH ANNUAL REPORT  
1981**

**Nairobi, October 1982**

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# ICIPE GOVERNING BOARD, MARCH 1981

## 1982 RETIREMENT CLASS (April 1982)

- . Professor R. Galun 1979 (C, I)
- . Professor M. Kassas 1979 (C, I)
- . Professor Kenneth Prewitt 1979 (C, I)
- . Professor A. Semb-Johansson 1979 (F, I)
- . Dr. F. J. Wang'ati 1979 (K, I)

## 1983 RETIREMENT CLASS (April 1983)

- . Professor H. Brooks 1977 (F, II)
- . Professor Guy Camus 1977 (C, II)
- . Mr. Peter Nderu 1977 (K, II)
- . Dr. T. K. Arap Siongok 1977 (K, II)
- . Dr. O. M. Solandt 1977 (C, II)

## 1984 RETIREMENT CLASS (April 1984)

- . Professor L. K. H. Goma 1981 (C, I)  
(Succeeded Dr. H. Idris after the latter had completed only one term)
- . Dr. Peter T. Haskell 1981 (F, I)  
(Succeeded Prof. J. W. S. Pringle after his completion of 2nd term)
- . Professor John H. Law 1981 (F, I)  
(Succeeded Prof. T. A. Taylor whose term ended in March 1980. Professor Law was not appointed until April 1981)
- . Mr. R. B. Stedman 1978 (C, II)

## 1985 RETIREMENT CLASS (April 1985)

- . Professor Dr. Heinrich C. Weltzien 1982 (C, I)  
(To succeed, in April 1982, Prof. Alf Jolmels, whose 2nd term should have ended in April 1983. If Article 32 is

changed, Dr. Weltzien's  
first term will end in  
April 1985)

**NOTES:** C = Company nominee  
F = ICIPE Foundation nominee  
K = Kenya Government nominee

Each term last 3 years; I, first  
term II, second term

# SCIENTIFIC AND TECHNICAL STAFF- RESEARCH PROGRAMMES

## BASES OF PLANT RESISTANCE TO INSECT ATTACK

Dr. Z. T. Dabrowski, Programme  
Leader  
Dr. R. C. Saxena, Senior Research  
Scientist  
Dr. R. S. Ochieng', Research Scientist  
Mr. G. T. Masina, Graduate Research  
Scholar  
Dr. C. McFoy, Postdoctoral Research  
Fellow  
Mr. E. O. Nyangiri, Senior Technician  
Mr. S. H. Okech, Senior Technician  
Mr. F. O. Onyango, Technician  
Miss A. Ragot, Technical Assistant  
Mr. E. L. Kidiavai, Junior Technician  
Mr. E. O. Omolo, Agronomist  
Mr. M. D. O. Bungu, Junior Technician  
Mr. E. O. Arigi, Technician  
Mr. P. E. W. Njoroge, Junior Technician

## CROP BORERS RESEARCH PROGRAMME

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Leader  
Dr. K. V. Seshu Reddy, Senior Research  
Scientist  
Dr. G. C. Unnithan, Research Scientist  
Mr. A. G. L. Delobel, Research Scientist  
Mr. K. Ogwaro, Research Scientist  
Mr. J. B. Okeyo-Owuor, Associate  
Scientific Officer  
Mr. J. J. Njokah, Associate Scientific  
Officer  
Mr. J. C. Olela, Principal Technician  
Mr. K. E. Kidega, Senior Technician  
Mr. S. M. Othieno, Technician  
Mr. G. O. Anala, Technical Assistant  
Mr. C. O. Jacomito-Simbi, Technician  
Mr. Philomen O. Agwaro, Junior  
Technician  
Mr. J. G. Kibuka, Technical Assistant  
Mr. M. Lubega, Technician  
Mr. D. B. Mathenge, Technician  
Dr. B. Amoako-Atta, Research Asso-  
ciate  
Dr. A. K. Raina, Research Scientist

Dr. Dang Thahn Ho, Postdoctoral Research Fellow  
Dr. J. B. Suh, Postdoctoral Research Fellow  
Mr. S. O. Paye, Junior Technician  
Mr. P. O. Ollimo, Technician  
Mr. M. O. Arwa, Technical Assistant  
Mr. P. W. Agina, Technical Assistant

#### TSFTSE RESEARCH PROGRAMME

Dr, A. Challier, Programme Leader  
Dr. L. H. Otieno, Senior Research Scientist  
Dr. M. F. B. Chaudhury, Senior Research Scientist  
Dr. M.B.A. Nyindo, Senior Research Scientist  
Dr. T. K. Golder, Research Scientist  
Dr. F. Snow, Senior Research Scientist  
Dr. D. A. Turner, Senior Research Scientist  
Dr. (Mrs). M. S. Ramasamy, Research Scientist  
Mrs. M. Vundla, Scientific Officer  
Mrs. M. Owaga, Scientific Officer  
Mr. T. S. Dhadialla, Scientific Officer  
Mr. Erasmus I. Onweuzo, Graduate Research Scholar  
Mr. J. Kawooya, Postgraduate Trainee  
Miss N. F. Darji, Research Assistant  
Mrs. R. W. Kunyiha, Research Assistant  
Mr. E. Mpanga, Technician  
Miss R. Chesang, Junior Technician  
Mr. D. Uvyu, Junior Technician  
Mr. D. K. Mungai, Technical Assistant/  
Driver  
Mr. R. Mutuaruhiu, Junior Technician  
Mr. J. Likhanga, Junior Technician  
Mr. J. M. Muchiri, Technical Assistant/  
Driver  
Mr. J. A. Makau, Technical Assistant  
Dr. G. P. Kaaya, Research Scientist  
Mr. J. K. Kiilu, Technical Assistant  
Mr. P. O. Agutu, Senior Technician  
Mr. J. Apale, Technician  
Mr. F. Mkunza, Junior Technician  
Mr. James A. Atema, Junior Technician

Mr. D. M. Omogo, Technical Assistant  
Mr. F. M. Kyai, Technical Assistant  
Mr. C. M. Mutero, Postgraduate Research Scholar  
Mr. P. Amuttala, Technical Assistant/  
Driver  
Dr. J. O. Olobo, Associate Scientific  
Officer  
Miss S. A. Tarimo, Graduate Research  
Scholar

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Dr. G. W. Oloo, Research Scientist  
Dr. B. M. Okot-Kotber, Research  
Scientist  
Dr. T. O. Oloya, Postdoctoral Research  
Fellow  
Dr. D. A. Ruyooka, Research Scientist  
Dr. Robert Sieber, Research Scientist  
Dr. T. Abe, Research Associate  
Mr. G. N. H. Nyamasyo, Graduate  
Research Scholar  
Mr. L. Busharizi, Senior Technician  
Mrs. N. M. Baraza, Technician  
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Mr. P. O. Amoke, Technical Assistant  
Mr. E. Nyandat, Junior Technician  
Mr. P. Muteria, Technical Assistant  
Miss G. M. Wanjiru, Technical Assistant

#### AFRICAN ARMYWORM RESEARCH PROGRAMME

Dr. D. J. Rose, Honorary Programme  
Leader  
Mr. B. O. Otindo, Associate Scientific  
Officer  
Mr. J. G. Yarro, Graduate Research  
Scholar

## SCIENTIFIC AND TECHNICAL STAFF- RESEARCH UNITS

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Dr. D. L. Whitehead, Senior Research  
Scientist

## LIVESTOCK TICKS RESEARCH PROGRAMME

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Leader  
Dr. R. M. Newson, Senior Research  
Scientist  
Dr. F. D. Obenchain, Research Scientist  
Dr. (Mrs.) C. K. A. Mango, Research  
Scientist  
Mr. D. Punyua, Scientific Officer  
Dr. (Miss) G. M. Binta, Associate Scien-  
tific Officer  
Mr. J. Kilori, Technician  
Mr. A. Mongi, Postgraduate Trainee  
Mr. A. Chiera, Research Assistant  
Mr. J. Ojowa, Junior Technician  
Mr. G. T. Thuo, Technical Assistant  
Mr. J. Mugane, Technical Assistant  
Mr. G. M. Hindi, Subordinate Assistant  
Mr. K. C. Wainaina, Subordinate  
Assistant  
Mr. A. Bwire, Technician

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Dr. W. A. Otieno, Research Scientist  
Dr. F. W. Mosha, Research Scientist  
Dr. M. J. Mutinga, Senior Research  
Scientist  
Dr. J. B. Kaddu, Research Scientist  
Dr. Maurice Odindo, Postdoctoral Re-  
search Fellow  
Mrs. Lucy M. Rogo, Graduate Research  
Scholar  
Miss D. Sabwa, Graduate Research  
Scholar  
Miss Lucy Irungu, Graduate Research  
Scholar  
Mr. P. Mwanisi, Junior Technician  
Mr. S. Muti, Junior Technician  
Mr. E. Mkuzi, Technical Assistant  
Mr. J. Mwandandu, Technical Assistant  
Mr. J. A. Otieno, Junior Technician  
Mr. M. C. Ngumah, Junior Technician  
Mr. R. C. Odhiambo, Junior Technician  
Mr. M. P. Nyamori, Senior Technician

Dr. D. A. Otieno, Research Scientist  
Dr P. G. McDowell, Research Scientist  
Mr. M. I. Jondiko, Graduate Research  
Scholar  
Mr. A. Chapya, Chief Technician  
Dr. A. Hassanali, Senior Research  
Scientist  
Mr. W. Lwande, Graduate Scholar  
Mr. C. S. Thomas, Graduate Research  
Scholar  
Mr. E. O. Osir, Graduate Research  
Scholar  
Mr. N. Juma, Technician

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Mr. P. Lisamulla, Principal Technician  
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Research Fellow

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Mr. P. G. Njagi, Graduate Research  
Scholar  
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Mr. P. Ahuya, Technical Assistant  
Mr. S. K. Kirui, Graduate Research  
Scholar

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Mr. G. Achieng', Technical Assistant  
Mr. L. Moreka, Technical Assistant  
Dr. T. Gebreyesus, Research Scientist  
(on leave of absence)

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Mr. S. M. Mbugua, Technical Assistant  
Mr. B. O. S. Ogal, Technical Assistant  
Mr. R. Okello, Technical Assistant  
Mr. G. M. Ng'ang'a, Technical Assistant

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Mrs. S. W. Mwanycky, Associate Editor

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Mr. D. Odhiambo Aoko, Messenger  
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Mr. S. K. Langat, Driver

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Mr. D. Kigera, Librarian  
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Mrs. L. Kimani, Accounts Assistant  
Mr. F. Otieno, Messenger

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Mr. S. M. Kimaita, Administrative  
Officer  
Mrs. A. Okumali, Senior Secretary  
Mrs. P. Owitti, Secretary  
Mrs. T. Lohay, Secretary  
Mrs. M. Antao, Secretary  
Mrs. G. Wasike, Secretary  
Mrs. R. Okoth, Assistant Secretary  
Miss R. Vugaba, Secretary

Mrs. G. Weya, Senior Telephonist/  
Receptionist  
Mr. R. Nyaridi, Messenger  
Mr. L. Kisutia, Cleaner  
Miss S. Christopher, Cleaner/Tea Maker  
Mr. C. Otieno, Messenger  
Miss E. Afandi, Assistant Secretary

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Security Services  
Mr. J. Atiche, Security Officer  
Mr. S. Akhaya, Assistant Janitor  
Mr. S. Mitalo, Watchman  
Mr. L. Okong'o, Watchman  
Mr. W. W. Achiroma, Watchman  
Mr. E. K. Shilengo, Watchman  
Mr. A. M. Ouma, Watchman  
Mr. L. L. Ayekha, Gardening Assistant  
Mr. J. Elegwa, Gardening Assistant  
Mr. A. M. Bubusi, Cleaner  
Mr. D. Chege, Cleaner  
Mr. E. Asami, Cleaner  
Mr. J. A. Onyango, Watchman  
Mr. Z. O. Nyandere, Watchman  
Mr. D. O. Singe, Watchman  
Mr. A. O. Ogaja, Watchman  
Mr. J. K. Opere, Watchman  
Mr. M. M. Ogolla, Watchman  
Mr. A. N. Makori, Watchman  
Mr. J. D. Nyawalo, Watchman  
Mr. R. K. Milgo, Watchman  
Mr. A. Muyanda, Watchman  
Mr. C. O. Okello, Gardener

## TECHNICAL SUPPORT SERVICES STAFF

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Technical Services  
Mr. A. Lobo, Electronics Engineer  
Mr. P. Nyachio, Senior Technician  
Mr. S. Karanja, Senior Technician  
Mr. J. M. Maina, Technician  
Mr. J. N. Mtei, Technician  
Mr. H. Gichinga, Technician  
Mr. J. N. Musisi, Maintenance Engineer

Mr. J. O. Onyango, Plumber/Mason  
Mr. B. Omulo, Junior Technician  
Mr. K. Kibe, Technical Assistant  
Mr. J. Omondi, Technical Assistant  
Mr. P. Oluya, Junior Technician/Glass  
Blower  
Mr. E. N. Nyoike, Junior Technician/  
Glass Blower  
Mr. S. O. Obiero, Technician

#### TRANSPORT UNIT

Mr. A. D. Sheikh, Automobile Foreman  
Mr. J. O. Oduol, Senior Mechanic  
Mr. G. M. Kinyanjui, Driver  
Mr. E. W. Muchiri, Technical Assistant  
Driver  
Mr. F. Ombija, Senior Mechanic  
Mr. F. O. Hamala, Mechanic/Driver  
Mr. B. Oyondi, Driver  
Mr. J. K. Maina, Technical Assistant/  
Driver  
Mr. R. B. Gathu, Technical Assistant/  
Driver  
Mr. P. Mahogo, Senior Driver  
Miss E. N. Mwangi, Driver  
Mr. J. G. Mahinda, Driver  
Mr. A. Orina, Mechanic/Driver

#### LABORATORY MANAGEMENT

Mr. E. E. Muro, Engineer & Controller  
for Laboratory Services

## FIELD STATIONS

#### MBITA POINT FIELD STATION

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tive Officer  
Mr. B. S. K. Masyanga, Farm Controller  
Mr. F. K. Ongola, Assistant Accountant  
Mr. J. O. Angado, Maintenance Engineer  
Mrs. M. N. Okach, Assistant Secretary  
Mr. H. Purcell, Project Manager  
Mr. E. Sonye, Watchman  
Mr. S. O. Aol, Watchman  
Mr. D. Oyoto, Watchman  
Mr. G. N. Harshe, Site Engineer II  
Mr. R. C. Joshi, Site Engineer I

Mr. J. O. Ohato, Mechanic/Driver  
Mr. P. Mbuya, Driver  
Mr. B. Mogendi, Watchman  
Mr. J. N. Asanyo, Mechanic/Driver  
Mr. P. O. Ouma, Farm Assistant  
Mr. N. M. Sangura, Farm Assistant  
Mr. P. O. Auta, Farm Assistant  
Mr. J. Sagini, Farm Assistant  
Mr. Wolukau, Technician

**MBITA POINT INTERNATIONAL  
SCHOOL**

Mrs. P. A. Ogada, Principal  
Mr. F. O. Omolo, Teacher  
Mrs. C. O. Ndiege, Teacher  
Mrs. S. A. Omune, Cleaner

**KAJIADO FIELD STATION**

Mr. J. M. Ole Kobaai, Watchman

**COASTAL FIELD STATION –  
MOMBASA**

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Mrs. C. Rarieya, Assistant Secretary  
Mr. P. O. Ngugi, Senior Accounts Clerk  
Mr. J. B. Kariuki, Mechanical/Driver  
Mr. S. Abdalla, Watchman  
Mr. S. Wanjohi, Cleaner  
Mr. L. O. Odongo, Technical Assistant/  
Driver  
Mr. S. M. Kibati, Security Assistant  
Mr. E. M. Sowah, Driver

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GUEST CENTRE, RUARAKA**

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Mr. J. E. Mwangi, Head Cook  
Mr. A. I. Okapesi, Assistant Head Cook  
Mrs. J. A. Musiga, Housekeeper  
Mr. P. H. Opondo, Kitchen Assistant  
Mr. B. O. Randiga, Assistant Launder  
Mr. A. M. Mutwoli, Room Steward  
Miss S. M. Kagondu, Secretary  
Mrs. E. Kwach, Telephonist/Receptionist  
Mr. C. B. Oyieyo, Room Steward  
Mr. A. Lweya, Cook

Mr. H. M. Kibisu, Assistant Launder  
Mr. G. Gichuru, Kitchen Assistant  
Mr. A. E. Mulae, Room Steward  
Mr. Omutimu, Room Steward

**SENIOR MANAGEMENT STAFF**

Professor Thomas R. Odhiambo,  
Director  
Mr. J. M. Ojal, Manager for Communi-  
cation Systems  
Professor A. S. Tahori, Deputy Director  
(Research)  
Mr. L. Z. Mosha, Financial Manager

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## SCIENTIFIC DEVELOPMENT: THE TRIAL OF THE ICIPE COMMUNITY

Last year, in commenting on the Centre's Scientific Development: Planning for the Future, I was able to confidently state that, "... our preeminent goal in the first decade of ICIPE's existence has now been accomplished — that of establishing a critical mass of talented, highly motivated, interacting scientific community at the ICIPE all cooperatively targeted on a few, carefully selected pest management goals, collaborating productively with the wider scientific and practitioner community in Africa and other tropical regions, and concentrating on the major pest problems of its constituency. The next phase for us now seems to be one of giving this ICIPE scientific community the means to accomplish its mandate on a continuing and rationalised basis." These sentiments could not have experienced a more searching test than what the Centre went through in the year 1981.

Stemming from the worldwide economic recession, the ICIPE experienced the most acute financial situation that it has ever undergone in its eleven years of existence. This situation tested the institute as a whole to the very core. It is a testament to the resilience of the entire ICIPE community that, in spite of the very reduced circumstances, the scientific work and its quality continued unabated. The pioneering spirit is virile, the staff morale is high, the Governing Board has exhibited an exceptional concern and responsibility, and the ICIPE donors have shown a tenacious faith in the ICIPE.

The traditional donors of the ICIPE have, since November 1980, established an informal platform, a sort of consortium, which enables them to review ICIPE's performance and progress, and the support it needs to accomplish its tasks, in a more regular manner than was possible to do before. This group — the SPONSORING GROUP FOR THE ICIPE (SGI) — presently consists of the United Nations Development Programme (UNDP), the Food and Agriculture Organization of the United Nations (FAO), the International Fund for International Development (IFAD), the World Bank, The OPEC Fund for International Development, the U. S. Agency for International Development (USAID), and the International Aid Agencies of Australia, Sweden, Norway, Denmark, the Netherlands, France, Switzerland, the Federal Republic of Germany, the United Kingdom, and Belgium, as well as the host country — Kenya. The SGI met formally in Paris (May 1981) and Washington, D. C. (November 1981) at which time it set in motion a detailed examination of the programme and needs of the ICIPE over the medium-term perspective. This would form a basis of a continuing scheme for the support of the Centre.

One important point that has emerged from a reappraisal of the ICIPE programme during the year 1981 is that the most crucial target of its research effort is the satisfaction of the resource-poor farmer in Africa and other tropical regions in his pest management needs. We are therefore reassessing all of our research activities to ensure that their goals will correspond closely with these needs, and that research training we are undertaking for Africa are such that it will strengthen the national programmes in this critical area.

Thomas R. Odhiambo  
Director, ICIPE

14th August 1982

## PROFILES

### MR. J. M. OJAL



Mr. J. M. Ojal, Manager for communication Systems, heads all the communication activities of the Centre that include Training, Library and Documentation, communication and Information,

Public Relations and Visitor Service. Mr. Ojal is by training, a geologist, a botanist and a geographer and has had a distinguished public service in Kenya, notably as a science teacher in a number of the reputable boys' schools in the country.

Mr. Ojal has been associated with the ICIPE since 1970 when he was a member of the ICIPE Company and Governing Board. He was then the Permanent Secretary in the Ministry of Natural Resources.

In March 1972, Mr. Ojal joined the staff of the ICIPE as the Centre's Chief Administrative Officer. His position was later changed to that of Deputy Director (Administration). When the position of Administrative Manager, Deputy Director (Research) and Financial Manager were established, Mr. Ojal became the Manager for Communication Systems.

### DR. ELIZABETH KOKWARO



Dr. E. Kokwaro heads the Histology and Fine Structure Research Unit at the ICIPE. She was educated at the University of Nairobi where she obtained her Ph.D. in Entomology; the topic

for her thesis being "Oocyte Development in the fleshfly, *Sarcophaga tibialis*". She later received advanced training in Fine Structure and Cytochemistry at the Karolinska Institutet in Stockholm, Sweden and at the Institut de Biologie Moleculaire in Paris.

Dr. Kokwaro is also involved in the teaching of insect developmental biology, histological techniques and electron microscopy. She is a member of a number of scientific societies including membership to the African Association of Insect Scientists.

### DR. MATHEW P. CUNNINGHAM



Dr. Cunningham attended the University of Glasgow Veterinary School from 1947 - 1952, where he qualified as a Member of the Royal College of Veterinary Surgeons. His first appointment was

in the University of Glasgow Veterinary School where he taught and conducted research on Bovine Paratuberculosis and Canine Leptaspirosis. He also participated in the development of a vaccine for cattle against *Dictyocaulus vivipara*.

From 1958 to 1967 he worked for the East African Trypanosomiasis Research Organization at Tororo, Uganda. Some notable contributions resulting from his work at EATRO include establishment of a trypanosome bank; immunological, physiological studies and control of tsetse; and treatment of trypanosomiasis.

As a Project Manager of the FAO/UNDP PROJECT ON Tick control in Kenya, he participated in the development of methods for immunization of cattle against ECF and identification of an ECF curative drug. He joined the ICIPE in 1977 as the

Programme Leader of the Livestock Ticks Programme

**DR. G.W. OLOO**



Dr. Oloo, Programme Co-ordinator, Insect Pathology and Pest Management Programme (IPPM), obtained his first degree from the Department of Entomology and Environmental Sciences, Rutgers

State University, USA in 1968. On his return, he served in the Entomology Section of the Ministry of Agriculture, Kenya. He carried out applied research on sugar-cane pests for a period of six years leading to his Ph.D. degree in 1973. He joined the Grassland Termite Programme at the ICIPE in 1974, where he worked on the role of chemical communication in the co-ordination of foraging behaviour. He has recently been appointed the programme coordinator of the newly established IPPM Programme.

**DR. R. S. PATHAK**



Dr. Pathak, Acting Programme Leader of the Crop Borers Research Programme joined the ICIPE in 1980. He obtained his Ph.D. degree in Genetics from the Punjab Agricultural University in

1968. He then joined the Haryana where he later headed the Cotton Breeding Section and taught in the Department of Plant Breeding.

From 1972 to 1980, Dr. Pathak worked in the Faculty of Agriculture, University of

Nairobi. He is credited with initiating the first Postgraduate Programme in Plant Breeding.

At the ICIPE Dr. Pathak has initiated research on Genetics of Host Plant Resistance to Insect Pests in cowpea, sorghum, maize and rice.

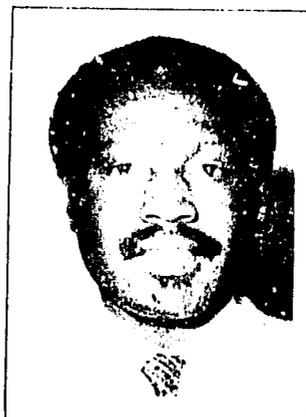
**DR. CORNELIS J. DEN OTTER**



Dr. Den Otter was born in Holland and received his M.Sc. (cum laude) in 1960 from the State University at Leiden, Holland and his Ph.D. in 1971 from the State University at Groningen,

Holland. From 1960 to 1966, Dr. Den Otter taught Animal Physiology at the Leiden State University and built up and directed the Sensory Physiology Research Unit. In 1966 he was appointed the Head of the Sensory Physiology Research Group at the Department of Zoology, State University at Groningen. In 1974 he spent a sabbatical year at the Max Planck-Institute fur Verhaltensphysiologie at Seewiesen (Federal Republic of Germany) at the Laboratory of Professor Dietrich Schneider. In January 1981, he was granted special leave to join the ICIPE as Head of the Sensory Physiology Research Unit.

**DR. LUKA O. ABE**



Dr. Abe was born in Uganda in 1945. He received his university education at Washington State University, where he obtained B.Sc. Agriculture, B.Sc. Entomology in 1971. He completed his Ph.D. stu-

dies at Rutgers University, New Jersey in the area of Plant Physiology and Biochemistry. Dr. Abe taught Biology at York College of the City University of New York and at Douglass College of New Jersey, before joining the University of Nairobi in 1974. In 1977, he joined the ICIPE, where he is currently the Head of the Training Unit. His special interest is now in institutional management with emphasis on scientific research institutions.

#### MR. ATASHILI MANDO



Mr. A. Mando, Principal Controller for Technical Services was born in Nigeria and received his early technical training in Cameroon and the University of Nigeria. From 1967 to 1971 he studied

Physics at the State Engineering College, Heilbronn in West Germany. For a period of two years he was attached to various technical firms in West Germany for practical training. In 1971, he joined the Phillips Forschungs Laboratorium, Aachen in West Germany where he worked as laboratory engineer investigating methods of improving the quality and intensity of high pressure gas discharge lamps and was later involved with the design and development of laboratory instruments. He joined the ICIPE in January 1973.

# ADMINISTRATION

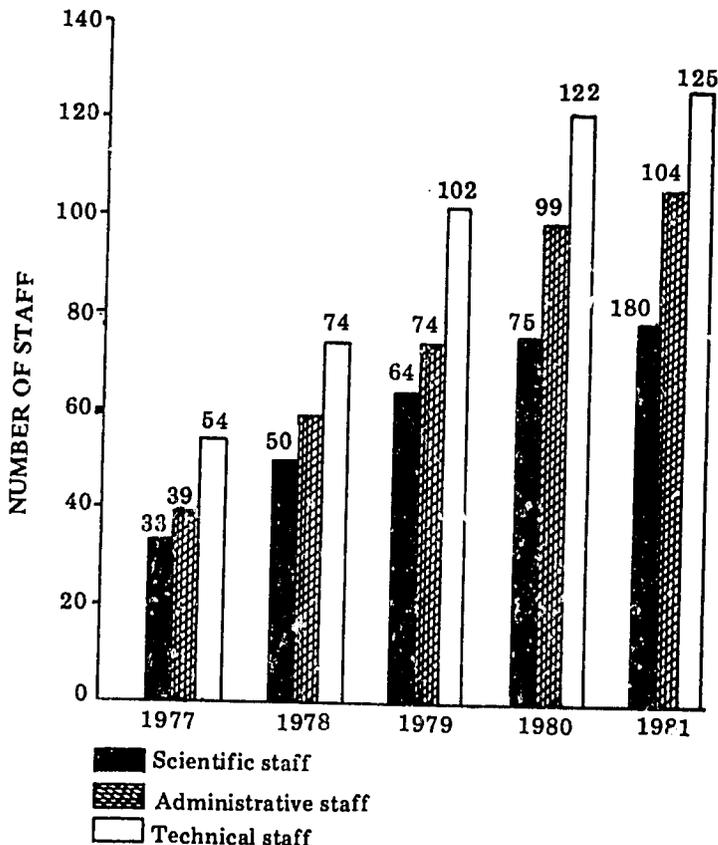
## STAFF GROWTH

ICIPE Staff has grown from 126 in January 1977 to 309 in December, 1981. Except for 1981 when the staff growth rate of 10.31% was recorded, the growth rate between 1977 and 1980 was 45% per annum on the average. The staff categories have been divided into Scientific, Technical and Administrative as shown in the following figure.

Scientific staff includes Postdoctoral Research Fellows, Scientific Research Officers, Associate Scientific Officers, Research

Assistants and Graduate Research Scholars. Administrative staff includes all staff in the Office of the Director, Communication and Training Division, Finance Division, Personnel and Office Management Division, Duduville International Guest Centre and Field Stations. Technical staff refers to those members of staff who render technical support to Scientific staff and includes Principal Technicians, Senior Technicians, Technicians, Junior Technicians, Technical Assistants and Technical Assistant/Drivers. Staff in the Workshops and Transport Unit loosely fall under this category.

STAFF GROWTH



# TRAINING AT THE ICIPE

## INTRODUCTION

Training continued along the same lines as in 1980. However, there were several new developments within the year which will now bring new dimensions into the ICIPE's approach to meet its goals of building capacity for mission-oriented research into pest management.

One of the major tasks of 1981 was continuation of review of the training programmes, which had been started in 1980. Part of this review resulted in the rationalised programmes which are reported below.

## TRAINING PROGRAMMES

### Staff development training

As part of its strategy to strengthen its capacity, the ICIPE has evolved several approaches to meet its institutional goals.

**Research Training.** Several members of the scientific staff were involved in research training at the ICIPE or overseas. At least fifteen such staff undertook training at institutions in Canada, Australia, Philippines, Sweden, Italy, United Kingdom, Japan, the United States of America, and the ICIPE itself.

**Technical Training.** As has been in the past, the ICIPE continued to support members of its technical staff in courses to upgrade their skills. This was done mainly at the Kenya Polytechnic, with one staff undertaking a course at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) at Hyderabad, India.

**Management Training.** As a means of sharpening managerial skills,

the ICIPE Management Staff also continued to receive advanced training in research management.

**Training in Communication Skills.** The Secretarial staff continued to train in advanced secretarial courses at the secretarial colleges in Nairobi.

### Pest Management Research Training

**Postdoctoral Research Fellowships.** At least 5 postdoctoral fellowships were supported in 1981 in the following programmes: Crop-Borers Research, Bases of Plant Resistance to Insect Attack, Grassland Termite Research, Histology and Fine Structure Research.

**The International Postgraduate Studies in Insect Science.** As a means of rationalizing the ICIPE's input in high-level postgraduate training in pest management, the Planning Workshop convened in Bellagio, Italy, recommended the establishment of African Regional Postgraduate Programme in Insect Science, ARPPIS. The ARPPIS is intended to be collaborative with African Universities; the ICIPE is to act as the managing agency and to offer coursework and research, while the universities are to award the degrees.

The programme is expected to start in January 1982; the first intake is to consist of up to ten postgraduate trainees from the participating universities.

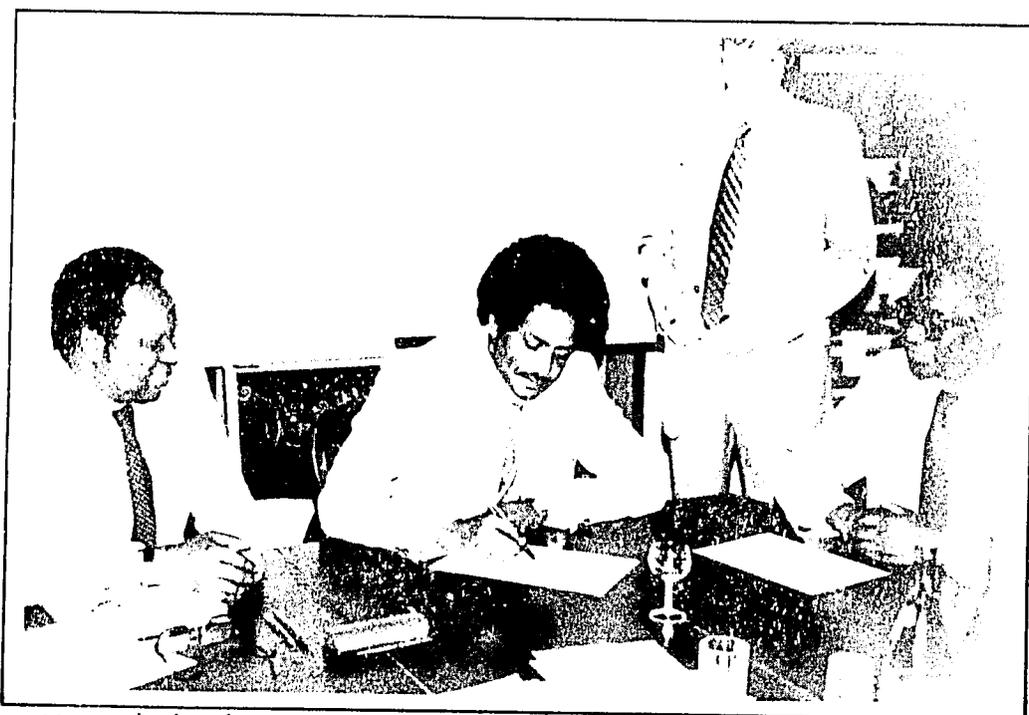
Under the ARPPIS, trainees are expected to undertake research in such areas as: ecology of arthropods of economic importance in

agriculture and rural health, insect endocrinology, developmental and pheromonal biology, parasitology, insect pathology, bases of plant resistance to insect attack, insect behaviour, morphology and anatomy, biochemistry and natural products chemistry, toxicology, insect nutrition and mass-rearing, development of specialized bioassay techniques etc.

The programme will consider and admit Postgraduates with M.Sc. or graduates with good first degrees in biological sciences, entomology, biochemistry, agriculture forestry, medicine or veterinary science.

International Study Workshop. The Planning Conference on the African Regional Postgraduate Programme in Insect Science (ARPPIS) held at the Rockefeller Foundation Conference and Study Centre, Villa Serbelloni, Bellagio,

Italy from 7 to 11 September 1981, was the major event in this category. This was a historic Conference and should form a very important milestone in the training programme. In all, 23 participants from the following institutions attended the Conference: the ICIPE, the University of Khartoum, the University of Ife, West African Rice Development Association (WARDA) University of Montreal, University of Ibadan, University of Malawi, Australia Development Assistance Bureau (ADAB), University of Ghana, University of Dar-es-Salaam, The Rockefeller Foundation, the UN Economic Commission for Africa (ECA), The Institute Senegalais de Recherches Agricoles (ISRA), the Danish International Development Agency (DANIDA), Makerere University and the United Nations Educational Scientific and Cultural Organization (UNESCO).



Professor Mutamad Ahmed Amin, Professor of Medicine, University of Khartoum and Director, Regional Ministry of Health and Social Welfare, Sudan signs the agreement for the establishment of the African Regional Postgraduate Programme in Insect Science (ARPPIS), reached at Bellagio, Italy. Witnessing are Professor Anthony Youdeowei (left), Professor Thomas R. Odhiambo (right) and Dr. Rodney C. Hills, who served as chairman of the Conference.

## TRAINING FOR PRACTITIONERS

The International Group Training Course on Components Essential for Ecological Sound Pest and Vector Management Systems.

The fifth course in this programme was held from 19 July to 7 August 1981. The United Nations Environment Programme, UNEP has con-



Dr. R. Subra (left, front) Programme Leader, the ICIPE Medical Vector Research Programme explains the transmission of filariasis in a pit latrine in a village at the Kenyan Coast.



Participants at the ICIPE/UNEP Group Training Course look into the problems of pest management in sugar cane plantation in Nyanza, Kenya.

tinued the co-sponsoring of this course. This year, the course attracted 32 participants from 18 countries: Brazil, Colombia, Ethiopia, Ghana, Kenya, Lesotho, Mozambique, Nigeria, Philippines, Saudi Arabia, Senegal, Somali, Sudan, Tanzania, Uganda, United Arab Emirates, Zambia and Zimbabwe. Since 1977 when the course was first held, 132 trainees have participated in this course.

The International Group Training Course in Insect Growth, Development and Behaviour: The first

course, in the series under the the theme "Insect Growth, Development and Behaviour", was held at the ICIPE from 10 to 21 August 1981. The purpose of the course is to provide an opportunity for young scientists who have already completed their academic training, and are at the beginning of their career in research or teaching, to learn new research techniques and advances in the area of insect physiology and developmental systems and behaviour. The first course, which concentrated on physiology and developmental systems



Practical demonstrations of laboratory techniques being conducted by Professor F. C. Kafatos (centre, seated) of the Harvard Biological Laboratories, U.S.A.

### *Training.*

was divided into lectures, practicals, tutorials and seminars.

A total of 17 trainees from 13 countries participated; the countries represented were: Ghana, Nigeria, Kuwait, Ethiopia, Mauritius, Malawi, Poland, India, Sierra Leone,

Kenya Uganda, Egypt and Vietnam. The course was co-sponsored by the International Cell Research Organization (ICRO), the UNESCO, the International Society for Developmental Biologists (ISDB) and the ICIPE.

## COMMUNICATION

The Communication Department continued to provide editorial, print, photographic and graphic art support for research, training and administrative activities of the ICIPE. These services culminated in a grand poster display of ICIPE activities during the laying of the foundation stone of the ICIPE's Mbita Point Research Station by His Excellency the President of Kenya, Hon. Daniel Arap Moi in April 1980.

### Seminars, Conferences and Study Workshops

Seminars, conferences and study workshops are regarded as ICIPE's main thrust for disseminating research information,

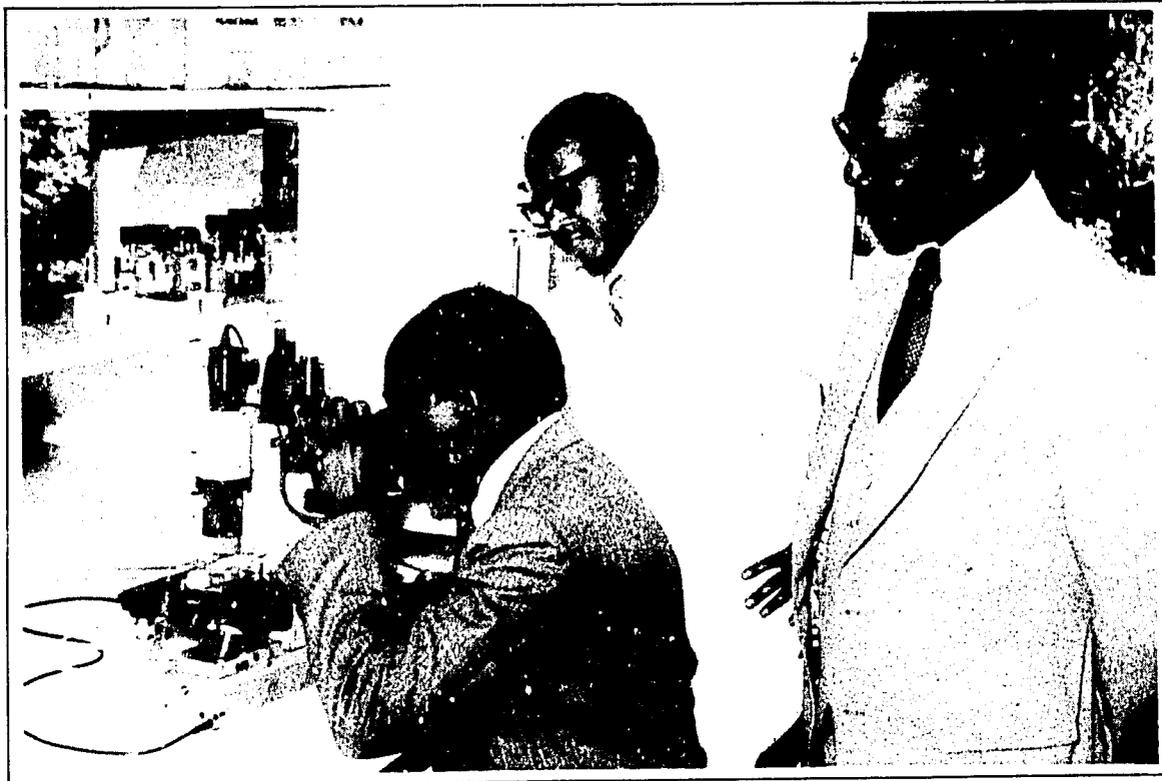
discussing new scientific advances, reviewing current research and planning future projects.

In 1981, a total of 41 institute-level seminars were presented at the Headquarters in Nairobi, at Mbita Point Field Station and at Muhaka Field Station. Over one third of these were presented by visiting scientists. Programme-level seminars or internal discussions continued to take place throughout the year in all ICIPE Research programmes and Research Units.

At the 11th Annual Research Conference, the Tsetse and Grassland Termite Research Programmes were reviewed in depth.



His excellency the President of Kenya, Hon. Daniel Arap Moi signing the visitors book at the end of his tour of the ICIPE's farm complex. Standing in the extreme left is Professor Thomas R. Odhiambo, Director of the ICIPE and at the centre is Hon. Alphonse Okuku, M. P. for Mbita.



The Vice President of Kenya, Hon. Mwai Kibaki seen looking at *Trypanosoma brucei*, the micro-organisms that cause Nagana in cattle under the microscope. Standing behind him, to his left is Professor Thomas R. Odhiambo, Director of the ICIPE and Dr. M. B. Nyindo (in white coat), a senior research scientist in the Tsetse Research Programme.



Dr. A. Hassanali (Bioassay Unit) is seen showing Dr. Bradford Morse, Administrator, UNDP, a synthetic sample of tsetse (*G. pallidipes*) sex pheromone synthesized at the ICIPE.

## Audio Visual Services

The ICIPE aims to improve her communication and outreach services by incorporating the use of relevant and advanced audio-visual material particularly in the training programmes, for visitor service, and for exhibition and demonstration in laboratories and in the field. In this regard, the department started the planning and the production of synchronized slide/tape presentations which are planned to cover all the ICIPE research projects.

## VISITORS SERVICE

The Communication Department received 800 visitors in 1981 at the ICIPE. These included His Excellency the President of Kenya Hon. Daniel Arap Moi who visited Mbita Point Research Station in April 1981. The Vice President Hon. Mwai Kibaki in August 1981, several cabinet Ministers, Senior Government officials, International and National Research Scientists, ICIPE donors, and college and school parties.



Hon. G. K. M'biijiwe, Kenya Minister for Agriculture seen holding jars containing armyworm larvae and pupae while he listens to Dr. D. J. Rose explaining the work going on in the African Armyworm Research Laboratory. In the centre is Professor Thomas R. Odhiambo, Director of the ICIPE and Dr. F. J. Wangati from the Ministry of Agriculture.

# LIBRARY/DOCUMENTATION SERVICES

## STAFF CHANGES

Mr. D. R. Kigera, who was the Librarian, left ICIPE in the middle of 1981. A Chief Librarian — Mr. William E. Umbima — was appointed in November. Before joining the ICIPE, Mr. Umbima worked with the University of Nairobi.

## ACQUISITIONS

As usual off-prints on entomology and related fields dominated library acquisitions. Many of these originated from the ICIPE sponsored journal, *Insect Science and its Application*, while others came from renowned scientific journals all over the world. A supplementary stock of off-prints emanated from photocopies largely through the British library. In all, about 207 off-prints were received. There has been a great demand for these not just from the ICIPE scientists but also from scientists in other parts of the world. Multiple copies of some of the papers are available for exchange purposes.

The ever rising cost of books has meant a reduction in the number the library can afford to purchase. Only very essential textbooks were acquired, making a total of 101 new monographs for the year.

The same strain was experienced in the area of periodical subscriptions forcing the ICIPE holdings to be restricted to leading entomological and a few scientific journals of the world. These include major abstracting and indexing services in the same fields. The Swiss Academy of Sciences must be singled out for its generosity. Its support has enabled the ICIPE to maintain subscriptions of about 125 journals.

## VISITS

Mr. R. Labelle, Project Advisor (Information and Documentation), International

Council for Research in Agroforestry, visited the ICIPE Library/Documentation Centre in an advisory capacity for one day. Valuable discussions centering on the proposed Pest Management Documentation Project at the ICIPE were held between him and Senior ICIPE staff. Similar discussions were held with UNESCO through correspondence. Mr. G.A.R. Davis of the British Council also visited the Centre and discussed with ICIPE officials the possibility of British Council book aid for the new library to be established at Mbita Point Field Station.

## MBITA POINT FIELD STATION LIBRARY

The new building at Mbita Point Field Station has provision for a library. A Librarian has been appointed for May 1982. It is hoped that the new library will begin functioning fully towards the end of 1982. A grant of £1,200 for books from the British Council is a great boost for Mbita library. The books have already been ordered and should begin arriving soon.

## COOPERATION WITH OTHER ORGANIZATIONS

Arrangements for exchange of publications with organizations such as (IRRI) ICRISSAT etc. are working quite well. Similarly, inter-library loan arrangements especially with Kenya Agricultural Research Institute (KARI) and the University of Nairobi, have been a great help to the ICIPE.

## FUTURE PLANS

The ICIPE has plans for a Pest Management Documentation Service whose aims are:

- (1) To enhance and facilitate its research on crop pest and medical vectors of tropical diseases by providing an efficient and effective

*Library & Documentation*

documentation service to its scientists.

- (2) To serve as a nucleus for a network of information on insect science research. This will enable the ICIPE to play its role in increasing the capabilities of developing nations to tackle development oriented research objectives in the field of insect pests.
- (3) To supplement its own resources by having easy access to international information banks in the relevant field, in an effort to

devise alternative pest management systems that may be adopted for pest control in the tropics. Eventually, the results should contribute to finding a solution to real and urgent human needs such as food and health.

A modest start has already been made by compiling lists of documents and collecting basic literature. Detailed subject lists, indexes, information packages and subject files are envisaged. The project will go into full swing as soon as necessary help is received from donors.

# PROGRAMME ON BASES OF PLANT RESISTANCE TO INSECT ATTACK

## INTRODUCTION

Resistance to insect in crop species is governed either by major (vertical) genes expressed in one mechanism of resistance (or few closely related factors) or by many polygenes (horizontal) that each has a small effect on the trait and express a number of mechanisms involved in low levels of resistance. Plant resistance governed by major or vertical genes is generally believed to be of high level but short-lived because with a single gene conditioning resistance, only a single gene controlling nutrition or behaviour in the insect is required to overcome it. Plant resistance governed by polygenic or horizontal resistance is considered to be more stable and longer lasting than vertical resistance. This type of resistance is biotype nonspecific and there is no gene-for-gene relationships, so there is very little danger that biotypes will develop. Horizontal resistance is generally of low level and is difficult to work with because a number of various mechanisms are involved and it is difficult to develop methods which are sufficiently sensitive to separate lines possessing small differences of resistance.

Both types of biological systems are included in our studies on plant resistance in our Programme at the ICIPE. The rice resistance to the brown planthopper, *Nilaparvata lugens* and cowpea resistance to the cowpea aphid, *Aphis craccivora* represents the biotype specific resistance, and the maize and sorghum resistance to *Chilo partellus* and cowpea resistance to *Maruca testulalis* represents the moderate multifactorial resistance.

In 1981, research on plant resistance concentrated on six aspects:

Confirmation and expression of stem borer's resistance in maize

lines originated from CIMMYT and the Kenya National Programme;

Effect of maize and sorghum resistant lines to the spotted stalk borer, *Chilo partellus* with regard to behaviour, survival and development

New methodology of cowpea screening for resistance to the legume pod borer, *Maruca testulalis* under artificial infestation;

Effect of resistant cowpea lines originated from IITA on *Maruca testulalis* and the cowpea aphid, *Aphis craccivora* behaviour and development

Biochemical, physiological and genetical relationships between resistant rice cultivars released by IRRI and *Nilaparvata lugens* biotypes; and

Experimental bases of mass rearing of *Maruca testulalis*, *Chilo partellus*, *Eldana saccharina*, *Busseola fusca* and *Atherigona soccata* on artificial diets.

The joint project between the FAO/UNDP Kenya Sorghum and Millet Development Programme (F. Pinto, H. J. Enserink, Petra Penninkhoff) and the ICIPE was initiated to screen large germplasm collection by the Kenya Programme for resistance to stem-borers using the methodology developed by the ICIPE and two experiments were planted in the Mbita Point Experimental Station to identify mechanisms of resistance/non-acceptance, antibiosis and tolerance in their selected advanced lines. The breeders intend to use different sources of sorghum resistance to insects.

part of plant: extensive on the highly susceptible Inbred A reduced on all resistant lines tested.

- (4) Penetration of young larval instars into stem: reduced on CIMMYT lines 34 and 125 and Inbred G.
- (5) Feeding of older larval instars in stem: reduced on CIMMYT lines 324, 104, 125, 181, 28 and Inbred D.

Extensive larval feeding on the whorl and upper part of stem of susceptible Inbred A and low damage on the resistant lines may suggest that the plant resistance may result from the plant failing to provide positive gustatory stimuli required by *Chilo* larvae or by the possession of characteristics (biophysical factors, dense trichomes or biochemical) having adverse effects on feeding activities. The chemicals acting as feeding suppressants or deterrents may

also affect the larval survival (physiological inhibitors).

#### Behaviour and Development of *Chilo partellus* on Sorghum Cultivars

Although several breakthroughs have been made in studies on sorghum resistance to *Chilo* a clear picture has not emerged. This is mainly due to the fact that different workers have tended to concentrate on particular aspects of resistance. However, sorghum plant resistance is actually on several levels chemical, ecological, physical and even related to the phenology of the plant.

Seven field, screenhouse and laboratory experiments were conducted in 1981 on the effect of resistant sorghum cultivars on the behaviour, development, survival and fecundity of the spotted stalk borer, *Chilo partellus*.

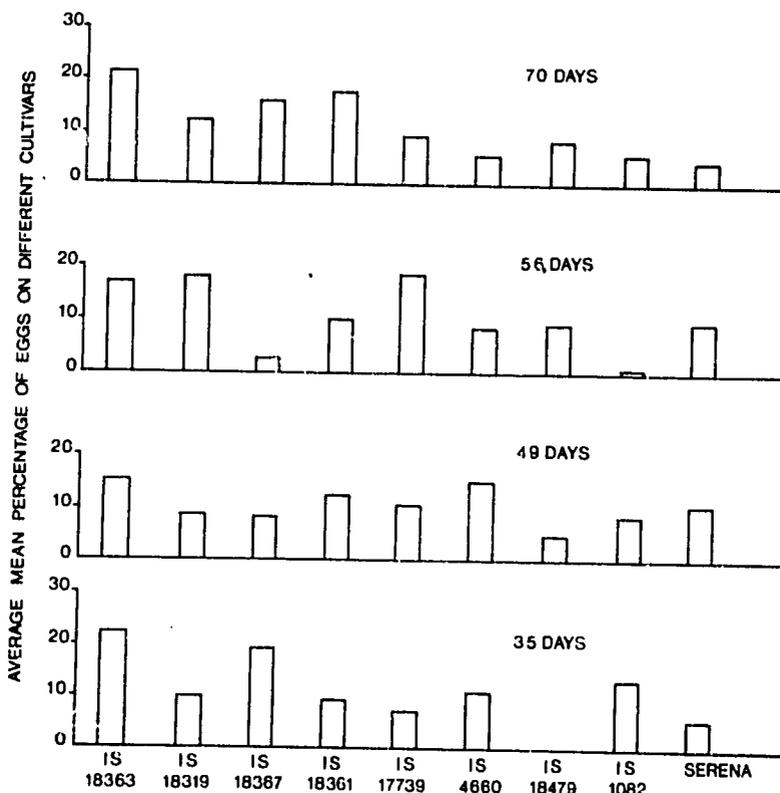


Fig. 3. Effect of cultivar and age of sorghum on the relative oviposition preference of *Chilo* females under screenhouse conditions.

The cooperation established by our Programme with Ahero Irrigation Research Station (W. Malinga) on the preliminary field screening of rice for resistance to *Maliarpha separata* under natural infestation was continued. We assist also in the methodology of bionomic studies on *Maliarpha* and other borer populations on rice.

The joint project on maize resistance to stem borers and the effect of cultivar upon the economic injury level of borers on maize was initiated by Kenyan entomologists working at the Coast National Agricultural Research Station, Mtwapa (C. M. Warui and J. K. Kuria). The Mtwapa station possesses specific conditions for collecting additional information on maize resistance to stem-borers for our studies in Western Kenya at Mbita Point Field Station. The species composition of stem-borers differ in these two locations. Besides, *Chilo partellus* there are two other species common on maize at the coast — *Chilo orichalcociliella* (the coastal stalk borer) and *Sesamia calamistis* (the pink stalk borer).

In the period under review, the programme created research facilities for Mr. G. T. Masina the entomologist from Swaziland who joined the ICIPE in January 1981 to work on mechanisms of sorghum resistance to *Chilo partellus* for his Ph.D. thesis. It should be mentioned that there is great interest among young African scientists to get training in research on mechanisms of plant resistance to insects in the Programme.

#### SCREENING MAIZE FOR STEM-BORER RESISTANCE

It is assumed that maize germplasm contain many genes for resistance to insects and gradual accumulation of the favourable genes in selected populations, the maize crop would be able to resist or tolerate pest hazards much better. New resistant varieties would benefit all classes of

farmers, particularly the small scale farmers in the rural areas.

The objective of screening therefore is to test and confirm the reported resistance of lines from other International Research Centres to the East African stem-borer complex under our local conditions. On a limited scale, local cultivars are also screened under field condition to select and develop local lines that are resistant to this stem-borer complex. Screening for *Chilo partellus* resistance will be carried out at Mbita Point Field Station, screening for *Busseola fusca* resistance will take place in the highlands of Kisii at the Nyanza Agricultural Research Station, Kisii.

Screening was carried out at Mbita Point Field Station and in farmers' fields. Observations were made on the plants to give the percentage damage based on the leaf and plant damage scale. At harvest, stems of individual plants were split open and the number of larvae/pupae including even pupal cases per plant were recorded. The length of the tunnelling of the stem and of magnitude of resistance was also recorded. Based on these data, in order of magnitude of resistance, the first ten and the last five were selected for testing and confirmation during the following season. Katumani composite maize is an early maturing variety (3–4 months maturity period) and Kitale Synthetic II is a late maturing cultivar (6–7 months maturity period). A cross between them is therefore medium maturing (4–5 months) and would be suitable for growing in the lake region of East Africa. A recombination of the Katumani lines and Kitambi lines separately in isolation will form new population of Katumani lines and Kitambi lines separately in isolation will form new population of Katumani composite and Kitale synthetic II which possess, to their credit, a higher level of resistance to crop borer complex. A cross between the improved populations of Katumani composite and Kitale Synthetic II will be a hybrid that in turn will be advanced to

three generations and tested before it is released to the national programmes for further testing and final release to the farmers.

A total of 460 maize lines from CIMMYT, Mexico reported to be resistant to the most widely spread borers affecting maize in the western hemisphere (*Ostrinia*; *Diatraea*) were planted under natural conditions in Mbita Point. Most of the maize lines showed good adaptability and high yield potential under dry conditions of Western Kenya. Twelve lines showing resistance to *Chilo* and good agronomic performance were selected and planted in the experiment on mechanisms of resistance.

### BEHAVIOUR AND SURVIVAL OF *CHILO PARTELLUS* ON RESISTANT MAIZE LINES

Fifteen maize lines identified previously as showing the lowest leaf and overall plant damage under natural field infestation were repeatedly planted in April and September 1981 at the Mbita Point Experimental Farm. The highest number of egg masses were always found on foliage of susceptible Inbred A line used as the control in both experiments. The lowest number of eggs were oviposited on the CIMMYT lines 324, 125, 34 and Inbred D. The number of holes and tunnels in stems was always higher on the suscepti-

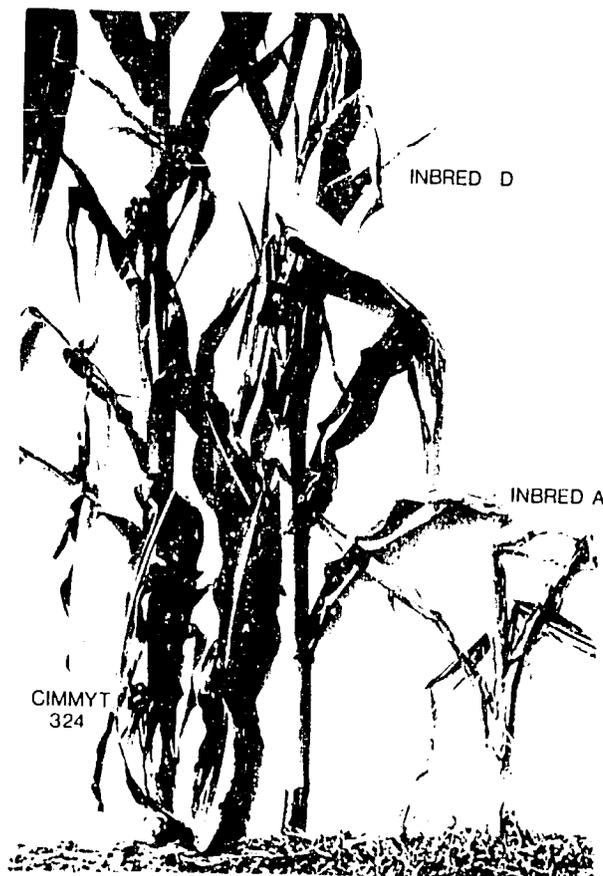


Fig. 1 Differences in infestation of the susceptible Inbred A and the moderate resistant live Inbred D and the resistant CIMMYT 324.

ble Inbred A. More significant differences were found when the frequency distribution of stem tunnelling (in % of plant height) by stem borer's larvae on Inbred A and D were compared.

It was also found that 60–70% of plants of the susceptible Inbred were completely destroyed in stage 4–6 *Chilo* larvae and only 0–4% of the moderately resistant CIMMYT lines and Inbred D and G (Fig. 1). The highest number of larvae and the longest tunnels were observed on the susceptible Inbred A followed by lines Mz5, CIMMYT 33, 34 and 71. The lowest number of larvae and the shortest tunnels occurred in lines 125 and 324. Non-preferred for oviposition line 34, showed moderate damage by the first generation larvae, but a high stem damage by the second generation.

Most of the larvae concentrated their feeding on the top 20cm portion of stem of the susceptible Inbred A line, whereas they were uniformly distributed on stems

of Inbred D and G. The extent of tunnelling on Inbred A was twice as long as on plants of Inbred D or G (Fig. 2). Those two factors: the longer tunnels and their concentration on the young growing portions of stems were responsible for destruction of susceptible plants by *Chilo* larvae.

The results of all experiments on maize resistance on *Chilo partellus* suggest that there are at least five levels of relationships affecting plant colonization, larval survival and damage level:

- (1) Non acceptance for oviposition: Inbred D and G and CIMMYT lines 125, 33, 324, and 178. Preference for the Inbred A (2–3 x more eggs).
- (2) Feeding of young larval instars on young leaves or leaf sheaths: reduced on CIMMYT lines 324, 125 and 33.
- (3) Concentration of feeding of young larval instars on the upper growing

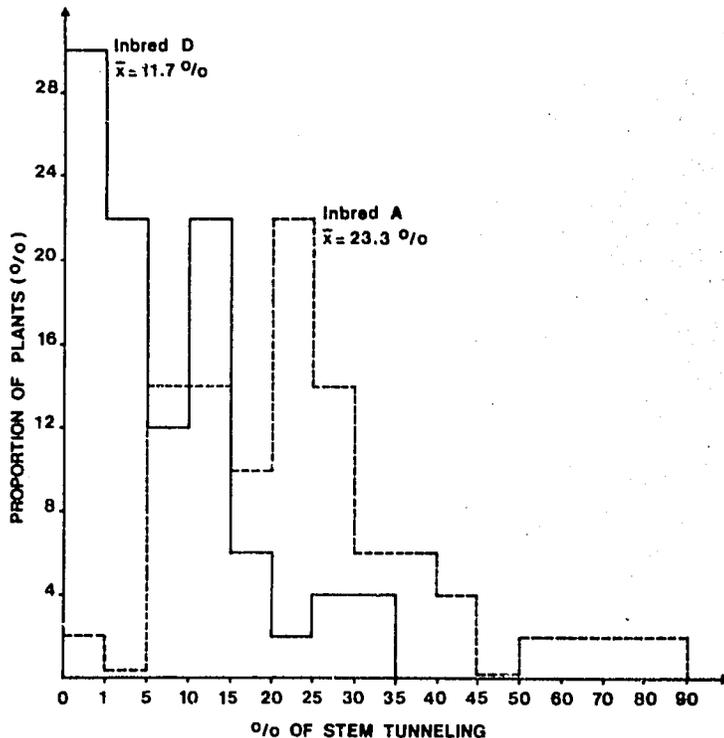


Fig. 2. Frequency distribution of stem tunnelling (in % of plant height) by stem borers larvae in two maize inbred lines. Mbita Point Field Station, 1981.

(1) Non-acceptance for oviposition

The aims of our experiments on the oviposition preference of *Chilo* females was to find out whether: the host plant variety is oviposited on by chance or there are some factors involved in its location; oviposition preference for any particular variety exists and if this is also related to the age of the plant; there is visual, olfactory and contact stimuli both physical and chemical in orientation and oviposition.

Most of the eggs were oviposited on the middle leaves. Slightly more eggs were laid on the upper side, mostly on the midrib, than on the lower side. The ratio was about 60:40 and did not vary considerably on different cultivars except on IS 17739 and IS 18361, where the ratio was reversed. It was observed that oviposition preference of *Chilo* females to sorghum cultivars is determined by several factors modified by the age of the plant. Some of these factors interact or even negate each other. The coefficient of variation was 15.3%.

IS 18363 was consistently the most preferred cultivar for *Chilo* — oviposition and thus possibly releasing positive chemical or physical stimuli for the females (Fig. 3). IS 18479 IS 1082 and Serena are the least preferred, probably due to different factors. IS 18479 may not be preferred because of its short height among other things. In that event, it would not have any advantage when planted alone. IS 1082 is one of the least preferred from 42 days onwards. It needs to be established as to what changes take place at this stage. IS 4660 seems to contain either a repellent or some undesirable physical characteristics affecting *Chilo* oviposition. At the early stages, IS 17739 is also non-accepted and thus need to establish the reason.

(2) Migration and establishment of first larval instars on sorghum cultivars

Five first instar larvae were released onto

22 selected cultivars of sorghum in the four screenhouses in the Mbita Point Field Station. The larvae were then monitored for migration, mortality and the damage they caused to plants. This experiment was replicated five times.

IS 1044, IS 18489 and IS 18349 had the highest migration, low mortality and low plant damage. This leads to the suspicion that these cultivars have some deterrents or suppressants affecting feeding of the first instars. In IS 1151, IS 4660 and IS 18479, there was little migration but leaves were extensively damaged and a large number of larvae died. From this, it is inferred that some factor(s) (chemical or physical) caused high mortality but there were no deterrents. Finally, in some cultivars there was high migration, high mortality but still extensive leaf damage. These cultivars are IS 1082, IS 2205 and IS 18367. The differences observed in migration of larvae from various cultivars indicate presence of some chemical or biophysical factors restricting initiation of feeding by first instars.

(3) Stem damage by *C. partellus* larvae

Selected sorghum cultivars were planted in randomized complete block with six replications under field conditions. From 28 days after planting the plants were sampled for eggs and larvae. Although *C. partellus* was the predominant species (74.7%), other borers were also found (*Sesamia calamistis* — 14.6%; *Busseola fusca* 6.6%; *Eldana saccharina* — 4.2%). This ratio varied, however, during the course of the long and short rain seasons. The resulting plant damage was therefore due to all four species of borers.

The least tunnelled cultivars were: IS 18489, Serena, IS 2122 and IS 1151. However, IS 2122 also had the highest incidence of lodging. This suggests that the cultivars tensile strength causing tolerance is not very high and the plant may contain some chemicals restricting larval tunnelling.

The least number of plants killed were IS 1151, IS 18349, IS 2122 and IS 2205.

(4) Effect of sorghum cultivar on larval survival, pupation and fecundity

When plants were sampled for borers, larvae were brought to the laboratory for antibiosis studies. They were reared in pieces of stem of the same cultivar until pupation. Pupal periods in each cultivar were recorded as well as pupal weights, mortality, sex and finally the number of egg batches from each resulting female moth. The least pupal weights were from IS 2122 Serena and IS 2205. The lowest adult emergence was from IS 18489 and IS 1082, while the fertile moths were raised from IS 4660, E 303 and IS 1082.

METHODOLOGY OF COWPEA  
SCREENING FOR RESISTANCE TO  
*MARUCA TESTULALIS*

Methodology of cowpea screening for resistance has necessitated some preliminary screenhouse experiments to be conducted on: Identification of the optimal number of insects for artificial infestation that will allow differentiation among susceptible and resistant lines; most suitable developmental stage of *Maruca* for artificial infestation in mass screening programme; and effect of plant growing stage on the expression of resistance. The TVu 946 line was included in our experiments as resistant, Ife Brown as moderately resistant and Vita I as the susceptible cultivar.

Eight different treatments on potted cowpea plants at the pre-flowering stage and on plants growing on the ground of four screenhouses under Mbita Point conditions were used in the experiment: (1) 5 larvae per potted plant; (2) 10 larvae per potted plant; (3) 5 larvae per plant grown on the ground; (4) 10 larvae per plant grown on the ground; (5) 5 eggs per potted plant; (6) 10 eggs per potted plant; (7) 5 eggs per plant grown on the ground and (8) 10 eggs per plant grown on the ground. The treatments with 5 and

10 eggs per plant released at the pre-flowering stage were repeated under natural field conditions. Four additional treatments were used at the flowering stage of plants grown under natural field conditions: (1) 10 eggs per plant (2) 20 eggs per plant in both pre-flowering and flowering stage; (3) 5 eggs per plant in the pre-flowering stage and 10 eggs per flowering stage and 10 eggs per flowering plant; and (4) 5 eggs per pre-flowering plant and 20 eggs per flowering plant. Small pieces of leaves with oviposited eggs cut out from plants exposed to the females in the oviposition cages were fixed with office glue to the stem.

Larval survival after 5, 10, and 15 days following artificial infestation of three cowpea lines by *Maruca* eggs or larvae was used as the index measuring the plant suitability for the pod-borer development. The suitability of using young plants or seedlings as the cheapest and the most rapid method in the screening of cowpea for *Maruca* resistance was also verified. The potted cowpea plants were kept on the tables in one of the screenhouses at Mbita Point to protect young *Maruca* larvae from predation by ants. It was previously observed that ants collected first larval instars of *Maruca* from cowpea plants grown on the ground, consequently changing the original level of infestation and increasing the range of error between plants of the same treatment. The number of insects (larvae or eggs) used for artificial infestation would therefore depend upon two factors: destructive effect of ants' population on *Maruca* population; and minimal level of larval population segregating cowpea line for resistance after elimination of the ant's effect.

Artificial infestation of young cowpea seedlings (average number of plant shoots 1-3) did not show clear differences in larval survival on lines tested. Significant differences in larval survival were observed on plants with average number of terminal shoots varying from 5-7 in the pre-flower-

Table 1. Survival of *Maruca testulalis* larvae on different parts of three cowpea lines under artificial infestation.

Treatment: Cowpea plants grown on the ground in four screenhouses infested with 5 larvae/plant

Plant parts	VITA 1			IFE BROWN			TVu 946			
	After Infestation	5 d	10 d	15 days	5 d	10 d	15 d	5 d	10 d	15 d
Av. no. of plant shoots		4.1	4.8	5.0	5.6	6.0	6.1	7.0	7.3	7.4
Larval survival (%)		45.5	29.6	20.0	44.0	21.6	16.8	24.0	12.0	12.8
Fraction of larvae feeding on (%):										
Stems		—	24.3	36.0	—	14.8	19.8	—	—	6.3
Leaves		21.1	10.8	16.0	7.3	25.9	33.3	30.0	46.7	31.1
Leaf stalks		—	35.2	32.0	—	14.8	42.9	—	—	18.8
Terminal buds		78.9	29.7	12.0	92.7	44.5	4.8	70.0	53.3	—
Flower buds		—	—	4.0	—	—	—	—	—	18.8
Flowers		—	—	—	—	—	—	—	—	25.0

ing stage grown on the ground and treated with 5 larvae/plant. After 5 days, 45.5% survival was recorded on Vita 1, 44.0% on Ife Brown and 24.0% on TVu 946 (Table 1). Observations on the distribution of larvae on different cowpea plants showed significant differences between the lines tested. An average of 24.3% *Maruca* larvae were found feeding in stems of Vita 1; 14.8% on Ife Brown and none in stems of TVu 946. However, 46.7% of pod-borer were found feeding on leaves of TVu 946 line on the tenth day after infestation; 25.9% on Ife Brown and only 10.8% on leaves of Vita 1.

The preference of *Maruca* feeding on different parts of young cowpea plants at pre-flowering stage has modified the expression of resistance and allowed

*Maruca* to establish its colony on all tested cowpea cultivars. It was often recorded that *Maruca* larvae moved from terminal buds to young cowpea leaves on TVu 946 line, probably avoiding the adverse effect of these points on their development.

Plant growing stage has modified the expression of cowpea resistance to *Maruca* larvae. Five to seven shoot stage (not younger) was found to be the most suitable for screening for resistance in pre-flowering period. By using 10 eggs per plant in pre-flowering stage and 20 eggs per plant in flowering stage, it was possible to differentiate between the resistant and susceptible lines. Higher larval population used for artificial infestation did not improve the screening results.

EFFECT OF RESISTANT COWPEA  
LINES ON THE LEGUME POD-BORER  
*MARUCA TESTULALIS*

Our screenhouse and field experiments on cowpea resistance to the legume pod-borer showed that the resistance in some cowpea lines of IITA origin is expressed in the pre-flowering and flowering stage. At present we may only identify three levels of *Maruca* — cowpea relationships expressed in the resistance.

- (1) Vita I plants showed significantly higher number of eggs per plant, followed by Ife Brown and 946. Presently planted cage experiments using only one cultivar in the cage in the non-choice situation should allow us to distinguish if we deal with the oviposition preference on *Maruca* to Vita I, resulting in the low oviposition of TVu 946 or with true resistance expressed by the non-acceptance for oviposition in TVu 946 line acting in the choice and non-choice situation. In the current laboratory experiments on *Maruca* oviposition behaviour, we try to estimate the differences in the females' orientation to various cowpea cultivars (response to odours) and their behaviour on the plant after landing (response to biophysical and contact chemical stimuli).
- (2) Low level of colonization of TVu 946 line in pre-flowering stage. Significantly less larvae of *Maruca* were found feeding on stems of TVu 946 than on Ife Brown and Vita I.
- (3) Low level of colonization of stems, flower buds, flowers and pods of TVu 946 line in flowering stage.

BIOCHEMICAL BASES OF COWPEA  
RESISTANCE TO *MARUCA TESTULALIS*

Preliminary results at the ICIPE on three

of the cowpea cultivars selected for study, indicate that there is a biochemical component to the overall resistance of stems of TVu 946 to *Maruca testulalis*, when this cultivar was compared with the susceptible Vita I and the moderate Ife Brown.

In an attempt to elucidate the mechanisms of resistance in stems of these cultivars, estimation of nutrients show that the total sugars and total amino acids are quantitatively less in the resistant TVu 946 (15.1 mg per g and 13.8 umole per g fresh weight respectively) than in both the moderate Ife Brown (18.68 and 21.52) and the susceptible Vita I (21.50 and 22.65). For total phenols and total flavonoids, results indicate that TVu 946 stems contained a higher concentration of these secondary metabolites (i.e. 0.58 and 0.55 mg chlorogenic acid equiv. per g fresh weight respectively than Ife Brown (0.49 and 0.47) and Vita I (0.45 and 0.44)

In a preliminary experiment, finely ground lyophilised cowpea stems of the three cultivars were incorporated in a synthetic diet (Diet 6) of first instar *Maruca* larvae, at concentrations of 1%, 0.5% and 0.25% dry weight. There was a clear difference between the duration of larval development to adult of all the treatments (20–25 days) compared with the control (18–22 days). However, there was no difference between pupal weights of control and treated at these concentrations. No difference between cultivars or between concentrations were observed for duration of larval development or pupal weight at these concentrations.

An extraction procedure which resulted in several extracts of different polarity, yielded hexane, ethyl and aqueous extracts (lipophilic, intermediate and hydrophilic, respectively) together with the residue. These were incorporated in diet 6 and the larval development and mortality of first instar *Maruca* larvae monitored. After 7 days, results show that there is a slightly

higher mortality in the hexane extracts of the cultivars (55–60%) compared with the control (35%). With the ethyl acetate extracts on the other hand, although there was a slightly higher larval mortality in extracts from Ife Brown (65%) and Vita I (60%) compared to control (40%), there was a clearly larger difference in mortality of larvae in extracts of TVu 946 (80%) than in the control. No difference (35–40%) was observed between mortality in control diet 6 and hexane or ethyl acetate solvents (4%) incorporated in the diet. These results indicate that the ethyl acetate extract from TVu 946 contains a higher concentration of compound(s) (allelochemicals) which have a deleterious effect on the survival of *Maruca* larvae.

#### RESISTANCE OF COWPEA TO THE COWPEA APHID, *APHIS CRACCIVORA*

Cowpea aphid or groundnut aphid, *Aphis craccivora* is the main aphid pest of cowpea. It was previously reported as a major pest of cowpea in Asia and a minor pest in Africa. However, recent observations indicate that heavy aphid populations are more frequently and widespread in Africa. Cowpea aphids infest the crop at seedling stage and the direct damage to the host plant is by this pest, even when populations are small, it transmits the cowpea aphid-borer mosaic.

Four cowpea lines selected previously by the IITA scientists as resistant (TVu 310, 410, 408-P-2 and 2806) and two susceptible cultivars (Vita I and TVu 946) to *A. craccivora* were included in our cage and field experiments on the effect of resistance upon aphids behaviour, survival and development. The cage observations showed that there were significant differences in the developmental rate of the initial population of 10 young females on the resistant and susceptible cultivars (Fig. 4). Detailed observation on: (1) aphids selection of host plant under choice and non-choice situation; (2) fecundity of

females on 2, 4, 6 and 8 weeks old plants; and (3) survival of females and nymphs on various cultivars showed that:

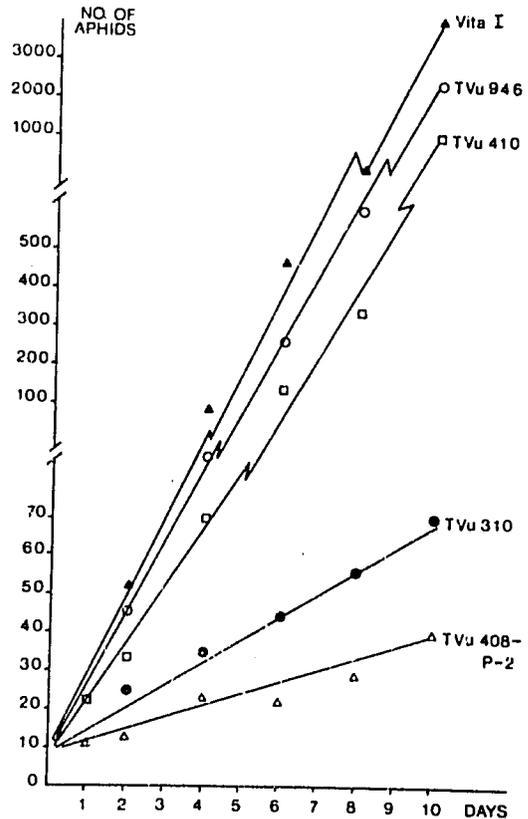


Fig. 4. Effect of cowpea cultivar on the development of original population of 10 females of *Aphis craccivora* on two weeks old plants undercage conditions.

- (1) the same number of alatae females responded to the resistant and susceptible cowpea lines;
- (2) *A. craccivora* females did not accept resistant lines for feeding (presence of suppressants or deterrents or lack of feeding incitants or stimulants) suggesting the existence of chemicals reducing the aphid's feeding;
- (3) *A. craccivora* females and nymphs showed clear gustatory preference to susceptible lines Vita I and TVu 946 suggesting existence of strong feeding stimulants;

- (4) Fecundity of *A. craccivora* females was very low (if ever) on TVu 310, 2896 and TVu 408-P-2 and very high on susceptible line TVu 946 and Vita I, suggesting the existence of antibiosis in the resistant lines;
- (5) TVu 410 line selected as resistant under IITA, Nigeria conditions showed only a low level of resistance in comparison with the susceptible line TVu 964 and Vita I. The difference in TVu 410 suitability for Kenyan and Nigerian population of *A. craccivora* may suggest the existence of different aphid biotypes;
- (6) The susceptible cultivar Vita I being a suitable host for *A. craccivora* development showed some level of tolerance than the susceptible line TVu 946 (all 2, week - old plants with five females were destroyed by aphid feeding during the next 2 weeks).

The present glass house and laboratory experiments with application of radioactive materials should allow us to disclose the role of feeding incitants, stimulants, suppressants and deterrents in the acceptance of host plant by *A. craccivora* and identify the nature of antibiosis (physiological inhibitors or nutritional deficiencies).

#### BIOCHEMICAL BASES OF COWPEA RESISTANCE TO *APHIS CRACCIVORA*

Several cultivars have been chosen for the detailed study of the biochemical factors involved in the resistance mechanisms of cowpea to *Aphis craccivora*. Estimation of total sugars and total amino acids in finely ground lyophilised whole stems of these three cultivars - TVu 310 - resistant; 408-P-2 - resistant and Vita I - susceptible - reveal that there is slightly more sugars in TVu 310 (18.3mg/g fresh wt) than in Vita I (16.5 mg/g and 408-P-2 (13.9 mg/g) and slightly more amino acids in 408-P-2 (30.5  $\mu$ mole/g fresh

wt) and TVu 310 (29.4  $\mu$ mole/g) than in Vita I (21.8  $\mu$ mole/g).

Total phenols are higher in the two resistant cultivars 408-P-2 (1.02mg chlorogenic acid equiv./g fresh wt and TVu 310 (0.91) than in the susceptible vita I (0.68mg). Similar results were obtained for total flavonoids, 408-P-2 (0.81 mg/chlorogenic acid equiv./g fresh wt) TVu 310 (0.65mg) and Vita I (0.59mg).

Confirmatory experiments are in progress together with the development of a bioassay for the presence of feeding stimulants/deterrents in extracts of the various cultivars and on the induced production, utilisation and/or redistribution of nutrients and secondary metabolites in the plant. Also in progress are experiments designed to decipher the exact tissues of the plant on which the aphids feed, the importance of which cannot be overemphasized.

#### MECHANISMS OF RICE RESISTANT TO BIOTYPE I BROWN PLANTHOPPER, *NILAPARVATA LUGENS*

Mechanisms and possible causes of resistance in rice to Biotype I brown planthopper (BPH) was studied on six resistant and one susceptible variety. Resistant varieties were selected on the basis of the different genes possessed by them. These included Mudgo with BPH 1 gene, ASD 7 BPH 2 gene natha Haenati with BPH 3 gene, Babawee with BPH 4 gene and PTB 33 with 2 genes for resistance. ARC 6650 is a moderately resistant variety but its genetic composition has not been studied. TNI served as the standard susceptible check variety.

The study was divided into two major phases. Phase I involved the study of the biology of the BPH on the test varieties. The experiments conducted included settling response of adults, nymphal development, female longevity and fecundity, oviposition and egg hatch

Table 2. Longevity, fecundity, oviposition, and hatching of eggs of Biotype 1 brown planthopper females on susceptible and resistant rice varieties (IRRI, 1980-81)<sup>1</sup>

Variety <sup>2</sup>		Longevity <sup>3</sup> days	Fecundity <sup>3</sup> (No. of Eggs/ Female)	Oviposition <sup>4</sup> (No. of Eggs/ 5 Females/18h)	Egg hatch (%) <sup>5</sup>
TNL	(S)	10.6 a	444.2 a	112 a	88.0 a
Mudgo	(R)	3.8 b	89.9 b	111 a	69.3 bc
ASD7	(R)	4.0 b	124.5 b	111 a	82.7 ab
Rathu Heenati	(R)	4.0 b	105.7 b	112 a	65.3 c
Babawee	(R)	4.7 b	113.9 b	120 a	69.0 bc
Ptb 33	(R)	3.8 b	88.3 b	101 a	69.3 bc
ARC 6650	(R)	4.8 b	100.2 b	95 a	68.0 bc

<sup>1</sup>In a column means followed by a common letter are not significantly different at the 5% level by DMRT.

<sup>2</sup>S = susceptible check; R = resistant

<sup>3</sup>Average of 10 replications

<sup>4</sup>Average of 5 replications

<sup>5</sup>Average of 6 replications

(Table 2). Also the quantity of food ingested and the metabolic utilization of ingested food from the test varieties by adult females was determined.

BPH adults preferred to settle on the susceptible variety (TNI) over the resistant varieties after 6 hr of introduction into the test cage. Mortality of the nymphs was very high on resistant varieties and the nymphal growth index was correspondingly very low. All the resistant varieties had a lower amino acid composition than that in the susceptible TNI variety.

Plant allelochemicals were obtained by stem distillation of leaf sheaths followed by diethyl ether extraction. Bioassays of allelochemicals included orientation, settling and feeding response of adults, toxicity to both nymphs and adults, and phagostimulation or inhibition when added to 10% sucrose solution when offered to the BPH. Effect of extracts on egg hatch was also examined.

The insects were significantly more

attracted to the rice plant odours than to the blank source in an olfactometer. However, they were unable to differentiate between odours of resistant and susceptible varieties. Significantly, more insects settled and fed on TNI plants treated with TNI extract and control plants than on plants treated with extracts from resistant varieties. Allelochemicals from resistant varieties were more toxic to both nymphs and adults of the BPH than allelochemicals from TNI (Table 3). Toxicity of the allelochemicals from resistant varieties when applied topically to adults, increased with plant age up to 60 days after seeding and then decreased from 100 days after seeding which suggests that the level of resistance changes with plant age. TNI allelochemicals stimulated more feeding on 10% sucrose solution than that of the resistant varieties. Egg hatch was not affected by these allelochemicals (Table 4).

Although the allelochemicals evaluated in the present study are not likely to be found in the phloem which is the feeding site of the BPH, their odoriferous and volatile

Table 3. Toxicity of steam distillate extracts of susceptible and resistant rice varieties to 1st instar Bio-type 1 brown planthopper. IRRI, 1981 (Extracts were sprayed on the susceptible rice plant tillers).

Variety 1		Mortality (%) <sup>2</sup>			
		Dose (mg Extract/30-Day-Old Plant)			
		0.01	0.1	0.5	1
TN1	(S)	2.5 ab	50 bc	0.0 d	40.2 b
Mudgo	(R)	0.0 b	7.5 abc	10.0 cd	70.2 a
ASD 7	(R)	13.1 a	10.6 abc	15.8 bc	61.1 a
Rathu Heenati	(R)	5.0 ab	13.4 ab	20.8 abc	73.3 a
Babawee	(R)	5.0 ab	2.5 c	24.3 ab	71.1 a
Ptb 33	(R)	7.5 ab	18.1 a	32.1 a	70.1 a
ARC 66 50	(R)	7.5 ab	16.2 a	34.6 a	51.5 a

<sup>1</sup>S = susceptible; R = resistant

<sup>2</sup>Average of 4 replications. In a column, means followed by a common letter are not significantly at the 5% level by DMRT. Extract sprayed on the susceptible rice plant tillers.

Table 4. Phagostimulation by steam distillate extracts of susceptible and resistant rice varieties IRRI, 1981

Variety <sup>1</sup>		Quantity of sucrose solution <sup>2</sup> ingested (Mg) <sup>1</sup>
TNI	(S)	12.037 a
udgo	(R)	4.550 bcd
ASD 7	(R)	12.502 a
Rathu Heenati	(R)	2.350 cd
Babawee	(R)	6.532 bc
Ptb 33	(R)	0.657 d
ARC 6650	(R)	7.927 b
SUCROSE (control)		6.312 bc

<sup>1</sup>S = susceptible; R = resistant.

0.1 mg of extract per parafilm sachet was incorporated with 10% sucrose solution offered to 20 newly-emerged females.

<sup>2</sup>Average of 4 replications. Means followed by a common letter are not significantly different at the 5% level by DMRT.

nature makes them have a strong influence on the internal and external chemical environment of the rice plant. These allelochemicals are therefore of ecological significance in the BPH resistance in rice plant. Gas chromatograms showed distinct

qualitative and quantitative differences among the steam distillate volatiles. These 'fingerprints' of the steam distillate volatiles illustrate basic chemotaxonomic differences between the BPH-susceptible and resistant rice varieties.

## CYTOGENETIC VARIATIONS IN THE BROWN PLANTHOPPER BIOTYPES 1 AND 2

Chromosome number, morphology and behaviour have often been relied upon as complementary taxonomic indicators in a number of species complexes. The sex chromosomes are especially useful in cytotaxonomy because they may show from marked to subtle differences within a genus, or a species. In 1981, cytological investigations of the meiotic chromosomes of the brown planthopper Biotype 1 and 2 populations, maintained as stock cultures at IRRI for several years, revealed that the first meiotic division was reductional and the second division equational for all the components of the species' genome. The male diploid number was  $2n = 30$ , consisting of 14 bivalent autosomal pairs and XY sex chromosomes. Thus, *Nilaparvata lugens* has an XY sex — determining mechanism, the males being heterogametic ( $14\text{II} + \text{XY}$ ) or producing two types of secondary spermatocytes and the females homogametic ( $14\text{II} + \text{XX}$ ) or producing only one type of secondary oocytes.

Chromosomal behaviour during metaphase 1 featured clustering of highly condensed and shortened autosomes at the equatorial portion of the reproductive cell and separation of the highly heterochromatic, unequally synapsed sex chromosome from the autosomal grouping. The clustering together of autosomes is mainly due to intrachiasmatic and interchiasmatic matrices between the homologous bivalent chromosomes and among the tetrads or homologues.

A total of 218 and 200 metaphase 1 stages in testicular cells from 60 newly-emerged males of each of BPH Biotypes 1 and 2, respectively, were examined. In Biotype 1, 147 (68%) cells showed complete aggregation of sex chromosomes with autosomes, while the rest of the cells revealed a slight separation of sex chromosomes from the autosomes. On the other hand, almost 100% of the observed meta-

phase 1 chromosomes of Biotype 2 testicular cells manifested complete isolation of the sex chromosomes from the autosomal groupings. Thus, the sex chromosome is more isolated from autosomes in Biotype 2 than in Biotype 1. Also, the extent of chromosome clustering was detected to be higher in the former than in the latter. The occurrence of chromosomal aberrations as 'loose pairings' of paired homologous bivalents as well as fragmentations or chromosomal deletions were found to be more frequent among Biotype 1 than among Biotype 2 chromosomes. Further cytogenetic studies are in progress.

## EXPERIMENTAL BASES OF INSECT MASS REARING

Insect rearing is a complex and specialized field. It is a backbone of modern experimental entomology. The development of new pest control strategies depends upon our ability to rear and manage quality insects in the insectary. In realization of the importance of insect rearing, in the ICIPE, an experimental unit for mass rearing has been set up to research and develop methods for insect pests of tropical Africa. The target species include sorghum shootfly (*Atherigona soccata*) and several lepidopterous crop borers: *Chilo partellus*, *Busseola fusca*, *Maruca testulalis*, *Maliarpha separata*, *Eldana saccharina* and *Sesamia calamistis*.

There are four basic requirements in developing a practical rearing programme: (1) formulation of a satisfactory artificial diet; (2) precise study of the life cycle on the diet under laboratory conditions; (3) development of a mass rearing procedure; and (4) manipulation or management of a colony for supply of experimental insects.

## THE SORGHUM SHOOTFLY, *ATHERIGONA SOCCATA*

Over 35 artificial diets were formulated and evaluated in the laboratory. Several diets based on casein, yeast and pepper

(shredded kleenex tissue paper) were found equally satisfactory. The rearing was done in 75 x 25 mm glass vials at 27°C. On the best diet, the larval period was 13–15 days and adult emerged in 21–25 days. Larval survival was 64%. To date four generations have been completed and the third is on its way. Fecundity study is continuing; indications are that egg production is comparable to that of wild flies. It seems that a reasonably satisfactory diet and a rearing method has been developed. The most serious problem at present is the microbial contamination, which is partly due to unsatisfactory laboratory conditions and use of unbalanced antimicrobial inhibitor. Efforts are now concentrated to solve this problem. When contamination is controlled, it is expected that larval survival will considerably improve, resulting in higher adult yield. The work will continue until a practical rearing method is developed.

#### THE SPOTTED STALK-BORER, *CHILO PARTELLUS*

The larvae were reared at 20, 25, and 30°C on two diets: the wheat germ diet without plant powder added and the diet with plant powder added (already in use at the ICIPE). Both diets were found equally satisfactory. The average life cycle from egg to adult was 36 days at 30°C, 48 days at 25°C, and 68 days (estimated, adults still to emerge) at 20°C, respectively. On the whole rearing performance at 25°C was better; this temperature was recommended for developing mass rearing procedure because of higher yield and better quality insects. Evaluation of mass-rearing method using locally available glass bottles and plastic

boxes is in progress. A draft of model for mass-rearing method was prepared which will be improved as data becomes available in due course of time.

The microbial contamination is under control. At present the culture is in its second generation and in excellent health. In 2–3 months, we expect phase 3, that is management of the colony to start and then supply of insects for experimentation can begin.

#### THE LEGUME POD-BORER, *MARUCA TESTULALIS*

Several diets were evaluated including the previous diets with cowpea flower powder added and already in use at the ICIPE. An improved diet without cowpea flowers was formulated and compared to the diet made by Pritam Singh and patented in New Zealand. The life cycle data on the best diet at 25°C are: larval period 10–12 days, pupal period 6 days; adult recovery from neonate larvae, 60%. The adults have not reproduced in the laboratory due to lack of suitable oviposition method. This should be investigated and given priority. We also discovered a protozoan disease which is under investigation with collaboration of W. A. Otieno.

#### THE MAIZE-BORER, *BUSSEOLA FUSCA*

This species has a life cycle than the two species described earlier. The insect has developed on Pritam Singh's artificial diet and larvae have pupated starting from day 35; some larvae have entered diapause. It is expected that normal adults will emerge in due course. Special attention should be paid to diapause behaviour.

# CROP BORERS RESEARCH PROGRAMME

## INTRODUCTION

The Crop Borers Research Programme was established in late 1979 with its main objective to develop environmentally safe and economically feasible Integrated Pest Management practices for crop-borers of major food crops of Africa. The target crop-borer species under active investigation are: sorghum shootfly, *Atherigona soccata*; maize and sorghum stem-borer, *Chilo partellus*; rice stem borer, *Maliarpha separatella*; and cowpea pod-borer, *Maruca testulalis*. In addition, information on the importance of other stem-borers such as *Busseola fusca*, *Sesamia calamistis* and *Eldana saccharina* of maize and sorghum, and aphids and thrips of cowpea is being collected.

To formulate effective integrated pest management practices investigations cover the following areas:

- (i) Biological and ecological studies on the target pests.
- (ii) Physiology and behaviour of the target pests.
- (iii) Biological control agents.
- (iv) Crop loss assessment for sorghum, maize, rice and cowpea in respect of economic thresholds of the target pests.
- (v) Intercropping in terms of pest management.
- (vi) Screening for resistance and tolerance, confirmation of pre-selected materials from International Agricultural Research Centres and National Programmes under Mbita Point Field Station conditions and other representative ecological conditions.
- (vii) Genetics of resistance and tolerance.

Apart from laboratory studies on the sorghum shootfly, Crop-Borers Research is carried out at the Mbita Point Field Station

(MPFS) and at nearby farmers' fields where the hosts and their pests occur in their natural habitat. Existence of a high pest population at MPFS facilitates screening of cultivars under natural infestation.

The Crop Borers Research Programme witnessed a great expansion in its scientific manpower and research activities during this period reflected by a number of findings. Sorghum Shootfly population level depends on rainfall, availability of host plants, rate of parasitism and predation and availability of food to adult. Honeydew produced by cereal aphids and cowpea aphids is an efficient food source for shootfly adults and may influence the shootfly population in the field. *Chilo* is the most abundant borer (over 90%) infesting sorghum and maize, particularly in the warmer and low altitude areas of Kenya. In the cooler and high altitude areas, however, *Busseola* is found to be the most important. Preliminary studies on the rice stem borers have indicated that *Maliarpha* is the most abundant. However, *Diopsis* and *Sesamia* caused high incidence of 'dead-hearts' at vegetative stage and 'white-heads' at reproductive or ripening stages, respectively. Studies on cowpea pod-borer, have shown that under both pure and mixed crops, *Maruca* is the most abundant accounting for more than 90% of the total borer population. Studies on the development of cowpea plants and damage/injury levels or threshold of *Maruca* infestation have been initiated. Intercropping experiments to determine the importance of pests within mixed ecosystem of maize, sorghum and cowpea are being conducted. Studies have been initiated on the genetics of host plant resistance and tolerance to target insect pests.

The Crop Borers Research Programme collaborates with CIMMYT, ICRISAT, IITA, IRRI and WARDA, and National Research Programmes of Kenya.

## SORGHUM SHOOTFLY

## Population dynamics

*Atherigona soccata* population levels are closely linked with the availability of sorghum (wild and cultivated) stems of a suitable stage. Field experiments using susceptible sorghum hybrid (CSH-1) at MPFS have shown that *A. soccata* females lay more eggs on young sorghum seedlings measuring 4 to 8 cm in height than on plants of any other size and newly hatched larvae survive only in shoots measuring less than 24 cm in height. Survival of the first instar larva depends on the size of the host plant, resistance to penetration of the leaf sheaths and the distance between infestation site and growing point. Sorghum plant density affects oviposition by *A. soccata*. Low density plots with stouter plants bearing broader and greener leaves received 3.35 times more eggs than plants in higher density plots. Larval mortality resulting from competition increased from high to low plant densities. These results support the farming practice of using higher seeding rates and subsequent thinning of infested plants.

Heavy rainfall after dry months was found to be detrimental to shootfly adult population. It took 3 months for shootfly population levels to become re-established after the rainy season. However, water plays a central and preponderant role in increasing shootfly population: it promotes the growth of wild and cultivated sorghums and has a direct effect on survival of every stage of the pest, except for the second and third instars, which are completely protected inside the plant tissues. A sufficient air humidity is required by the egg to complete its development and for hatching, dew is necessary for the progression of the newly hatched larva from egg site to the point where it enters the stem. Mortality increased rapidly with decreasing humidity (from 100%), especially at temperatures below 12°C and above 32.5°C. No development occurred at

humidities lower than 67%.

In local farming practice, cattle graze on sorghum stubbles after harvest and certainly help reduce larval populations during the dry season, Aphid honeydew is a major source of food for shootfly adults. Flies fed on honeydew of cowpea aphids and cereal aphids had very high fecundity. The part played by aphid population fluctuations may be of considerable significance to understanding of shootfly population fluctuations.

Among the major parasites and predators of the sorghum shootfly, the following have been identified: (i) *Tetrastichus nyemitawus* — a larval parasite, (ii) *Trichogramma kalkae* Sch. & Feij. : — an egg parasite, and (iii) *Scymnus trepidulus* Weise — an egg predator.

## MAIZE AND SORGHUM STEM-BORERS

## Stem-borers complex

Previous studies on stem-borers complex have shown that *Chilo partellus* contributed to over 90% of all the borer species infesting the lowland maize and sorghum. Recent studies on the incidence of stem-borers complex on sorghum variety Serena at MPFS have confirmed that although all the four stem-borer species, viz: *Chilo*, *Eldana*, *Busseola* and *Sesamia* were found damaging, *C. partellus* was the predominant species (87 to 99%) followed by *Eldana* (9–32%). It was found that *Chilo* larvae infested the crop at 3 weeks from emergence and continued infesting till harvest. The incidence of *Busseola* and *Sesamia* was very low. At the Nyanza Agricultural Research Station, Kisii, which is at an altitude of 1806m above sea level, *Busseola* was found to be the major stem-borer species.

During the long rainy season of 1981 in farmers' field at Ruri, *Chilo* damage started when the crop was 3 Weeks old. No other stem-borers were seen up to tenth week.

*Chilo* infestation was observed up to sixth week and the maximum was 8%. *Sesamia* attack was started at eleventh week and continued till harvest with a maximum of 5% infestation.

#### Survey of stem-borers of sorghum and maize in Kenya

A field survey on the distribution of stem-borers of sorghum and maize was conducted in Nyanza, Western Rift Valley, Central, Eastern and Coastal Provinces of Kenya. *Chilo partellus* was found to be the major stem-borer species on both sorghum and maize. In coastal areas, however, two species of *Chilo*, viz: *orichalcociliellus* and *C. partellus* were present. *Busseola fusca* was the second most important stem-borer. It appears that the distribution of stem-borer species is influenced by altitude, rainfall and temperature. In the warmer and lower altitude areas, particularly in the lake basin and coastal areas, *Chilo* is the most important stem borer. It was recovered from 70 and 5500 ft. *Busseola fusca* was found to be the dominant stem borer species in cooler and higher altitude areas, above 3,750 ft. The presence of *E. saccharina* on sorghum and maize in the sugar belt of Western and Nyanza Provinces and *S. calamistis* in many areas of Kenya except in higher altitudes were noted.

#### Pest carryover studies

In order to study the carryover of stem borer species, Serena sorghum stalks with their stubbles were stocked after harvest in February 1981. Monthly sampling of 100 stalks was carried out for seven months for the presence of larvae and pupae. Initially, a very high proportion of stalks contained larvae and pupae of *C. partellus* followed by *Eldana*, *Busseola* and *Sesamia*. The presence of *Chilo* and *Sesamia* larvae was observed up to 180 days in the dry stalks and that of *Eldana* and *Busseola* larvae up to 90 days

after stocking.

#### Light trap studies

At MPFS a pressure lantern was operated partially for 19 nights before the break of long rains. All the four stem borer species, viz: *C. partellus*, *E. saccharina*, *B. fusca* and *S. calamistis* were attracted to light and the catches were 109, 15, 7 and 5 respectively, thus confirming that *Chilo* population was the highest.

#### Screening for confirmation of resistance

Selected 140 sorghum lines which had already been screened for resistance to stem borer at ICRISAT, India were tested at MPFS for confirmation of resistance to *Chilo*. Preliminary observations showed that some lines possess a fair degree of resistance.

#### Physiology of aestivation-diapause in *Chilo partellus*

Physiological aspects of aestivation-diapause in the pyralid borer, *C. partellus* were investigated with the main objective to find out endocrinological relationships involved in aestivation-diapause. Effects of 20-hydroxyecdysone on last instar *Chilo* larvae were investigated by injecting larvae with 4ug of the hormone. It was observed that the number of diapausing larvae increased as the dry season started.

## RICE STEM BORERS

#### Borers composition

In general, rice areas in Kenya comprise of mainly lowland irrigated (LLI), upland rain fed (URF) and swamp flooded (SF) conditions. In Western Kenya, MPFS and Ahero Irrigation scheme where rice is grown under LLI condition, four species of stem borers, namely, *Maliarpha separata* Rag., *Sesamia* sp., *Chilo* sp. and *Diopsis thoracica* West. are present. Dissection of rice plants at various growth stages

revealed that though *Maliarpha* was the most abundant, it caused least 'dead-hearts' or 'white-heads'. *Diopsis* caused highest incidence of 'dead-hearts' at vegetative stage. *Sesamia* occurred at reproductive stage and caused the highest number of 'white-heads'. *Chilo* rarely occurred and caused the least damage.

In Central Province, at the Mwea Tebere Irrigation Scheme it was found out that *Maliarpha* was the most abundant followed by *Sesamia*. 'Dead-hearts' were caused by *Sesamia*.

In Coastal Province where rice is believed to have been introduced earlier than in other parts of Kenya, the occurrence of various borer species differed from the rest of Kenya. In Mazeras under URF condition, *Chilo* and *Sesamia* were as abundant as *Maliarpha* at various plant growth stages. In Ramisi, under SF conditions, only *Maliarpha* was recorded. It may be noted that the Coastal and Central locations where the surveys were done are separated by a large strip of semi-arid vegetation in hundreds of kilometres where rice is not grown and only sorghum and maize are abundant. That might be the reason for the striking differences between the distribution of stem borer in Coastal Province and the other parts of Kenya.

#### Copulation oviposition and egg mass characteristics

One hundred and ten pairs of adult *Maliarpha* were used for observations on reproduction. Adults emerged late after sunset (at about 1900 hours) and were capable of copulation a few hours after emergence. Oviposition under field conditions started at early vegetative stage and egg masses could be found throughout the growth stage of the rice crop but were most abundant at late vegetative stage. Average preoviposition period was 2 days (ranged from 1 to 6 days).

#### Post-embryonic development and larval activity

Under field conditions, dissection and marking were made to determine the duration of larval stages of the insect. Larval period ranged from 30 to 50 days depending on the varietal resistance. Soon after hatching the larva moves towards the leaf sheath, penetrates downwards into the hollow internodes. Larvae can survive only if the stem has been differentiated with internal hollow. Older larvae and pupae were found at the lower internodes at ripening stage. Larvae are gregarious and can survive in stems previously infested with other borer species. Combined infestation with *Sesamia* caused high incidence of 'white head'. Pupal period was about 14 days.

#### Adult longevity, fecundity and sex ratio

Average longevity of the male and female adults was 3 and 4 days respectively. Each female on average produced 2 egg masses with 50 eggs each. Hatchability was 81% and incubation period was 8 days. Light trap and field collected data revealed that the sex ratio is very close to 1: 1.

#### Population dynamics

In both long and short rainy seasons, oviposition started before tillering stage and reached a peak at vegetative stage (from 45 to 60 days after transplanting, (DT). Larval population reached its peak at flowering stage (75DT). Tiller damage was highly correlated with larval population. Per cent 'dead heart' was very low although a maximum of 70% of the tillers were infested in IR579-48-1 and Sindano. The economic importance of this pest is questionable because, the damage appears to reduce the vigor of the plants without causing 'dead-heart' or 'white head'. The peak of pupal population occurred at 90 DT indicating that *Maliarpha* would complete only one generation on short duration rice varieties.

### Effect of nitrogen fertilizer on stem borer infestation

Preliminary studies showed that 'dead heart' incidence was maximum (9.4%) at tillering and flowering stages of the rice variety Sindano applied with 60kg N/ha nitrogen, and was significantly higher than that of the crop without N fertilizer (0.9%). Further, the effect of nitrogen fertilizer was not only on increase in total incidence of damage but also in distribution pattern of the stem borer species.

### COWPEA POD-BORER, *MARUCA TESTULALIS* GEYER

#### Population studies under pure and mixed crop situation

It was observed that on cowpea (local variety — Nyar milambo) under both pure and mixed crop, *M. testulalis* was the most abundant borer accounting for more than 90% of the total borer population at all sampling stages. Among other borers, the Lycaenid butterflies, especially *Euchrysops* spp. and *Lampides* spp. were most prominent at MPFS and the farmers' fields. These studies revealed that population of *M. testulalis* is lower in pure than in mixed crop and that the mixed crop ecosystem might be favourable for the pest's survival.

#### Studies on pest-host relations and yield loss assessment

Work was initiated at MPFS to study the development of cowpea plants and determine damage/injury levels or thresholds of *M. testulalis* infestation on cowpea. The purpose was to understand the partitioning process in cowpea, i.e. how it channels its energy in coping with the consistently changing biotic and abiotic relationships in the agro-ecosystem, as a basis for establishing damage thresholds or indices for managing the pest complex of the crop.

Phenological studies showed that pods

matured in an average of 30 days in field plants; only 29 to 30% of flower buds resulted in mature pods, i.e. up to 70% of flowers and pods arising from buds are 'wasted'.

Results of this preliminary trial on field infestation and yield loss assessment suggested that:

- (i) The number of pods and pod weight per plant, pod dimensions, number of seeds per pod and shelling per cent were comparable among treatments
- (ii) Pod deformation caused by sap sucking insects and/or plants stress, was much higher when related to numbers of pods (50–60%) but was considerably lower (less than 20% compared to 35% in infested and non-infested plants, respectively) in relation to pod weight.
- (iii) An assessment of pod sampled showed that only 7 and 6% seed damage resulted from infestations of 4 and 5 *M. testulalis* larvae, respectively per plant.

### GENETICS OF HOST PLANT RESISTANCE TO INSECT PESTS

It is envisaged to work out the genetic of host plant resistance to sorghum shootfly, *A. soccata* in sorghum; to stem borer, *C. partellus* in sorghum and in maize; to rice stem borer, *M. separatella* in rice; and in cowpeas to pod borer, *M. testulalis* and aphids. During the long rainy season at MPFS, efforts were made to screen some cowpea cultivars for resistance to aphids and *Maruca*, and some sorghum cultivars to sorghum shootfly and stem borer. The susceptible and resistant cultivars identified will be used in crosses for genetic studies.

#### Sorghum shootfly

On the basis of 'dead-heart' (count on

the main stem and tillers) some resistant cultivars identified were: IS 2146, IS 5613, IS 1044, IS 4660, IS 18361, IS 18363 and IS 18427. The most susceptible were Serena, IS 1522, CSH-1, IS 8595.

#### Stem borer, *C. partellus*

The same set of 25 sorghum cultivars were also evaluated for stem borer resistance. Based on the tunnel length, the cultivars such as IS 5613, IS 1044, and IS 18489 were found resistant to *Chilo* damage. The cultivar IS 8595 was found to be the most susceptible followed by IS 1522 and local variety Serena.

#### Aphids

A severe natural infestation of aphids during long rainy season at MPFS made it possible to screen a total of 275 cowpea cultivars. Among the local cultivars Emma -- 60 was found to be resistant to aphids. Other local cultivars such as Katuli-107, Katuli-108, Machakos-66 and Machakos-68 when attacked, showed total damage. Some of the IITA cultivars, namely Tvx 66-2H, Tvx 337-3F, Tvx 33-iJ and Tvx 2394-02F were found resistant though late in maturity. It was encouraging to note that some F<sub>4</sub> progenies of the cross involving Emma-60 with IITA cultivars were found resistant to aphids and appear promising from grain yield and early maturity. Among other Tvx 1999-01F was identified as highly susceptible to aphid attack.

#### Pod borer

*Maruca* infestation was also very severe during the long rainy season of 1981. The pod infestation ranged from 25 to 100% under natural field conditions. The IITA cultivar Tvu 946 appears promising as it showed the lowest pod infestation of 25%.

Tvu 946 being the earliest in flowering and maturity, may have escaped the pod borer attack and hence needs confirmation of resistance. The local high yielding cultivar Katuli-108, and Emma-60 appear to possess some tolerance. Most of the selected IITA cultivars were found to be highly susceptible to pod borer attack. It may be encouraging to note that some F<sub>4</sub> progenies involving Emma-60, Tvu 1509 and other cultivars appear promising for resistance to *Maruca* attack and show high yield potential and earliness.

### INTERCROPPING EXPERIMENTS

Pest complex with ecosystem of maize, sorghum and cowpea

The main objectives of this experiment are:

- (i) to standardize sampling procedure in intercropping experiments;
- (ii) to assess pest complex within mixed ecosystem of maize, sorghum and cowpea; and
- (iii) to test the influence of the special pattern of different plant species of the pest abundance within intercropping system.

The experimental material included the early maturing cultivars of maize (Katumani), sorghum (serena) and local cowpea (ex-Luanda). They were planted in monocrop, dicrop and tricrop combinations. Target insect pest species monitored were categorized into three: specialized feeders, *M. testulalis* and *A. soccata*; relative specialist feeders, *B. fusca*, *C. partellus*, *S. calamistis* and *E. saccharina*; and general feeders, *Spodoptera littoralis*, *Heliothis armigera* and aphid sp.

## AFRICAN ARMYWORM RESEARCH PROGRAMME

This has remained at low ebb during the year with no new investigations started; and the remaining two scientists winding up their work, preparatory to expiry of their contracts at the end of the year. Although the study of techniques for determining the physiological age of armyworm moths did not reach completion, it has provided the basis for estimating the age of female moths obtained in the moth trap network, widespread in many African countries, and with further refinement it is being used to provide consistent results for moths up to 48 hr old. It gives an indication of the distance of moths from the source based on various assumptions derived from other studies of moth flight. The most useful parameter is the width of the developing ova in the proximal follicles of the ovarioles.

*Cynodon dactylon* grass is superior to all other host plants for *Spodoptera exempta*. *Cynodon dactylon* is the preferred

grass in choice experiments in the laboratory and in mixed pastures in the field. Star grass is the natural habitat for low-density populations of armyworm caterpillars, and the parts of grasses with high nitrogen levels are preferred. This may have some importance in understanding the common occurrence of outbreaks of caterpillars on new flush of grasses with high nitrogen content; and it also raises speculation about the consequences of applying nitrogenous fertilizers to cereal crops.

Field surveys by scientists in collaborating institutes and organizations, (COPR, KARI and DLCO-EA) have shown that armyworm moths and caterpillars are present during the off-season in Kenya, particularly in the *C. dactylon* pastures in the uplands where grasses are green throughout the year. Population studies are therefore necessary to measure their abundance and distribution and to assess their significance.

# LIVESTOCK TICK RESEARCH PROGRAMME

## INTRODUCTION

Ticks are found in all areas of Africa which are suitable for livestock including approximately 10,000,000 sq km presently infested with tsetse, and livestock production is seriously affected because of disease transmission and debility caused by tick infestation.

The most important diseases transmitted to cattle by ticks are theileriosis, anaplasmosis, babesiosis and rickettsiosis all of which occur throughout the continent. Theileriosis caused by *T. annulata* occurs in North Africa and extends into Sudan. Theileriosis caused by *T. parva* and *T. lawrencei* occurs in East, Central and parts of Southern Africa. Drugs and methods of vaccination have been available for sometime for the control of anaplasmosis, babesiosis and rickettsiosis. As a result of work carried out by the UNDP/FAO Regional Project (Research on tick-borne diseases and tick control) RAF/67/077 from 1967 to 1977 at Muguga in Kenya, and since then continued at the Veterinary Research Department of the Kenya Agricultural Research Institute (KARI) and at ILRAD Nairobi, curative drugs and experimental vaccine against East Coast Fever (caused by *T. parva*) have been produced. Successful field trials with the experimental vaccine have been carried out in Kenya and Tanzania, and plans are in hand to carry out extensive trials in Western Kenya and in coastal province. In addition, a Danida/FAO project has been established in Malawi which is expected to produce ECF vaccine for Malawi and neighbouring countries. A successful field trial has recently been carried out by the staff of the Veterinary Research Department, Muguga, in Western Kenya using two curative drugs against East Coast Fever.

The traditional method for controlling ticks and the diseases they transmit is the close interval application of acaricides in

dips or sprays to livestock. To control East Coast Fever (ECF), two weekly applications are necessary. This procedure has many disadvantages: the high cost of installing, maintaining and staffing dips and spray races, the high cost of acaricides, the development by the ticks of resistance to acaricides, high levels of acaricide residues in beef and dairy produce. But perhaps the greatest disadvantage of using acaricides for the control of ticks is the fact that an inherently unstable situation results where regularly dipped cattle are completely susceptible to disease and naive to tick infestation. If for any reason acaricide application fails, catastrophies can occur. This happened recently in Zimbabwe on a very large scale when acaricide control broke down as a result of hostilities, and more than 1,000,000 cattle died of tick-borne diseases and debility caused by massive tick infestation.

At a recent conference on theileriosis at ILRAD in Nairobi, attended by delegates from many countries in Africa and elsewhere, concern was noted regarding the present situation of using acaricides to control ticks and great support was given for the development of biological methods to control ticks, to be used in conjunction with curative drugs and vaccines against the tick-borne diseases. It was considered that such an integrated approach would create a stable situation which would obviate most of the disadvantages associated with the use of acaricides.

What are the prospects? As stated above, curative drugs and vaccines are now or shortly will be available for the control of the diseases of cattle transmitted by ticks. In addition, it has been known for many years that cattle can be made resistant to ticks, and the technique is now being used in Australia to control *Boophilus microplus*, the vector of babesiosis.

Bovine resistance to tick infestation was reported in the early part of this century by Johnson and Bancroft (1918) who found that cattle in Queensland, Australia developed resistance to *B. microplus* following infestation. However, it was only within the last 10 years that this finding has been applied in the field, necessitated by the widespread development of resistance to acaricides by *B. microplus* and made possible by the availability of curative drugs and a vaccine for the control of babesiosis. Over the years, a great deal of work has been carried out on this subject but very little in Africa.

It has been confirmed that cattle which develop resistance to tick infestation can control field populations of *R. appendiculatus*. The reduced numbers of larval and nymphal ticks which engorge on resistant cattle, moult into nymphal and adult ticks which are smaller than ticks which feed on non-resistant host animals. These stunted ticks have a reduced survival potential when exposed to temperature and humidity stress.

When resistant cattle are exposed to ticks infested with East Coast Fever, fewer parasites are transmitted, and a less severe disease reaction is produced. Furthermore, when transmission of the parasite is attempted by applying ticks to infected tick resistant cattle, fewer ticks become infected, and since these stunted ticks have a reduced survival potential, the field challenge is greatly reduced. Thus the use of tick resistant cattle in ECF vaccination programmes will probably enhance the efficacy of vaccination by reducing transmission of the parasite to and from the exposed cattle.

Antigens have been identified from engorged tick homogenates which have been used to produce antibodies in rabbits. Ticks which feed on these rabbits have 80 – 90% reduction in egg viability. It is anticipated that when these antigens are inoculated into cattle which have deve-

loped resistance following tick infestation, that an improved resistance will be produced for the control of field populations of *R. appendiculatus*.

During the year (1981), the Tick Research Programme has collaborated with the Chemistry and Biochemistry and Sensory Physiology Research Units at the ICIPE on various aspects of research. Soon trials will be carried out in co-operation with Insect Pathology and Pest Management Programmes to demonstrate the use of resistant cattle to control field populations of ticks.

There has also been collaboration with the Veterinary Research Department of the Kenya Agricultural Research Institute (Dr. A. Young, Dr. T. Dolan and Mr. B. Leach); the International Laboratory for Research on Animal Diseases (Dr. A. D. Irvin, Dr. S. Shapiro and Dr. M. Murray); the FAO Sheep and Goat Project Veterinary Laboratory, Kabete (Dr. E. Allonby; Dr. J. Allonby).

#### CATTLE RESISTANCE TO TICKS

Investigations were started at the ICIPE in an attempt to produce resistance in cattle to ticks. All of the work so far has been done on *R. appendiculatus* because of its importance as a vector of *T. parva* and because of its widespread distribution in East, Central and Southern Africa.

Following the Australian work, it was quickly found that cattle become resistant to *R. appendiculatus* when approximately 500 adult ticks are allowed to feed on them. Resistance also develops when cattle are exposed to ticks in paddocks. Resistance has been produced in *Bos taurus* as well as in *B. indicus* cattle, and is long lasting – at least 2 years.

Resistance appears to be stimulated in cattle in response to antigens inoculated in the saliva of the feeding tick. When

ticks feed on a resistant animal, they excrete antigens in the saliva. These antigens stimulate an immediate type hypersensitivity reaction, and marked swelling occurs at the site of attachment of the tick, within 20 min of attachment. This reaction interferes with the ability of the tick to feed properly. The effect is most marked against larvae, less against nymphs and least against adult ticks. In highly resistant cattle, very few larvae, less than 25% of nymphs and less than 50% of adult ticks are able to complete feeding, and the engorged weight is also reduced. The smaller ticks which are produced after moulting have a reduced survival potential when exposed to temperature and humidity stress and the females produce smaller egg batches.

Two experiments have been carried out which demonstrate that resistant cattle can control field populations of *R. appendiculatus*:

- (1) Individual tick-naive cattle were exposed in each of five paddocks infested with ticks. Initially, the tick populations in each paddock increased and reached a peak after 9 months, when hundreds of adult ticks were seen on the cattle. Thereafter, the ticks decreased in number and after 2 years had almost disappeared. On removal from the paddocks the cattle were found to be highly resistant.
- (2) In the second experiment, a pair of resistant cattle were exposed in a paddock infested with larval ticks, and pairs of tick-naive cattle were exposed successively in a similar paddock. Thousands of adult ticks were produced in the tick-naive cattle paddock and very few adults in the tick-resistant cattle paddock.

Another important finding is that tick-resistant cattle infected with ECF, greatly affect the infection rates in larvae

or nymphae which feed on them, resulting in reduced transmission of the disease. It is probable that resistance to ticks in cattle will enhance the efficacy of vaccination against ECF by reducing challenge.

Another approach to produce resistance in cattle depends on the observation that bovine gamma globulins ingested by ticks pass unchanged from the tick blood meal into the haemolymph (Galun, 1975). When target antigens from the ticks are inoculated into rabbits or cattle, antibodies are produced against the target antigens. When ticks are fed on these animals, a high tick mortality results and the reduced numbers of female ticks which do engorge, either do not lay eggs or produce eggs which have markedly reduced viability.

Finally, it has been established that cattle quickly become resistant to infestation with *R. appendiculatus* after a relatively small number of adult ticks have fed on them and it is probable that this resistance can be enhanced by inoculation of target antigens from ticks into host cattle. Furthermore, it has been demonstrated that when resistant cattle are allowed to graze in *R. appendiculatus* infested paddocks, and no other host animals are available for the ticks to feed on, the tick population falls to very low levels and might even disappear. As a result of these findings, the ICIPE group has been invited to cooperate with colleagues from the Kenya Agricultural Research Institute (KARI), and from ILRAD to carry out trials using tick resistant cattle, vaccinated against East Coast Fever and exposed to challenge in enzootic areas in Western and Coastal Province in Kenya.

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# THE GRASSLAND TERMITES RESEARCH PROGRAMME

## INTRODUCTION

This programme had an active year, although some aspects of the programme were being phased out towards the end. Collaborative help was received from the Chemistry and Biochemistry Research Unit for several aspects of the programme.

The most interesting finding is the profound differences between our two target species, *Macrotermes michaelseni* and *M. subhyalinus*. Although they are morphologically almost indistinguishable, it has become clear that they are ecologically very different. This has shown up both in the work on populations and in the measurements of foraging offtake. The mounds built by the two species are quite different in structure, but the properties of their constituent soils are quite similar.

As the programme is closing down early in 1982, all aspects are either already complete or are being finished off and written up. The stated aims of the programme have been achieved, resulting in an unprecedented understanding (in quantitative terms) of the ecological role of the target insects in semi-arid grasslands, their physiology and development, and the role of pheromones in coordinating and regulating their social behaviour.

## POPULATION STUDIES

The long-term study of *Macrotermes michaelseni* populations was completed by measuring the duration time of juvenile stages in mature nests from which the royal pair had been removed. The results showed that larval development time in mature nests was about twice as long as in laboratory incipient colonies. It was thus possible to estimate the productivity of a mature nest, for which figures on mean population and biomass were already

available.

Comparative study on populations of mature nests of *M. subhyalinus* was also completed this year. The total population was about the same as in *M. michaelseni*, but the proportion of larvae in the population was much lower (about 25% compared with 45% in *M. michaelseni*). As a result of this and the higher mean weight of the adult castes, the nest biomass is nearly twice as great in *M. subhyalinus*, while the production is much lower.

## FORAGING ACTIVITIES OF TERMITE SPECIES

Measurements of foraging offtake by the two species also showed substantial differences. There were 2–3 times as many foraging holes opened per day per m<sup>2</sup> where *M. michaelseni* was the dominant species. The amount of grass litter taken overnight per hole opened, was found to be 0.28g dry weight for *M. subhyalinus* and only 0.17g for *M. michaelseni*, but because of the higher density of holes the overall foraging offtake by *M. michaelseni* was nearly twice that of *M. subhyalinus*. *Macrotermes michaelseni* shows two weak peaks in foraging offtake per year while *M. subhyalinus* shows only one, during the long dry season (Fig. 1). Thus, *M. subhyalinus* is a less active forager than *M. michaelseni* both in offtake per unit area and in weight specific consumption. This may be an adaptation to areas of low rainfall and grass production where *M. subhyalinus* is usually dominant. Conversely, *M. Michael-seni* predominates in areas of relatively high rainfall and production.

Measurements of grass standing crop and litter production showed that the maximum biomass of litter coincided with the peak in foraging activity of *M. subhyalinus*. Trampling by cattle increased the production of litter from

standing grass, and this may contribute to an increase in termite activity and population in heavily stocked pastures.

Trail-laying by foraging *M. michaelsoni* in the field was investigated. A pheromone trail was laid to a food source, which retained its activity for about 1hr when exposed to the atmosphere. When in use by foraging termites, such a trail would be continually reinforced. Similar trails were detected in the underground passages leading from the nest to the foraging site, which would presumably retain their activity longer in that enclosed air space.

The trials were laid mainly by major workers, which are the main foraging caste. They have the largest sternal gland of any adult caste, this gland being the site of production of the trail pheromone.

#### DEVELOPMENT OF PHYSOGASTRY

Development of physogastry in the queens of *M. michaelsoni* and corresponding changes in the kings, were studied in incipient colonies, in young field colonies, and in mature colonies in which replacement reproductives had been induced. It was found that the rate of development of physogastry was greatest in the large, mature colonies and least in the very small incipient colonies, and is therefore a function of colony size.

#### CASTE DIFFERENTIATION IN COLONIES

Work was continued on caste differentiation in incipient colonies. Determination of the soldier castes takes place in the third larval instar and is associated with increase in size of the corpora allata and the prothoracic gland. Any female larva has the competence to develop into a soldier if it is acted upon by hormones during the first half of the third instar. The hormones responsible are a juvenile hormone and ecdysone. Work is in progress to identify the type of juvenile

hormone, and to establish the quantities of both hormones present in a larva when the differentiation of soldiers is taking place. The probability of becoming a soldier is positively related to the size of the colony, and may be negatively related to the number of soldiers and pre-soldiers already present (thus forming a feedback loop).

#### PREDATION OF TERMITES

The extensive field studies on predation by the ponerine ant, *Megaponera foetens* (reported in earlier Annual Reports) were successfully completed. Offtake by the ants was estimated at about 2% of the termite production, but was not evenly distributed. The proportion of the worker production taken was very low, estimated at 25% of minor soldiers and 75% of major soldiers; taken by this one predator. As a follow-up laboratory experiments were undertaken to test the ants preference for different castes of termite, when offered freshly killed. The ants then took a much larger proportion of workers than they did in the field. This suggests that the termite soldiers are effective in protecting termite workers from attack by ants.

A study was carried out on the calliphorid fly *Bengalia* sp. which as an adult is a predator on termites and as larva a parasitoid on the alates of *Macrotermes* and *Odonotermes* spp.

#### SOIL TEXTURE OF MACROTERMES MOUNDS

*Macrotermes* mounds are composed of fine-texture, clay-rich soils in comparison to the surrounding soil. The organic matter content is slightly greater, perhaps because the termites add saliva as a cement. They also contain more calcium, potassium and nitrogen, and in dry areas also more available phosphorus. As a result, the soil washed down from mounds and forming a pediment around them is more productive than the surrounding soil and carries a much higher grass biomass. Also, the few

species of grass which flourish around the mounds are more palatable and contain more nutrients than the larger number of species that grow between the mounds

Thus the termite-altered soils contribute disproportionately to the food supply of the grazing mammals.

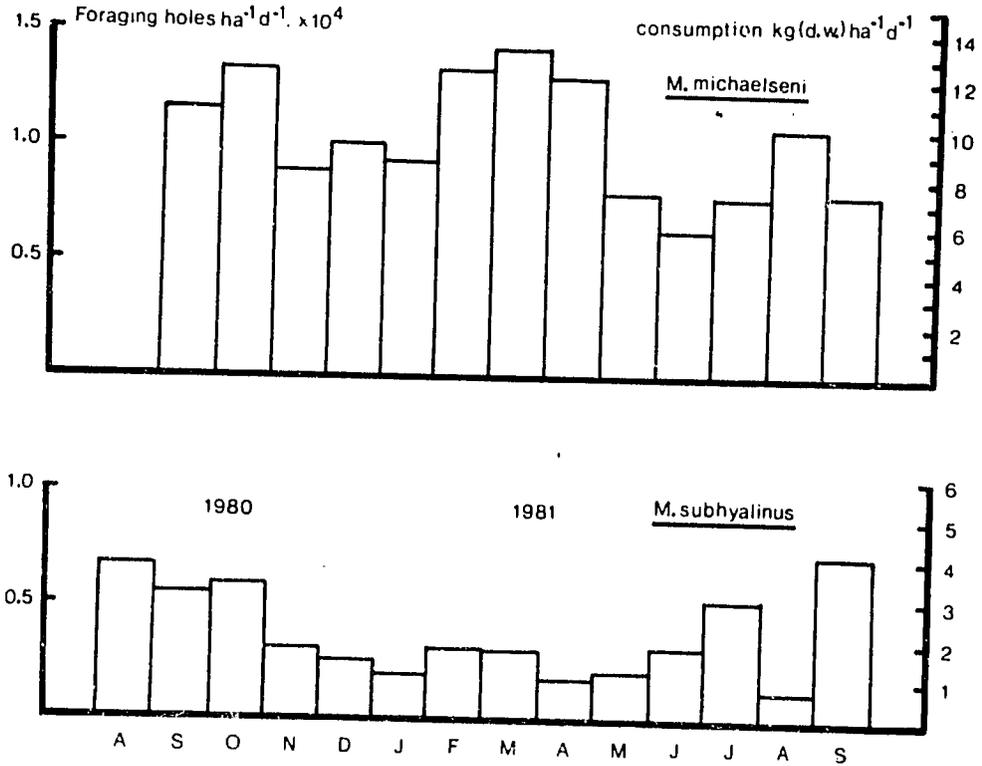


Fig. 1. Comparison of foraging activity and offtake by two species of *Macrotermes* in Kajiado district

# TSETSE RESEARCH PROGRAMME

## Introduction

During 1981 the Tsetse Research Programme pursued its activity through its three projects: Ecology, Reproductive Physiology and Trypanosome-Vector Physiology.

Studies of the ecology of *Glossina pallidipes* on the South Kenya Coast have continued; analysis of the bulk of data collected for several years is underway in order to elucidate mechanisms of population fluctuations in relation to density-dependent and density-independent factors. The studies undertaken in the Lambwe Valley since two years have been interrupted due to tsetse control measures applied to stop an outbreak of sleeping sickness; however, in collaboration with the authorities, an entomological evaluation of the effectiveness of spray operation was made on a monthly basis. Trapping trials have continued using cylindrical revolving screens as attachment to the biconical trap to improve trap performance.

In Reproductive Physiology Project, experiments have been pursued to investigate the role of Juvenile Hormone (JH III) in the control of egg maturation, and the effect of precocene on corpus allatum. Studies have been undertaken on the effect of feeding males and females on rabbits immunized against various reproductive tissues from males. In addition, preliminary observations have been made on immunomechanisms in tsetse.

In the Trypanosome-Vector Physiology Project, experiments have been carried out in order to determine the part played by temperature and number of Trypanosoma temperature and number of *Trypanosoma (Trypanozoon) brucei brucei* in the vectorial capacity of *G. morsitans morsitans*, and the effect of proteolytic enzyme on the survival of trypanosomes ingested by young

tsetse. Studies on the sensitivity of trypanosome-infected *G. m. morsitans* to toxic substances have been extended to include female flies and natural pyrethrin.

After 4 years of successful *in vitro* cultivation of *T. brucei* studies now concentrate on the metabolism of this microorganism. Epidemiological surveys have been carried out in the Lambwe Valley after the recent outbreak of sleeping sickness, while on the South Coast, patterns of trypanosome infections in *G. pallidipes* have been assessed in various habitats with different host availabilities.

## TSETSE ECOLOGY

### Tsetse ecology on the Kenya coast

Studies of the ecology and behaviour of *Glossina pallidipes* on the south Kenya coast have continued. Emphasis has been placed on the investigation of the population dynamics of this tsetse through a comparison of the characteristics of population at different localities and detailed monitoring of selected populations. Many aspects of this work are a continuation of preliminary investigations in the ICIPE and this study has now entered a phase of consolidation and analysis of field data.

The investigations have been aimed at an understanding of the processes involved in the natural regulation of numbers of tsetse at different localities on the south Kenya coast: why population densities differ at different localities and what factors govern the fluctuations of these populations. Mortality factors act at all stages of the tsetse life cycle, but the studies to date have only considered what is happening to adult tsetse and in particular to mature females since these are the productive members of the population.

The sampling project was based on the

use of biconical traps, which capture *G. pallidipes* in large numbers. Continuous,

The sampling project was based on the use of biconical traps, which capture *G. pallidipes* in large numbers. Continuous, monthly — 4 days in 28 days — observations were completed for 33 months at Muhaka and 21 in Shimba Hills. Occasional or shorter series of observations were also made at Diani, Mwalewa and Ukunda.

When the relative density, survival rates and month to month fluctuation in relative density of the different tsetse populations are considered, a number of points suggested in previous reports, have become clear with the accumulation of more data. Fluctuation in numbers of males and females roughly parallel each other, minor differences may be due to the shorter life-span of males. Population changes at different localities are not synchronised, despite the fact that the study sites are very close with marginal climatic differences between them. Each population has its own characteristic equilibrium level although there may be considerable fluctuations about this mean population density. This suggests that factors determining the equilibrium level of each population may be operating at other stages of the life cycle of this tsetse. Although mean population densities differ significantly between sites, mean survival rates are virtually the same. There is a relationship between population changes and the survival of adult female *G. pallidipes* at Muhaka, although this relationship is less clear for other localities

Mortality of tsetse population is affected by abiotic and biotic factors. The abiotic or climatic factors mainly act independently to density. Their effect is often termed Density — Independent Mortality (DIM). As noted in previous reports, there is a relationship, at Muhaka, between rainfall and change in population density. A rainfall between 50 and 200mm for 28 days, creates optimum conditions and the

population almost invariably increases. With more or less precipitation, numbers generally decline, with higher adult mortality. Although the main climatic effect is almost certainly through saturation deficit rather than rainfall, these observations do indicate that climatic factors, acting as DIM can contribute to the fluctuations in apparent density of tsetse at the study sites.

The biotic factors — predation parasitism and competition, act with increasing severity as population density increases and their effective is described as Density - Dependent Mortality (DDM). There is a relationship between change in density and apparent density of the *G. pallidipes* population at Muhaka. This indicates the density relatedness of some of the components involved in the regulation of adult tsetse population on the Kenya coast. The relationship between apparent density and survival of a population, shows that survival rates are optimal about the mean population level (Fig. 1). Lower survival rates at higher densities indicate DDM factors coming into play. However, they appear to come into operation at a different density at each locality, suggesting that these processes are different, or at least operate in a different way at each site.

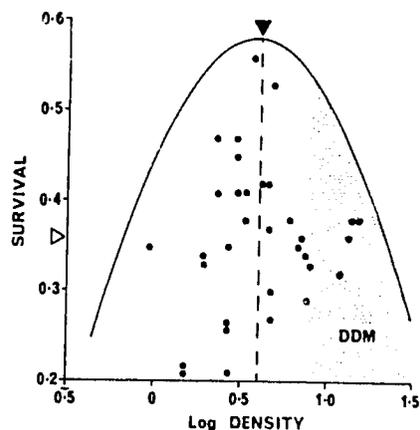


Fig. 1. Relationship between log apparent density and survival of female *G. pallidipes* at Muhaka. Lower survival rates at the higher densities indicate the influence of density dependent mortality (DDM) factors.

Although the fluctuation in numbers of adult tsetse can, to a large extent, be explained on the basis of the results outlined above, they cannot account for differences in population density of *G. pallidipes* at different sites. However, if young adult tsetse are considered, two categories can be recognised in females before their first ovulation: Oa teneral and ob non-teneral. These tsetse are poorly represented in biconical trap catches and the numbers in these two categories do not reflect their true relative numbers. Nevertheless, the difference between the two age groups if sampling biases are the same will give an indication of the relative survival from Oa to Ob at different localities. There is a clear inverse relationship between the proportion of Oa females in the catches and apparent density. Highest mortality rates are observed in the lowest density population. Newly emerged tsetse have limited food reserves and are very susceptible to stress. Host animals seem less abundant in areas with low tsetse densities and this factor, resulting in a greater delay between emergence and finding a first blood-meal, may be critical for the survival of newly emerged flies. This is the first indication of a factor which could explain differences in population size between different localities.

A number of mark-release-recapture trials have been carried out on the *G. pallidipes* population at Diani. This semi-isolated tsetse population is associated with 10ha forest relic. Although wild pigs and small antelopes are still present, a single small herd of cattle is occasionally grazed around the forest edge. The results of the mark-release-recapture experiments are providing data on tsetse population size in relation to apparent density assessed from the catch per trap per day and true sex ratios which seem similar to those observed in the trap catches. An attempt has been made to integrate these ecological techniques with the epidemiological work to estimate the number of trypanosome-infected bites received by the cattle, grazed around

this forest. It was estimated that each cow received from *G. pallidipes*, one infective inoculum of *T. congolense* every 5.8 days during the first experiment and 4.1 days in the second.

The ecology of *Glossina pallidipes* Aust. in the Lambwe Valley, South Nyanza

The planned long term study of the ecology and behaviour of *G. pallidipes* in the Lambwe Valley, started in August 1979, was terminated prematurely in February 1981 when the Kenya Government Authorities, in response to a serious epidemic outbreak of Rhodensian sleeping sickness in the valley during 1980, undertook measures aimed at eradicating the vector from the valley and its environs. Operations were based on sequential aerial applications of endosulfan aerosol, backed by ground spraying of residual dieldrin and bush clearance in areas of difficult terrain. While eradication was not in the end achieved, the population overall was reduced by over 99.5% which effectively precluded further studies of an ecological nature. The data obtained to date, relating to population dynamics feeding behaviour and mark-release-recapture studies, are now in the process of being analysed.

In collaboration with the Authorities, an entomological evaluation of the effectiveness of spraying operations was made by monthly sampling, using biconical traps, in three localities characterising the principal habitat types of *G. pallidipes* in the Lambwe Valley: thicket, acacia woodland and coniferous plantation. On the basis of the standard evaluation technique of ovarina age-grading of female flies caught in relation to the timing of spray applications, it was found that adults were unaffected by sprayings in all three localities, and that the measures were particularly ineffective in the coniferous plantation. This was because of the technical problems associated with aerial spraying in the hilly terrain on which the plantation is located and the difficulty of achieving adequate

spraying droplet penetration beneath the canopy of the conifers.

Post-spray monitoring to date (September, 1981), indicates that the population is recovering, albeit slowly at present. In the possible absence of future anti-tsetse measures, monthly sampling will continue in order to assess the rate of recovery of the population, and to predict duration of full re-establishment.

In view of the unlikelihood of the population recovering in the short term and the unpredictable epidemiological situation in the Lambwe Valley, it has been decided to transfer the operations of the Lambwe Valley Tsetse Ecological Research Team to the Nkruman *G. pallidipes* fly-belt near Lake Magadi in the Rift Valley Province. Steps are underway to establish the necessary basic support facilities there.

Our field and laboratory investigations in collaboration with the Insect Pathology Unit and the Fine Structure Unit) on the phenomenon of cuticular lesions on *Glossina* species have been concluded. Lesions present on the ventral abdominal integument are common among tsetse in Kenya: occurring in all species (*G. pallidipes*, *G. swynnertoni*, *G. austeni*, *G. fuscipes*, *G. brevipalpis* and *G. longipennis*), and in all populations of species sampled from areas widely apart. While considerable variation exists in number and size of lesions, four or five basic forms are distinguished: small pits; raised, irregular-shaped scabs or warts; tumour-like eruptions of soft cuticle with a hard black melanotic core; around shallow disc and flat, plate-like forms; and necrotic tracks or striations, running anterior-posteriorly. Between species, the occurrence of lesions ranged widely from 3 to 72%. Monthly monitoring of *G. pallidipes* populations in the Lambwe Valley over twelve months indicated that sex, seasonal and locality differences in incidence of lesions were related to the population age structure, being more common in older flies. Microbiological

and histopathological studies have failed to implicate bacterial, fungal or viral micro-organisms in lesion formation nor was there any evidence of transmissibility when purified suspensions of macerated cuticular lesions were applied by various pathways to laboratory-reared, newly emerged *G. morsitans*. Lesion formation and subsequent melanisation apparently result from integumental injury caused by non-infectious agents in the natural environment. Instances of penetration of the integument by foreign bodies have been observed in wild flies, and most forms of lesion can be induced by artificial wounding in the laboratory.

#### Revolving traps for *Glossina pallidipes* Austeni

Revolving screen around the base of the biconical trap has been developed and used for sampling *G. pallidipes*; and that a range of speeds, namely 15 revolutions per minute (rpm) 20 rpm, 35 rpm, and 40 rpm had been used. A similar trap with screens revolving at 20 rpm showed promise in capturing tsetse of age grades that were closest to theoretical lifetable structures; although the total yields of the trap were less than those of the ordinary stationary trap. These observations are based on one study area only — Nkruman escarpment in the Rift Valley.

When the flat revolving screens were replaced with cylindrical ones, and tested, stationary devices, with flat or cylindrical attachments gave higher catches than any other. The stationary flat screen yielded even higher numbers than the ordinary standard biconical trap. The total yields decreased with the speed of revolutions and 75 rpm gave the lowest numbers. However, the 20 rpm performed better than the 15 rpm trap.

The age structure of females from all stationary traps was similar with a large number of old flies. High speeds of revolution, e.g. 40 rpm and 75 rpm considerably

improved catches of very young flies, but not those of older females. The optimum for getting a life table-like curve of age grades was still 20 rpm (intervals), and 35 rpm both of which had flat screens. Speed 20 rpm has not been used on cylindrical screens yet. Flies caught by traps with cylindrical screens at speeds (40 rpm, 75 rpm and stationary) are not significantly different in age structure from those by traps with flat screens at the same speeds.

The highest number of females carrying fully developed larvae with black breathing lobes were captured by stationary traps of both flat and cylindrical screens. The proportion of females with empty uteri due to both abortions and after larviposition was rather high.

There was no significant difference in fat reserves between the individuals captured in revolving flat screens and revolving cylindrical screens. However, flies from stationary traps had lower fat reserves (not significant) than those from revolving traps.

## REPRODUCTIVE PHYSIOLOGY

### Control of egg maturation in *Glossina morsitans morsitans*

The corpus allatum (CA) produces the juvenile hormone (JH) in insects. In the adult state of most insects, JH is the gonadotropin that stimulates oocyte maturation, particularly the vitellogenesis. In some insects, the egg development neurosecretory hormone (EDNH) stored in the neurohemal organ, corpus cardiacum (CC), acts as a gonadotropin. Available evidence of the role of JH and neurohormones in the egg maturation process of *Glossina* is rather scanty and confusing. Allatectomized females produce at least some progeny before they stop giving birth. Ablation of median neurosecretory cells of the brain has little effect on the rate of egg maturation in *G. austeni*. Additionally, in the maturing oocyte of *G. austeni*, spaces

between the cells of the follicular epithelium do not appear during the vitellogenesis. In most insects studied, the appearance of vitellogenetic protein, is regulated by JH. Thus it appears that, unlike in most other insects studied, the vitellogenesis is independent of JH in *Glossina*. What then is the role of JH in the egg maturation process of *Glossina*. And how do the brain neurohormones influence the development of tsetse egg?

Results of our experiments showed that the females allatectomized 12hr after emergence developed mature follicles in all four ovarioles in about 37 days. About 40% of females which were allatectomized 6hr after emergence failed to develop mature follicles in at least two ovarioles (third and fourth) of most of the insects. These eggs did not develop past previtellogenic stage. In contrast, flies that had their CA-CC complex removed 12hr after emergence failed to mature the third and fourth eggs. These eggs completed the previtellogenic stage but did not undergo yolk incorporation. Females allatectomized at 1hr after emergence and subsequently treated with JH III developed the third and fourth eggs in about 30 – 70% cases, respectively. In contrast, JH III treatment of flies lacking the CA-CC complex did not produce any more eggs than the control insects.

It is therefore clear that JH is not required for vitellogenesis in *G. m. morsitans* but is perhaps an important factor in previtellogenic development of the oocyte. The JH probably activates a particular stage during previtellogenesis and once the follicle is activated and allowed to pass the JH dependent stage, the vitellogenesis proceed. A neurosecretory factor from the neurohemal organ of tsetse (comparable to EDNH) probably induces the final stage of previtellogenesis to undergo vitellogenesis.

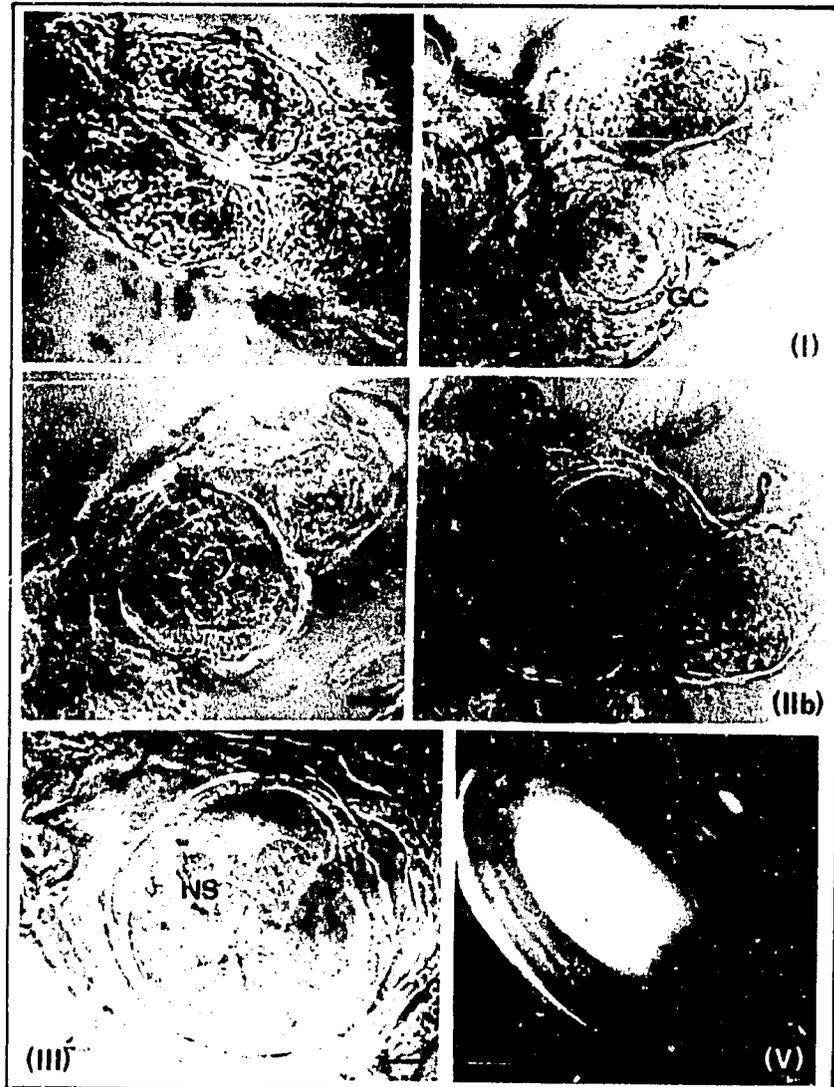


Fig. 2. Phase contrast micrographs showing stages in follicle development. Figures in parentheses indicate stage of development.

- (0) Only germarium is present and follicle absent. Scale – 12.5  $\mu\text{m}$
- (I) Undifferentiated germarial cyst descends into vitellarium forming a stage 1 follicle which is still attached to the germarium by a stalk (arrow). Scale – 12.5  $\mu\text{m}$
- (IIa) Follicle contains dividing cells; nurse cells visible. Scale 12.5  $\mu\text{m}$
- (IIb) Follicle contains dividing cells; oocyte and its nucleus visible (arrow). Scale – 12.5  $\mu\text{m}$
- (III) Oocyte and yolk occupies up to 10% of follicle. Scale – 5  $\mu\text{m}$
- (v) Mature chorionated egg. Scale – 32  $\mu\text{m}$

GM – germarium; VT – Vitellarium; RO – right ovary;  
 SD – Spermathecal duct; LO – left ovary;  
 GC – germarial cyst; FC – follicular epithelium;  
 NS – nurse cell; Y – Yolk

### Precocene--induced sterility in F<sub>1</sub> generation of *Glossina morsitans morsitans*

Precocene treatment of female *G. m. morsitans* does not disrupt its reproductive cycle but some F<sub>1</sub> females produced are sterile and their ovaries contain only germaria (ICIPE Annual Report, 1980). Further studies on the effects of precocene on female tsetse suggest that the 'critical' time for precocene action appears to be related to each ovulation and precocene could have its effect on the recently fertilised egg which could ultimately give rise to an adult female. Furthermore, each application of precocene, made either at the time of ovulation of the first egg or subsequently after each larviposition holds good for one reproductive cycle only. The occurrence of retardations/abnormalities in ovarian development among F<sub>1</sub> females, in addition to the total absence of oocytes in vitellaria has enabled the characterisation of stages in development of ovarian follicles in *G. m. morsitans* (Fig. 2).

It was reported that JH III reduced the incidence of sterility in F<sub>1</sub> females (ICIPE Annual Report, 1980). However, in the later investigation, initial sterility of females was not established and it is possible that these females already had oocytes in their ovaries at the time JH III was applied. Similar experiments were repeated with juvenile hormone analogues after sterility of the females was established. Topical application of ZR 512 and ZR 515 did not promote follicle development even 37-39 days after adult emergence. However, these compounds appeared to influence the development of the milk gland in sterile females.

Although precocene did not induce degeneration of corpus allatum of treated 'mothers', the corpus allatum of her sterile offspring appeared relatively inactive and degenerate. Toluidene blue stained semi-thin sections and electron micrographs suggest that the gland had undergone some degree of degeneration and that

they were not active at the time of fixation.

### Studies on male *G. m. morsitans*

Antibodies were raised in rabbits against some components of the reproductive organs and fat body of male *G. m. morsitans* to study the effects of ingested antibodies on the development of abnormalities/lesions in tsetse fed on immunised rabbits. Larviposition was normal and there was no significant mortality among male or female parents, pupal development, spermatogenesis, accessory gland development and inseminating ability of F<sub>1</sub> generation from parents fed on immunised rabbits were similar to those of controls.

Pharmacological studies on the mechanisms involved in the release of accessory gland material during copulation are being continued.

### Immunomechanisms in tsetse

Groups of 50 female newly emerged *Glossina morsitans* were maintained for two successive generations (F<sub>0</sub> and F<sub>1</sub>) on rabbits immunized with homogenates of whole crude tsetse (WCT), engorged tsetse guts (ETG), gravid tsetse uteri (GTU) or on untreated control rabbits. It was confirmed both by immunodiffusion and by immunoelectrophoresis that these tsetse-derived antigens provoked a strong antibody response in the inoculated rabbits. No mortality occurred in the flies maintained on the immunized rabbits but a decrease in fecundity and in the mean pupal weights, and a slight increase in pupal mortality were observed. For the two fly generations (F<sub>0</sub> and F<sub>1</sub>), a total fecundity decrease of approximately 31%, 36% and 47% were observed in the CWT, ETG, groups respectively when compared with the controls. The overall decreases in the mean pupal weights for F<sub>0</sub> and F<sub>1</sub> generations were approximately 12%, 9% and 12% for CWT, ETG and GTU groups respectively. No pupal mortality occurred in the pupae of F<sub>0</sub> flies, but in the F<sub>1</sub>

generation alone, a pupal mortality of approximately 7, 18, and 8% were observed in the CWT, ETG, and GTU groups respectively.

These observations strongly suggest that this immunological technique, if improved, e.g. by using purified insect antigens may prove to be useful for tsetse control. This view is strengthened by the fact that different fly generations feeding on the same immunized animals are affected. This suggests that in an isolated tsetse-infested area, if the animals favoured by the flies as the source of blood are immunized, the fly population will presumably decrease progressively. Lastly, it should be remembered that these data have been obtained from a laboratory maintained in almost ideal conditions and that in the field where adverse conditions exist, the pathological effects on both flies and pupae will be more pronounced.

#### TRYPANOSOME-VECTOR PHYSIOLOGY

##### Temperature effect on the vectorial capacity of *G. m. morsitans*

A variety of factors have been suggested to account for the variable trypanosome infection rates observed in the tsetse following engorgement on *T. brucei* infected animal. In this exercise the intention was to see if there was a difference in the infection rates between flies fed directly on the bellies (the number of trypanosomes circulating in the veins per unit time may not be uniform or constant) of infected rats and those fed through silicone membrane on the same population of trypanosomes suspended uniformly in defibrinated rat blood.

It was interesting to note that among the control group of membrane fed flies, the number of infected flies remained remarkably constant (12%) regardless of the height of parasitaemias. On the other hand, flies cooled 6hr after engorgement showed a steady increase (from 6.5 to 21%)

in infection rates. No consistent results were obtained with flies cooled immediately after engorgement, although the highest infection rates (31.5%) were observed among this group. Flies fed on the bellies of infected rats gave variable results.

##### Estimation of the least number of *T. brucei* infective to *G. m. morsitans*

The observation that *in vitro* feeding gave more consistent results than the *in vivo* method, made it important to see if it was possible to determine the least number of trypanosomes that would initiate an infection in a tsetse. The number of flies which showed salivary gland (mature) infection did not differ substantially whether the source was  $3.3 \times 10^{-1}$  or  $3.3 \times 10^5$  organisms/ml. Nevertheless, there was a significantly higher incidence of immature (gut) infections when the source was  $3.3 \times 10^5$  organisms/ml.

##### Relations between proteolytic enzyme activities and the survival of *T. brucei* by young *G. m. morsitans*

It has been demonstrated that ingested bloodstream form of *T. brucei* influenced the activity of proteolytic enzyme (trypsin) in young female *G. m. morsitans*. The decrease in enzymic activity is thought to be caused by the presence of trypanosomes. It is postulated that the enzymes are being utilized in the lysis of trypanosomes. In order to examine this possibility, young *G. m. morsitans* have been exposed to rats infected with *T. brucei* at various parasitaemic levels and enzyme activities of trypsin and aminopeptidase monitored over a 4hr period after trypanosome ingestion.

No significant differences were observed in aminopeptidase activities between control and flies which ingested trypanosomes; nor was there a difference between day 3 and 5 infections. Trypsin activities were constantly higher in control flies than in flies which fed on trypanosome

infected blood. The differences were however, not statistically significant.

Flies fed on rising parasitaemias (day 3 infections) destroyed almost all ingested trypanosomes within 24hr. On the other hand, flies fed at peak parasitaemias (day 5 infections) destroyed the ingested trypanosomes gradually. The surviving trypanosomes after 24hr of ingestion, were virtually all stumpy in shape. The sharp decrease in the number of slender trypanosomes in both cases indicated that these organisms were more susceptible to the lethal factor in the tsetse midgut.

#### Oxidative metabolism of metacyclic trypanosomes cultured *in vitro*

In the infected mammalian host *T. brucei* utilizes glucose in the plasma and in

about 2hr the parasites can consume glucose equal to their own dry weight. The end product of glucose metabolism is pyruvate. In this way bloodstream lack the cytochrome systems. Hence in terms of energy conservation they are poor doers, indeed, since only 2 ATP are formed from one molecule of glucose compared to 38 from eukaryotic cells.

The metacyclic trypanosomes we have grown *in vitro* for about 4 years now also utilize glucose to pyruvate but also yield glycerol (ratio of 3:1). The latter product is toxic to bloodstream forms of *T. brucei*. The studies indicate that the infective trypanosomes grown from the vector have a peculiar glucose utilisation pathway which would be of interest in the study of mechanism of energy production in *T. brucei* of tsetse origin and bloodstream forms (Fig. 3).

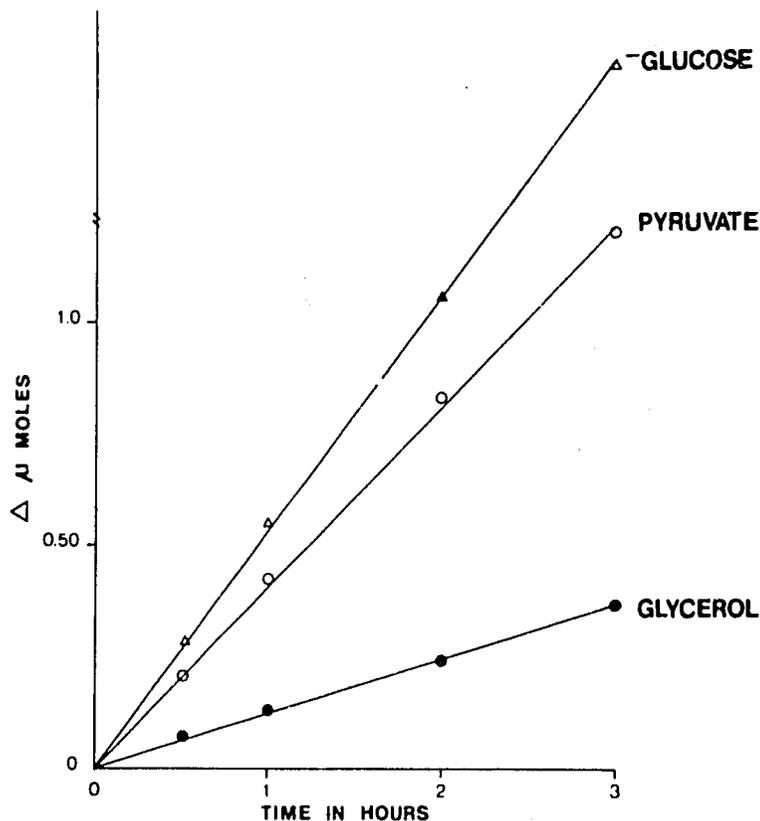


Fig. 3. Utilisation of glucose (GLUCOSE) and production of pyruvate and glycerol by metacyclic *T. brucei* propagated *in vitro*.

Possible existence of sexual forms of *T. brucei*

The possible existence of sexual forms of *T. brucei* and other pathogenic trypanosomes has been in the minds of many investigators for a long time. However, at no time has karyogamy (meiosis) been demonstrated. It has not been possible either to transfer the phenomenon of drug resistance of one strain of the parasite to another showing no resistance.

In our laboratory we have grown *T. brucei* as midgut and metacyclic forms and in both types of parasites a phenomenon was observed that suggested possible existence of sexual forms at light and electron microscopic observations. Two

parasites were seen to come into opposition at their posterior ends. In many cases one of the parasites was smaller or slender than its partner and displayed vigorous twitching movements at the region of the kinetoplast or flagellar pocket of the bigger parasite. Parasites were also seen to lie parallel to each other. Electron microscopic studies showed that the point of contact of the two parasites were either the kinetoplast, flagellar pocket or nucleus. Two of these sites (kinetoplast and nucleus) contain genetic material.

The classic definition of sex requires that a new individual arise by union of two sex cells or gametes. The formation of new individuals was not demonstrated in this study (Fig. 4).

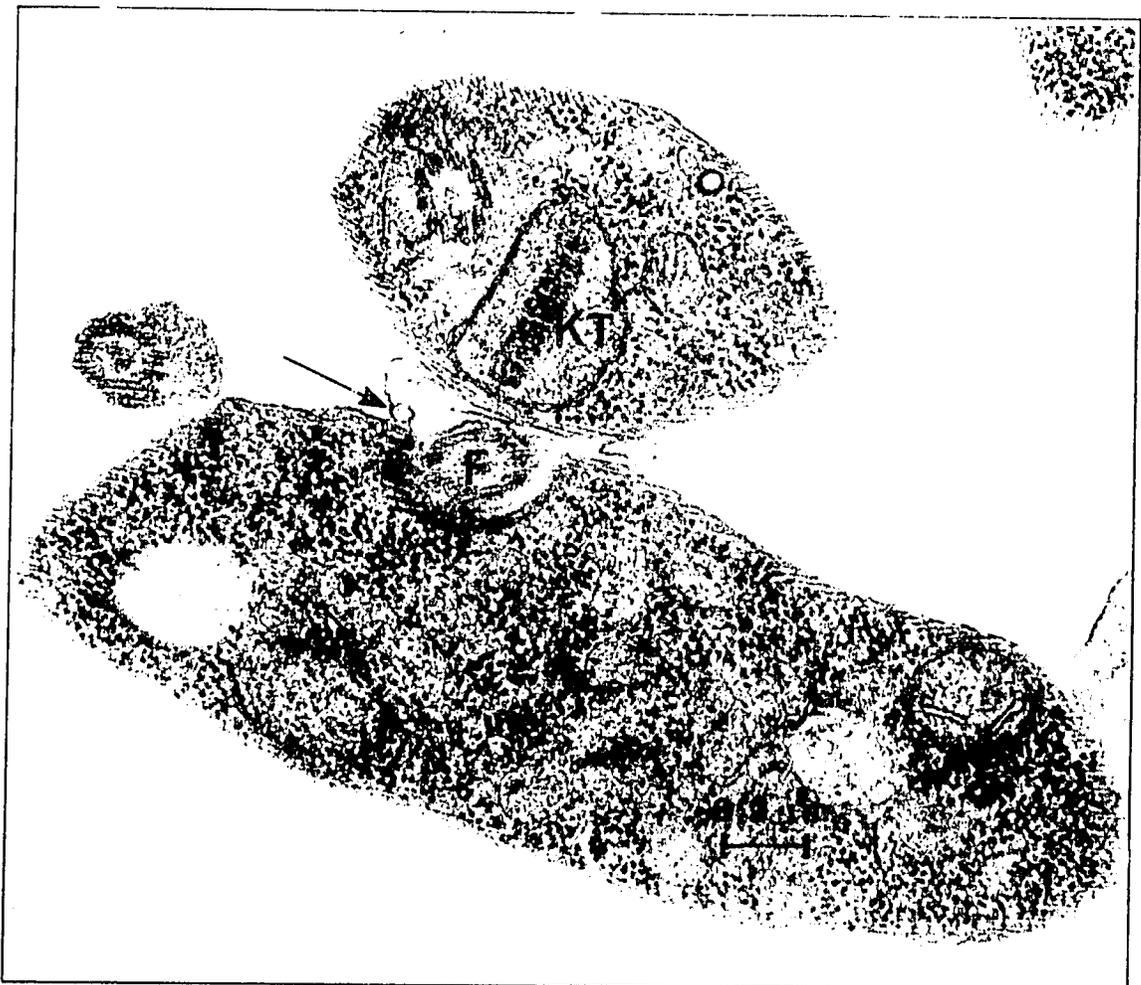


Fig. 4. Possible sexual forms of *T. brucei* in culture. Areas of contact were kinetoplast (KT) and flagellum (F). Arrow indicates reflected membrane.

The sensitivity of trypanosome-infected *Glossina morsitans* to toxic substances, endosulfan and pyrethrins

We previously reported (ICIPE Annual Report, 1980) that preliminary results suggested that trypanosome infected male *G. m. morsitans* Westwood were more sensitive to endosulfan than non-infected flies. We have extended our studies to include female flies and another toxic substance, natural pyrethrin. *G. m. morsitans* reared in our insectary were used.

Infected flies of both sexes showed statistically significant increase in sensitivity to both toxic substances. For endosulfan, there was 48% mortality in non-infected males (28 dead out of 58) compared to 90% mortality (18 dead out of 20) for the infected male flies. For the pregnant flies, non-infected flies showed 31% mortality (8 dead out of 26) as compared to 68% mortality (13 dead out of 19) in the infected group. For the pyrethrin extract, there was 14% mortality in the non-infected males (14 dead out of 101) compared to 50% mortality (13 dead out of 26) in the infected flies. In the pregnant group there was 40% mortality (70 dead out of 175) in the non-infected flies and 59% mortality (17 dead out of 29) in the infected group.

These results support our hypothesis that *G. m. morsitans* with mature salivary gland infections of *T. b. brucei* are not as healthy as non-infected flies. This contention will have to be tested further using other parameters of stress. Nevertheless, the data from this study indicate that *T. b. brucei* harms the fly. This view has not been widely held in the past. Several studies have provided data which suggest that trypanosome infections are harmless to the tsetse and may perhaps increase the fly's longevity.

We are continuing to explore changes in physiology and behaviour of infected flies in the hope that this information

will provide new insights for trypanosomiasis control.

## EPIDEMIOLOGICAL STUDIES

Epidemiology of human sleeping sickness in Lambwe Valley, Western Kenya

Lambwe Valley is one of the few areas in Kenya where human sleeping sickness is known to occur from time to time, occasionally reaching epidemic proportions. In June 1980, such an epidemic occurred. This outbreak was described by the local inhabitants as being much more severe than those of the recent past and even children were affected. The involvement of children suggested that apart from the disease being contracted in the game reserve area, active transmission was occurring close to or in the villages.

Three surveys have been carried out in the Riamakanga area of Wiga, to examine the role of *G. pallidipes* in the transmission of this disease. One survey was carried out during the outbreak of the disease and the next carried out at the beginning of endosulfan spraying operations. The last survey was carried out 6 months after endosulfan spraying. The isolated *T. brucei* sub group organisms were characterised using isoenzymes and the Blood Incubation Infectivity Tests (BIIT).

During the first survey, four stocks identical to stocks isolated from man in 1980 outbreak were isolated. And during the second survey, one *T. brucei* isolate gave similar enzyme patterns to *T. rhodesiense* isolated from a patient in this area. It was interesting to note that a stock with similar enzyme pattern combination has been found in patients in Zimbabwe. All these stocks are probably man infective.

## PATTERNS OF TRYPANOSOME INFECTION IN *GLOSSINA PALLIDIPES* ON THE KENYA COAST

Populations of *G. pallidipes* have been monitored for trypanosome infection rates,

factors affecting infection rate and trypanosome species, at five selected localities. These areas were chosen on basis of habitat with wild hosts. Diani and Ukunda are semi-rural areas with mainly domestic stock and few wild animals. Muhaka is an intermediate situation having both wild and domestic animals. Using biconical traps, *G. pallidipes* has been sampled quarterly, that is during the long rains (May), intermediate rains (August), short rains (November) and the dry season (February). The parasites have been identified through their location in the fly and the age of the female tsetse has been determined using ovarian age-grading techniques.

The trypanosome species observed in order of their abundance are: *Trypanosoma congolense* type, *T. vivax* and *T. brucei*. *Trypanosoma brucei* is rare, but has been identified in all the study areas except Muhaka. The Blood Incubation Infectivity Test (BIIT) has been carried on some isolates of *T. brucei*. No human strain(s) has been identified on the coast. *Trypanosoma simiae* has however, been identified from Muhaka. Since *T. congolense* and *T. congolense* and *T. simiae* inhabit the same location in tsetse, all infections of gut and proboscis have been included together as *T. congolense*. *Trypanosoma congolense* appears to be stable in the study localities with *T. vivax* accounting for most of the fluctuations.

Although *G. pallidipes* was feeding more on bovids than suids in Shimba Hills, Diani and Ukunda, a lower infection rate was noted at the former. This could be related to the presence of only wild hosts which have lived with the trypanosomes

for a long time and may have evolved some immunity to infection. *Glossina pallidipes* in Mwalewa and Muhaka appear to be feeding more in suids.

Highest infection rates were generally observed during the dry season and lowest during the main rains. Trypanosome infection rates increased with the age of the tsetse up to around 70 days beyond which they tended to decline. This suggests that older flies may lose their infection or disappear from the population due to higher mortality.

A detailed study is being made of the situation at Diani. A semi-isolated population of *G. pallidipes* is associated with a small area of forest. Tsetse are feeding both on a small herd of cattle grazed around the forest and on wild pigs using the forest as a refuge. Epidemiological and ecological techniques have been used together to estimate the intervals between infective bites received by each cow per day from *G. pallidipes*. The infection rate in animals has also been determined. Although the cattle are under regular chemoprophylaxis, an infection rate of more than 30% was observed in these animals. Infection rate in wild pigs has been reported to be lower.

Tsetse take more than 50% of their meals from bovids, mainly cattle as the other bovids in these areas come out only at night. Since *G. pallidipes* feed every 3 to 4 days, the observed infection rate of 5% in *G. pallidipes* suggests a considerable barrier to trypanosome infection from the mammalian host to tsetse. Studies are continuing at this locality in an attempt to develop an epidemiological model for this situation.

# MEDICAL VECTORS RESEARCH PROGRAMME

## INTRODUCTION

Since the Medical Vectors Programme was established, its aim has been to develop alternative control methods without using insecticides. As far as mosquitoes are concerned these methods are mainly aimed against preimaginal stages. In this respect, most of the work done in 1981 has dealt with taxonomic problems and various aspects of preimaginal ecology of malaria and filariasis vectors: oviposition behaviour, site-selection, egg ecology and other factors regulating preimaginal populations. Amongst these, a lot of attention has been paid to the role of mosquito natural enemies (especially predators and competitors) and to the effects of salinity, since brackish waters are an important part of coastal ecosystems.

Identification of the species of the *Anopheles gambiae* complex by using electrophoresis had been so far restricted to the adult stages. It has been demonstrated that preimaginal stages can also be identified by electrophoresis techniques.

From laboratory studies on the influence of salinity, it appears that an increase in the salinity at breeding sites of *Culex quinquefasciatus* could lead to partial control of this mosquito. This would be especially feasible along the Kenya coast where salt introduction into pit latrines is commonly practised in the belief that it lowers the water level of the latrines. Salinification could not, however, be applied against freshwater *A. gambiae* and *Anopheles funestus* since there is a risk of replacing these two mosquitoes by *Anopheles merus*. Field observations on mosquitoes breeding in brackish water sites of the Kenya South Coast have shown that five species can develop at various ranges of salinity, the species having the widest range of tolerance being *Culex sitiens*. This may confirm the hypothesis that the latter consists of several species. As far as the freshwater

species of the *A. gambiae* complex are concerned, it has been observed that in the laboratory, females prefer to oviposit on dark targets but this preference is overridden by pale turbid water from natural breeding sites. This is interpreted as meaning that probably there is an arrestant factor of chemical nature, at play. The prolonged duration of egg viability shows that egg ecology is a determinant factor of anopheline preimaginal population dynamics and that this factor should no longer be ignored.

In the study of competition between *C. quinquefasciatus* and *C. cinereus* it has been demonstrated that the latter is more susceptible to domestic detergents. This could explain, at least partially, its absence from breeding sites which contain those detergents.

Work on visceral leishmaniasis (=kala-azar) has continued on the different aspects of the epidemiology of this disease. In the field of vector studies, an attempt was made to find breeding sites and to identify eggs and preimaginal stages of species collected in various areas, but most of the work was carried out on adult stages, especially on resting sites. Investigations on parasites have dealt with *Leishmania* isolated from different sandfly species and also on the possible effect of these parasites on the vectors. As far as reservoirs are concerned an assay has been made to isolate antibodies from dogs which have been exposed to *Leishmania* infection.

Studies on visceral leishmaniasis epidemiology have led to two major achievements. The first one is on the finding of *Leishmania* parasites in the malpighian tubules of two sandfly species. This is the first time that this type of information has been encountered in Kenya Phlebotominae. The second achievement is on the uptake of promastigotes of a lizard

*Leishmania* by mouse macrophages. This may suggest a possible relationship between lizard and mammalian *Leishmania*.

The insect pathology group has been dealing with two possible ways of controlling mosquitoes and tsetse. Studies on the first one have concentrated on two categories of larval mosquito pathogens (a fungus species and three genera of microsporidia) and the second one on a virus-like particle infecting salivary glands of tsetse. On the other hand an attempt has been made to develop immunological methods for immunizing rabbits against tsetse.

Observations on mosquito pathogens have shown that the fungus *Coelomomyces indicus* is able to infect freshwater species of the *A. gambiae* complex all the year round. In pools where it occurs, the infection rate is almost always over 50% with the highest incidence of infection occurring during the short rainy season. In addition these mosquitoes, together with *A. merus* and *C. sitiens* have been found infected by microsporidia. Infection rates throughout the year were low and tended to be limited by rainfall and high salinity for brackish water breeders.

Research on tsetse pathology has shown that the incidence of salivary gland hypertrophy in field populations of *Glossina pallidipes* showed high site variation throughout the year while seasonal variation was only slight. The causative virus was found to be infective to *G. pallidipes* by intrahaemocoelic and *per os* infection, but not to *G. morsitans*. On the other hand tsetse fed in the laboratory on immunized rabbits were adversely affected in different ways. If the same is applicable to cattle under field conditions, it could lead to a reduction of fly populations and could then be considered as an alternative way of controlling *G. pallidipes*.

## MOSQUITOES

Identification of species of the *Anopheles*

*gambiae* complex by use of electrophoresis

Electrophoresis of soluble enzyme protein structural products on starch gel, has become one of the most important tools for identifying morphologically similar species. The advantages of this method are that the genotypes of an individual can be scored at a number of loci simultaneously and relative electrophoretic mobilities (electromorphs) usually show simple mendelian inheritance without dominance. More than one member of the *A. gambiae* complex are known to occur in the coastal region, therefore, it became necessary to be able to distinguish the freshwater and saltwater breeding members of the *A. gambiae* complex.

The method adopted was that of horizontal starch gel electrophoresis by Smithies (1955) employing the buffer system of Barlow and Ridgeway (1971). The electrode buffer pH 8.9 contained boric acid, hydroxymethyl aminomethane and disodium ethylene diaminetetra acetic acid. The gel contained 11–12% of starch per 100 ml of buffer prepared by reducing the pH of the electrode buffer to 8.2 with Boric acid and diluting to three tenths of original concentration. The mosquitoes to be identified are homogenized in 5 ul distilled water, adsorbed onto 4 x 4mm of chromatography paper and inserted into the gel. Electrical connections are effected and electrophoresis conducted in a refrigerator. After electrophoresis, the gel is sliced into two equal parts, one slice may be used to detect esterase enzyme activity and the other Octanol dehydrogenase (ODH) and superoxide dimutase (SOD) activity. These enzyme activities are made visible by incubating in stain until the bands become visible.

To distinguish freshwater from saltwater breeders the SOD bands are sufficient: *Anopheles merus* (a Kenya strain from Jimbo) has slower SOD white bands than freshwater *A. gambiae s. l.*

The ODH dark bands in E. Africa would easily identify *A. arabiensis* ODH 100/95, 100/90, 95/95, 95/90 and *A. gambiae* has a faster ODH than *A. arabiensis*. However, it must be noted that the probability of error for ODH genotype 100/100 is 0.002 for *A. gambiae s. s.* in E. Africa. This system of identification has advantages over the chromosome method because preimaginal stages, and all adults, whether male or female and at whatever stage of the genotrophic cycle or growth, may be identifiable. This method is to be used in future to find out the distribution of *A. merus* at the Kenya coast.

#### Role of salinity

Laboratory observations onto the freshwater species of the *A. gambiae* complex, *Anopheles funestus* and *Culex quinquefasciatus*. Salinity as a limiting factor in the breeding of these species has not been well documented but is known to have detrimental effects at high concentrations.

Newly hatched larvae of these species were reared up to the adult stage in serial dilutions of 0, 10, 30 and 40% sea water in distilled water. Their medium lethal concentrations ( $LC_{50}$ ) of sea water was also estimated, but for newly hatched larvae only, in the same manner as for larvicides.

No significant difference was observed in the larval survival rate of *C. quinquefasciatus* when sea water concentration was ranging from 0 to 20%. The differences observed for *A. funestus* were significant between 0 and 5% but not significant either between 5% and 10%, or between 0 and 10%. It can therefore be assumed that the survival of this mosquito was stable in the trend 0 to 10%. Survival rate of freshwater *A. gambiae s. l.* was significantly different between 0 and 5%, and not significant between 5 and 10%. Maximum salinities supporting full development for the larvae were 30–40% sea water for *C. quinquefasciatus* and 20–30% sea water for both *A. funestus* and *A. gambiae s. l.* The period

to full development of *A. gambiae s. l.* and *A. funestus* was the same in all the different solutions of sea water used, while the average period for *C. quinquefasciatus* was 9 days in all dilutions within the range 0–20% sea water and 12 days at 30% sea water.

$LC_{50}$  of sea water for *C. quinquefasciatus*, *A. funestus* and *A. gambiae s. l.* was 60.3%, 20.3% and 50.5% respectively.

Field observations in brackish water habitats in the south Kenya coast. An investigation was started to find out whether there is a mosquito faunal succession and potential dynamics related to salinity variation in a brackish breeding site in Jimbo, a coastal village at the Kenya-Tanzania border. Sampling at 2-week intervals began in October 1980 and ended in October 1981. The immature stages sampled were reared to adulthood for confirmation of identification. The range of salinity recorded in the pond was between 13–134% sea water although mosquito larvae were only recorded between 13 and 54% sea water.

Five different mosquito species were collected during the course of the present work: *Aedes albocephalus*, *A. gambiae s. l.*, *Culex sitiens*, *Culex thelassius* and *Culex tritaeniorhynchus*. *Culex sitiens* occurred abundantly at salinity range of 31–40% sea water and was the most abundant species in the pond. Least abundant was *Ae. albocephalus*, recorded only between salinity 21–30% sea water.

It seems apparent from the data that of these five species or species complexes, *C. sitiens* tolerates the widest range of salinity, an explanation for which may be either the efficiency of its anal papillae to regulate the salt content of the larval body, or that *C. sitiens* may be a complex of species capable of tolerating different ranges of salinity. This observation on *C. sitiens* requires further investigation to establish whether *C. sitiens* does indeed

consist of a species complex which is the opinion of other workers.

*Ae. albocephalus* larvae appear either to be incapable of surviving in salinities above 25.5%, or to occur only in freshly-flooded conditions. The species has nevertheless been reported from West Africa to occur in crabholes. Its possible inability to tolerate high salinity on the Kenya south coast seems in keeping with personal observations that the *Ae. albocephalus* collected in the Jimbo pond has certain slight morphological variation from the West African holotype specimen, and might therefore merit new taxonomic status.

*A. gambiae s. l.* seems to be the most tolerant species to high salinity. Due to that tolerance exhibited it is possible that the *A. gambiae* present in this pond is mainly the saltwater breeding species *Anopheles merus* of the *A. gambiae* complex.

The salt concentration in the pond seems to be a density regulator of the species present in it at any given time.

### 1.3. Preimaginal ecology of *A. gambiae* freshwater breeders

#### 1.3.1. Oviposition studies

Oviposition is one of the first priorities to be considered when one wants to study preimaginal ecology as whole. Aquatic habitat selection depends in the first place on the ovipositing female and from this springs the entire ecological life strategy of the species in its preimaginal stages. Experiments in large cages, involving overnight exposure of petri dishes differing in treatment in one respect only, in a 2 x 2 layout with one or both of the two freshwater-breeding malaria vector species ('fw') of the *A. gambiae* complex, ready to oviposit, have confirmed that:

(a) Neither the larvae nor the pupae of their own species attract or repel these ovipositing females.

(b) Although dishes of clear water backed with black receive more eggs than dishes with paler backing, they receive less eggs than dishes with turbid (and therefore paler) water from natural breeding sites. This difference is greater if the black-backed dishes contain distilled water than if they contain dechlorinated tap water, and these replicates finalise the conclusion that natural breeding site water appears to contain an oviposition arrestant of a chemical nature, not due to the prior presence of the preimaginal stages.

Studies on the circadian pattern of oviposition in 'fw' *A. gambiae* have been finally concluded with experiments on field-collected females re-fed at 20–2100 hr and 05–0600 hr sun time and offered oviposition targets 2 nights later. The first of these lots yielded a peak of oviposition in the first hr of the night and the second at 00–0200 hr. This confirms clearly that the timing of oviposition in both these species is not regulated by an endogenous activity rhythm as previously believed, but depends only on the temperature-dependent duration of ovarian development following blood-feeding.

In a sense, oviposition studies have progressed by a series of backward steps as it has been realised that results have been uninterpretable until first principles are reinvestigated. After discovering that 'fw' *A. gambiae* oviposit from a distinctive form of flight when optimal stimuli are presented, an observational scoring method was developed to quantify these responses. This involves holding females without access to oviposition targets until the end of the night on which they are due to oviposit and then, from 0430 or 0500 hr (sun time), their oviposition activity can be synchronised and is usually completed within 25–35 min. The number of females responding in each of the three different ways are then recorded every 2 min. As a standardised laboratory procedure this method has been used to reassess responses to HS oils, and could be used

for monolayers and other new larvicides for which such data are totally lacking. We have used it so far, with various modifications, for:

- (a) comparing specific oviposition characteristics of 'fw' *A. gambiae* with the 'seawater' *A. merus*, and of 'fw' *A. gambiae* with *A. funestus*.
- (b) assessing responses of *A. merus* to water and mud with and without salinity.

**Egg ecology.** The viability of 'fw' *A. gambiae* eggs has been assessed when transferred to hypertonic saline at various times after oviposition. A marked rise in viability, and thus in resistance to desiccation, occurs 8–10 hr after oviposition at ambient temperatures. This would mark the time when the developing embryo secretes its serosal membrane within the endochorion. Such changes in resistivity have important ecological as well as experimental implications.

The intrinsic viability of 'fw', *A. gambiae* and *A. merus* eggs has been tested on a variety of substrates. A moist substrate with no free water whatever yields the maximum delayed viability since it inhibits hatching most efficiently. Eggs remain viable for up to 16 and exceptionally to 17 or 18 days under these conditions.

The phenomenon of staggered hatching has been investigated, as well as hatching stimuli. At 25–30°C, egg incubation is completed in 33–36 hr but without stimuli such as sudden flooding, agitation or bright light, many eggs fail to hatch when due. Given stimuli it then takes just over 30 min for 50% of the eggs of 'fw' *A. gambiae* to hatch, and between 90 and 120 min for the same in *A. merus*, regardless of previous delays of up to 14 days.

The fate of eggs on various natural shortline substrate materials and the behaviour and viability of freshly-hatched

first instar larvae have also been investigated.

Development duration of preimaginal stages. Insectary data on development rates of *A. gambiae* are notoriously inaccurate. To circumvent this, and also to avoid the paucity of pupae from field data which make it impossible to assess durations of the fourth instar, rearing was conducted in large tubs containing highly turbid water from a natural site, held in the sun. Synchronised hatches allowed different lots to be started at contrasting times of day. The most striking feature here is that when first instars were commenced at 1000 hr, only 6 hr later than at 0400 hr, the "gate" of the endogenous pupation rhythm 4 or 5 days later (open only from 1430 hr to just after 1900 hr sun time) prolonged the mean duration of the fourth instar by some 40%.

The gross pupal deficit, derived from the proportions of each developmental stage taken in field samples, may be expressed by the formula  $n_4^2/n_3n_p$ , where  $n_3$ ,  $n_4$  and  $n_p$  are the sampled numbers of third and fourth instar larvae and pupae. When developmental durations of each stage ( $d_3$ ,  $d_4$ ,  $d_p$ ) are taken into account, the pupal deficit may be expressed more realistically by  $(n_4 d_4) / (n_3 d_3 n_p d_p)$ . The change in fourth instar duration ( $d_4$ ) brought about by the time of larval hatching and the pupation rhythm could then affect the pupal deficit by a further maximal factor of more than 3. Such effects have been considered in previous analyses of *A. gambiae* aquatic populations. Even so, they are far from sufficient to explain pupal deficits altogether as encountered in our population monitoring.

**Predators.** The greatly increased mortality in the late fourth instar/early pupal stages had been difficult to account for, except as a result of predation by adult notonectid populations. These adult water bugs occur at high densities only in deeper water, whereas *A. gambiae* preimaginal stages are

mainly confined to the shallows. Study of the behaviour of the developing larvae showed, however, that as pupation approaches, the pre-pupal fourth instar becomes progressively less responsive to disturbance or to a moving background and thus progressively more likely to drift to the hazardous deeper water. Field evaluation of this is required, but supporting evidence for it was obtained when high pupal yields occurred in deeper water in association with certain algal blooms, which afforded the larvae protection from notonectid predation. Even so, large pupal deficits in habitats where notonectids and indeed all predators are scarce or absent, remain extremely perplexing.

#### Competition between *C. quinquefasciatus* and *Culex cinereus*

*Culex cinereus*, a non man-biting mosquito, has been found able to replace *C. quinquefasciatus*, a vector of Bancroftian filariasis in some breeding sites of the Kenya coast. But *C. cinereus* is not equally distributed in this area. In 1980 the hypothesis had been put forward that the absence of this mosquito in some breeding sites could be explained by the adverse effect of domestic detergents contained in the used water of these breeding sites. An experiment had been set up using two latrines built in a small coastal village. Emerging males were monitored when leaving the breeding sites as explained in the 1980 annual report. *Culex quinquefasciatus* was the first species to colonize the breeding site, and *C. cinereus* was observed a few weeks later. After the two species had coexisted together, *C. quinquefasciatus* disappeared and by early October 1980, *C. cinereus* remained the only species present. In November 1980 detergent was introduced daily in one of the breeding places while the other one was kept as a control. After about 3 weeks *C. cinereus* had disappeared from the treated breeding site while it remained abundant in the control breeding introduced, only two males of *C. quinquefasciatus*

and 16 *C. cinereus* were collected when departing from the breeding site but there is no proof that they had actually developed in the site. Detergent introduction was stopped on 20 January 1981. *Culex cinereus* numbers then increased steadily until the end of our observations in early March.

Thus, it appears that detergents have a detrimental effect on *C. cinereus* and they can prevent this mosquito developing in breeding sites. It also appears that *C. quinquefasciatus* was unable to recolonize the breeding site after *C. cinereus* had been eliminated. In order to explain this, we conducted two series of observations, one on *C. quinquefasciatus* females departing from the breeding site during the period of detergent introduction, the other on the susceptibility of *C. quinquefasciatus* preimaginal stages to domestic detergents. In the study of departing mosquitoes we counted (a) the unfed parous females, (i.e. those which might have laid their eggs in the breeding-site), and (b) the gravid females (i.e. those which were leaving the site without having laid their eggs). Out of a total of 24 females there were nine unfed parous and 15 gravid. It therefore appeared that the site was not highly attractive to this species, since the overall numbers of females were low, and the majority of them departed without laying eggs. The susceptibility of *C. quinquefasciatus* and *C. cinereus* preimaginal stages to domestic detergents was compared by rearing batches of 20 first instar larva in water taken from the lower breeding site at different times through December 1980 and January 1981, i.e. after *C. cinereus* had been eliminated from the site. The effect of detergents was assessed by the pupal productivity i.e. the percentage of those first instar larvae which were able to become viable pupae. In the controls, pupal productivity was high for both *C. quinquefasciatus* and *C. cinereus*, 94.4% and 81.3%, respectively. In water containing detergent, none of the *C. cinereus* larvae could be reared as far



(2) Taxonomy. The studies on vector taxonomy have been conducted on *Sergentomyia* and *Phlebotomus* larval stages and egg sculpture. These investigations are still in progress. There appears to be differences in egg sculpture of different species of sandflies among the ones we have investigated but more numbers have to be screened to be sure the results are reproducible. This will be a valuable technique to tell apart the *Symphlebotomus* complex once they are colonized in the laboratory. The chetotaxy of the immature stages is in progress and so far larvae of five species have been reared in the laboratory and their differences are under investigation.

Investigations on directional flight of sandflies near homes: During the dry season, the animal enclosures for cows, goats or sheep which are located very close to human dwellings and are constantly wet from animal excreta and urine could serve as breeding sites. Traps were set up between the houses and coated with castor oil on both sides. During the dry season the fly population was monitored for houses and termite hills. The results indicated that more flies were captured on the side facing the animal enclosure.

Although only experimental data for one month is available in each case of dry wet season fly population, there is likelihood that animal enclosures may serve as possible breeding sites during the dry season only as indicated by the large number of flies caught in that direction.

Adult resting sites. Although it has been previously said that sandflies mainly rest in termite hills and has been demonstrated at the ICIPE that homes are also favoured resting places, during the dry and rainy seasons, the comparative studies on the distribution in various habitats had not been carried out previously. Open traps and square traps (sticky sandfly trap) were set at various habitats, i.e. open areas without bushes, thickets, around termite hills,

hanging on house walls etc.

An analysis of species prevalent during the dry season, beginning of rainy season and at the peak of rainy season (April-May) revealed the following species:

<i>Sergentomyia bedfordi</i>	<i>S. schwetzi</i>
<i>S. garulami</i>	<i>S. ingrami</i>
<i>Phlebotomus martini</i>	<i>S. antennatus</i>
<i>S. adami</i>	<i>S. affinis</i>
<i>S. rosannae</i>	<i>P. rodhaini</i>
<i>S. squamipleuris</i>	
<i>S. Kirki</i>	

The majority of the anthropophilic sandflies, *P. martini* and *S. garulami* were mainly encountered in houses and termite hills. Over 95% of these species were located in these resting sites. In habitats other than the houses and termite hills, there were comparatively fewer numbers of sandflies even during the rainy season when fly distribution in the environment is increased greatly. In these studies it is also noted that the domestic animal enclosures could be a source of fly population and we are investigating the possibility that these may be an alternate breeding site for sandflies. Comparison of open and square trap catches in the field, except termite hills and near houses, revealed that the open trap is more efficient.

Laboratory colony: Anthropophilic visceral leishmaniasis vectors are only in abundance during the rainy season. For research to continue laboratory colony was deemed a necessity. In collaboration with the Walter Reed Army Research Institute we have managed to start laboratory colony to F2 generation of *S. bedfordi*, *S. antennatus*, *S. africanus*, and *P. martini*. This has been a major achievement as it will be possible in future to have laboratory reared flies for experimental work for vectorial capacity and biochemical studies and for taxonomy of the sandfly species. In the F2 generation we have 200 *P. martini*, 300 *S. antennatus*, 230 *S. africanus* and a few *S. bedfordi* adult flies.

This achievement has been through very difficult experimentation on larval diet and feeding procedures which we feel may account for the success so far made.

We have also carried out experiments of watering the termite mounds during the dry season when sandfly population is very low. The results of this work carried out for 3 months revealed that watering did increase the number of flies in the resting sites. It was not clear whether the population increase was due to emergence of the population resting inside the termite hills. This aspect is being followed to answer these questions.

#### PARASITE STUDIES

Parasite isolation: Dissections of various species of sandflies continued at Kalawa (Machakos focus). Additionally, because of the kala-azar outbreak at Masinga and Kitui (see Fig. 1) comparative studies were carried out in these areas to assess the vector potential. These flies were caught from houses and termite hills and dissected in saline.

In the Machakos focus, promastigotes were encountered in the three species: *S. garnhami*, *S. bedfordi* and *S. schwetzi*. *Sergentomyia bedfordi* and *S. schwetzi* are mainly reptilian feeders in rock crevices, and since *Leishmania* parasites have been isolated from lizards, it would appear circumstantial that the parasites are of reptilian origin and determination of the type is pending.

In the Kitui focus more positive species were encountered including *S. garnhami*, *S. clydei*, *S. ingrani*, *S. graingeri*, *S. schwetzi*, *S. kirki* and *S. bedfordi*. As some of the flies were obtained from termite hills and it was in termite hills where *Leishmania* positive mongooses and a genet cat were obtained, it is not clear whether these parasites are of the same origin. Blood meals have been collected from these areas to assess what the flies are feeding

on and will be sent for analysis. Again determination of the type of strains is being awaited.

Vector-parasite relationships of *Leishmania* in Kenyan Phlebotomine sandflies.

Investigations are being carried out on naturally infected sandflies not only to isolate parasites in search of vectors of human leishmaniasis but also to bring out information regarding the infection rate of leishmanial parasites in sandflies; and the effect (if any) of the parasites on the vector.

Sandflies were captured from their natural resting sites in termitaria at Tseikuru (Kitui District), Msinga (Machakos District) and Marigat (Baringo District). They were processed in the same way as for parasite isolation. The guts were examined for leishmanial parasites and some of them were processed for ultrastructural investigation. The heads of those flies which were parasitemic in the gut were preserved for histological examination and species identification. From the results obtained, leishmanial infections were found in four species of sandflies in three sublocations (Muuna, Nziitu and Ngiluni) of Tseikuru. These species of sandflies included *Sergentomyia garnhami*, *S. kirki*, *S. graingeri* and *S. schwetzi*. At Masinga, leishmanial infections were found in three species of sandflies namely, *S. garnhami*, *S. bedfordi* and *S. antennatus*; and at Marigat in *S. antennatus* only. Parasites were found in the abdominal midgut, the hindgut and in the malpighian tubules. Only two species of sandflies namely *S. garnhami* and *S. antennatus* had parasites in the malpighian tubules. The infection in the malpighian tubules of *S. garnhami* was such that there were parasites both free in the lumen and in close contact with the "basement membrane". Cultures of parasites isolated from the malpighian tubules became positive so that the isolate was stabilized and incorporated in the ICIPE *Leishmania* cryobank.

It is not yet known whether the infection in the malpighian tubules of *S. garnhami* and *S. antematus* is accidental or whether it forms an important part of the route of migration of particular physiological types of leishmanial parasites in sandflies. The identity of the parasites isolated from the malpighian tubules has yet to be investigated.

Uptake of promastigotes of a lizard *Leishmania* sp. and *Leishmania donovani* by mice peritoneal macrophages: Inbred Balb/c mice strain of 7-8 weeks old were injected intraperitoneally with a known quantity of in vitro promastigotes of human leishmania (*Leishmania donovani*) and a lizard *Leishmania* species. The peritoneal macrophages of the mice were harvested at different time points after infection and the percentage of infected macrophages was determined after adhering the macrophages onto coverslips and staining with Giemsa. Results showed that when promastigotes were injected intraperitoneally into mice, the rate of infection of macrophages for the lizard *Leishmania* and *L. donovani* (human strain) were not markedly different.

Lizard *Leishmania* sp. assumed rounded or oval shapes like amastigotes of *L. donovani* and other mammalian *Leishmania*. After 24 hr intact lizard *Leishmania* parasites could not be seen in the infected macrophages' granules and clusters which appeared to be disintegrated parasites were numerous. *L. donovani* amastigotes were, however, clearly visible at this time.

These results suggest that amastigotes of this lizard *Leishmania* could be encountered in warm blooded mammals in nature. This could also explain partly why transient infections have been reported after some human volunteers and other lower warm-blooded mammals were inoculated with promastigotes of reptilian origin. The transformation of this lizard *Leishmania* sp. into amastigote forms, in mice macrophages which normally support

growth of *L. donovani*, is suggestive of some relationship between lizard and true mammalian *Leishmania*. More studies are being conducted to the role of reptiles in *Leishmania*. More studies are being conducted to the role of reptiles in *Leishmania* epidemiology in Kenya.

**Leishmania cryobank:** Well controlled investigations regarding vectorial capability and vector-parasite relationships of various species of sandflies require not only a supply of laboratory-bred sandflies but also a reliable source of known physiological types of leishmania parasites. It was because of this that a system called the "Leishmania Cryobank" was organized and established in 1979-1980 to make it possible to cryopreserve parasites which are isolated during field work.

During the period January-15 October 1981 a total of 23 isolates were made. Stabilates of the isolates were made and incorporated in the leishmania cryobank. The isolates were made from various vertebrate and invertebrate hosts including wild animals (mongooses and lizards), human and recovery laboratory mice.

**Serological investigation on Leishmania isolates from Kenya. Leishmania,**

Serological investigation on Leishmania isolates from Kenya: *Leishmania*, like other protozoan parasites, consist of a complex series of antigens. Some are species-specific, others are genus-specific and some are shared with related genera. Cross-reacting titres in antigenic analyses are generally lower than homologous titres. Particularly where speciation is actively going on, antigenic differences among emerging strains are to be expected. Studies on antigenic relationships of *Leishmania* is therefore important for strain differentiation and for the development of immunodiagnostic reagents with high specificity.

Human and lizard *Leishmania* strains stabilized as ICIPE 126 and 140 respecti-

vely were used as antigen. Large numbers of promastigotes were grown in Dulbecco's Modified Eagle's Medium (DMEM) supplemented with 20% foetal calf serum (FCS). Excreted Factor (EF) of the two species was partially purified from the infected medium. Antisera were raised in rabbits using promastigotes grown in what is the full name (NNN) culture media.

Using counter immunoelectrophoretic and double diffusion techniques the antigenic differences and/or similarities between the isolates were studied. Preliminary results showed that antiserum to the human isolate does not recognize any antigens or EF of the lizard leishmania. Immunoelectrophoretic studies employing homologous antisera to human and lizard leishmania isolates indicated a number of antigens peculiar to each isolate (not shown).

An attempt to develop an immunofluorescence assay for canine antibodies to *Leishmania*: Isolation of *L. donovani* sensu stricto from dogs in Kenya has raised the question as to whether more dogs from *leishmania* endemic areas could have been exposed to or actively infected by the parasite. This study was therefore initiated to screen domestic dogs in endemic areas to establish the degree of involvement in the spread of the disease.

Over 20 dog sera from kala-azar endemic areas were tested for antibodies to *Leishmania* by indirect FA technique. Three sera, which served as positive controls were obtained from dogs with parasitologically proven leishmania infections. Negative sera were obtained from dogs in non-leishmania areas.

Dog leishmania promastigotes stabilized as ICIPE 156 used as antigen were grown in RPMI-1640 supplemented with 20% foetal calf serum. They were harvested at peak growth, washed, fixed in formalin and smeared on clean glass slides. Serum dilutions were applied in encircled areas

containing the organisms. After incubating for half an hour the slides were washed and fluorescein conjugated rabbit anti dog (IgG) was applied and incubation and washing repeated. A drop of buffered glycerol was applied on the circles and surmounted with a cover glass. Fluorescence was assessed using an orthoplan microscope.

Preliminary results showed extensive cross reactivity among the different antisera. Studies are being conducted to perfect the test as a diagnostic tool.

### INSECT PATHOLOGY

Infection levels of a preimaginal population of *Anopheles gambiae* s. l. by the fungus *Coelomomyces indicus*

Of the parasitic fungi known to attack mosquitoes, *Coelomomyces* sp. are among the more promising for use in the control of preimaginal populations by biological agents. A preliminary survey of preimaginal populations of *A. gambiae* s. l. was conducted during two rainy seasons (1980 short rains and 1981 long rains) in the Rabai area (20km NW of Mombasa). Samples consisted of larvae collected at weekly intervals from a temporary infection pool (Fig. 2). The larvae were inspected microscopically for *C. indicus*, in the laboratory not later than the day after collection.

The time of first appearance of *C. indicus* in larvae of *A. gambiae* s. l. occurred about one week after the onset of short rainy season (on August 20 1980) and ten days after the onset of the long rains (on 23 March 1981). A severe drought affected the level of water in the pond during October 1980 and May, 1981 such that it dried up completely and no larvae were collected.

Infection levels for the long rains lagged behind those of the short rain season in terms of percentages of diseased larvae taken. The highest level of infection recor-

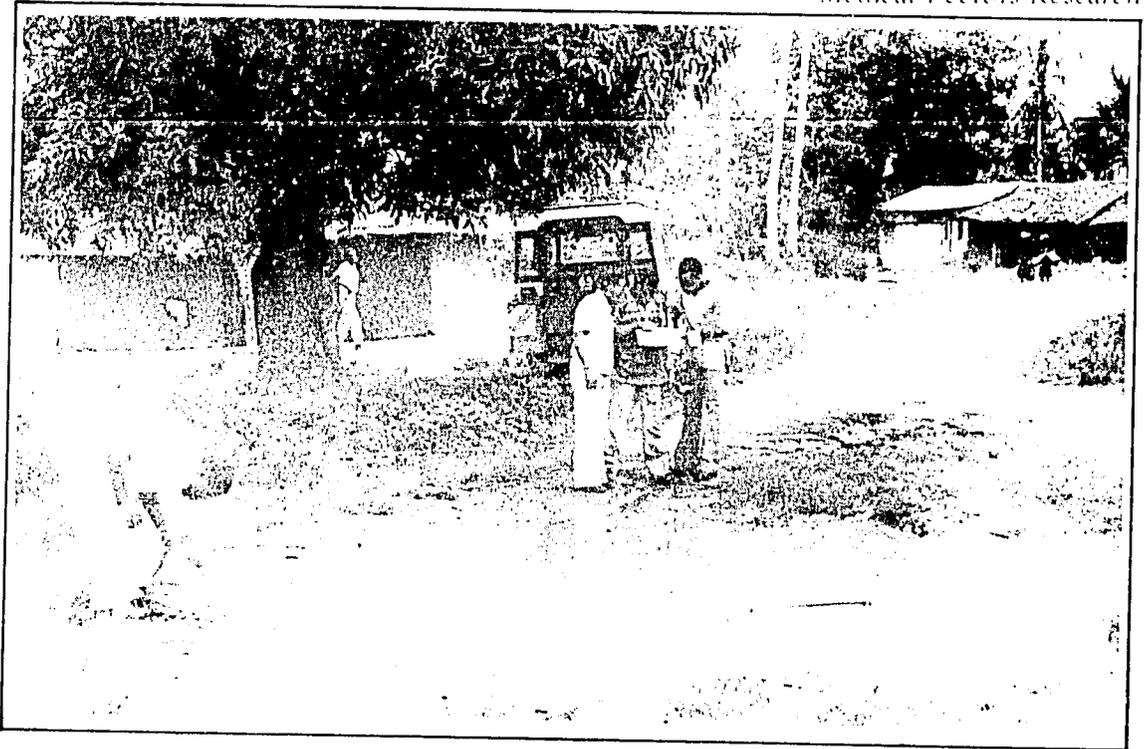


Fig. 2. A mosquito *Anopheles gambiae* s. l. breeding habitat: temporary pool, Mwamoni site, near Mombasa.

ded in any single monthly sample occurred in 1980 (November) when 82.41% of the larvae were stricken, but infection was generally high in that season. The high levels of infection noted in July occurred when numbers of larvae in the pond were very low; in that month the standard sample of 100 dip yielded 11 larvae out of which nine were infected.

Observations of levels of infection by *Coelomomyces* in field populations of mosquitoes have been reported by several workers, but these have covered periods of one growing season or less, or have been based on irregular collections. Longterm, systematically conducted field studies of *Coelomomyces* in natural populations of mosquitoes have not been available previously. As a part of a long term sampling regime, preliminary results covering two mosquito growing seasons is presented. Although the level of infection during the short rain is higher (71.81%) than the long rain season (53.53%) it is probably premature to say that the reduction in infection level was anything not within the realm of normalcy.

During the course of these investigations it has been noted that infected fourth instar larvae may live for several days, frequently more than 14, without pupating or dying. The extension of this phase of development of larva beyond the usual 2 to 4 days, causes an increase in the number of fourth instars over that normally expected to be in the population. The increase is a result of infected fourth instar larvae not leaving the larval population at the normal time, and the proportion of infected larvae in the fourth instar class is thereby increased.

The view is held by certain workers concerned with malaria control that retention of vector species in the larval stages can contribute to the control of vector population. The longer the mosquito remains a larva, the greater are the chances of its destruction by several forces, e.g. vertebrate and invertebrate predators, parasites, pathogens, wave action, drying of the habitat, etc. Moreover, the persistence of the fungus through period when the site dries out completely is a particularly important feature from the viewpoint of

controlling these mosquitoes.

Microsporidia infections of Anopheline and Culicine mosquitoes along the Kenya Coast

The potential of microsporidia as biological control agents have been demonstrated by various workers using different mosquito species. Microsporidian infections, which may be present at any developmental stage have been reported to have numerous effects on their host. They may

reduce the egg-laying capacity, longevity, and resistance of the mosquito to adverse climatic conditions, insecticides, predation and parasitism.

This report presents preliminary results of a study aimed at identifying major microsporidia of Anopheline and Culicine mosquitoes along the Kenya coast, and to determine the extent of distributions of microsporidia infections and the changes in rainfall, temperature, pH and salinity of the breeding sites.

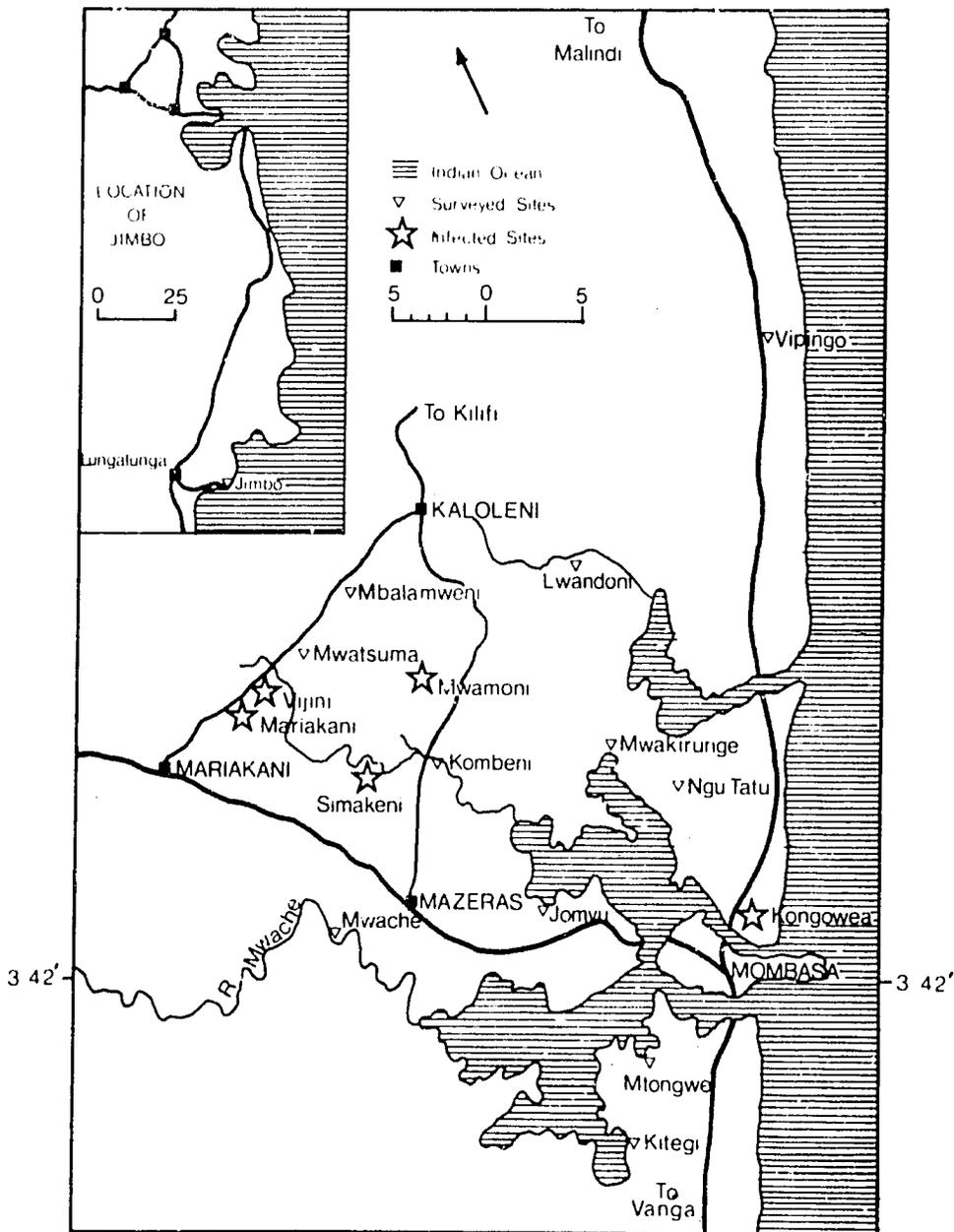


Fig. 3. A map of the Kenya Coast showing sites under survey for protozoan infections of mosquito larvae.

Initially, surveys of different types of breeding sites were carried out. These included salt, brackish and fresh water breeding sites. Larval samples were collected periodically from the sites and checked for infection. At the time of larval collection, the Ph, temperature, and salinity of the sites were recorded. Monthly rainfall figures were also taken. These surveys showed four sites to be infected with microsporidia : one freshwater pond and three brackish water pools. The fresh water site and one of the brackish water sites had infection only in September. The study therefore concentrated on the other two brackish water sites at Jimbo and Jomvu (Fig. 3).

Mosquito populations at Jimbo included *Anopheles merus*, *Culex sitiens*, and *Aedes albocephalus*. Microscopic examination of crushed samples of these mosquito larvae showed the presence of *Nosema* sp. (Fig. 4) and *Duboscqia* sp. in *A. merus* and *Nosema* sp. and *Thelochromia* sp. in *C. sitiens*. The unidentified *Culex* sp. and *Aedes albocephalus* were consistently free

of microsporidia wherever they occurred in the pools. In both pools, infections were low and occurred intermitently. In Jomvu, infection rates in *C. sitiens* were less than 15%. They were slightly higher in *A. merus*, going up to 20%. In Jimbo, infection rates were even lower in both species being below 5% at all times.

Culicine larvae infected with microsporidia were easy to detect in the field. The thorax and the abdominal segments were swollen and turned white. Infections began in the fat body of the middle abdominal segments and spread to the rest of the body tissues and organs. Heavily infected larvae did not successfully pupate but died at fourth instar. Infections in *A. merus* were not as easy to detect, except for the sluggishness in movement. In both mosquito species, heavy infections were confined mostly to the third and fourth instars. No first instar was ever found with mature spores.

Salinity ranges in the two study pools differed from each other. In Jomvu the

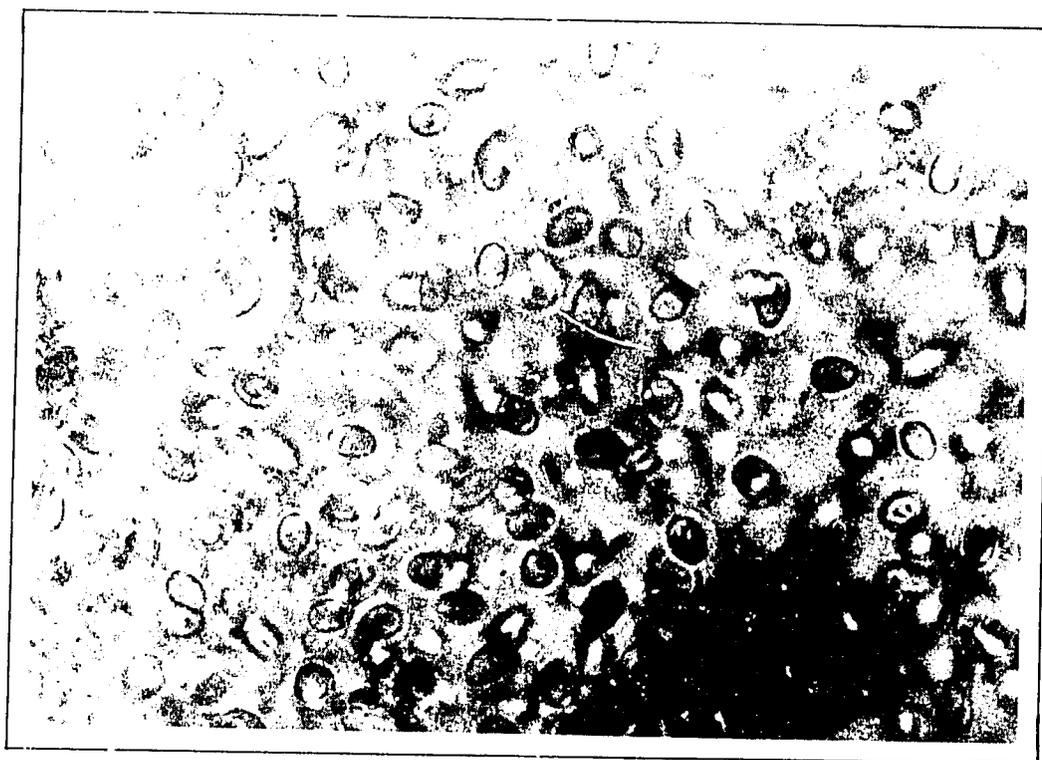


Fig. 4. A typical protozoan infection (Microsporidia; *Nosema* sp.) in its mosquito host *Anopheles* sp.

range was high (5–105% sea water). Infections occurred only during times of moderate salinity (18–45% sea water). Only on one occasion did infections occur briefly in 61% sea water, indicating that although high salinities deter microsporidian infections, infective spores are not completely destroyed. In contrast to the Jomvu pool, salinity in Jimbo was moderate at all time (10–47% sea water). The lapses between infections here were no more than one month in *C. sitiens* compared to the longer lapses in the Jomvu pool. Temperatures and pH of the pools remained fairly constant. pH was usually between 7.5–8, while temperatures were between 27°C and 32°C. On rare occasions temperatures went as low as 20°C and as high as 42°C. Water turbidity of both pools also remained constant, usually clear with a biota of beetles, crabs, snails, copepods and larviphagous fish.

It appears that the most prevalent groups of microsporidia along the coast are *Nosema*, *Thelohemia*, and *Duboscqia*, occurring mostly in semi-permanent sites and particularly in brackish water pools. The patterns of environmental factors at the study sites indicate that no one single factor is responsible for the occurrence, persistence, and distribution of these pathogens. Immediately after the start of the rains following the dry period, pools are filled with water and infections appear. A lot of continuous rain causes an overflow of the pools and infections disappear. Rainfall therefore seems to have an indirect effect on microsporidia. By causing an overflow of the breeding sites, the rain has the two effects: washing away the larvae and reducing their numbers in the pool and causing a dilution effect in volume of water. This reduces the chances of the few mosquito larvae being infected. It is also possible that the infections do not disappear completely but are reduced to such low rates that they fail to be monitored during sampling. Rainfall also serves to reduce the salinity of the pools, so that after moderate rainfall, salinity is maintai-

ned within suitable ranges and mosquito population rises, increasing the chances of infection. Infections were found to be higher during periods of high larval densities. As rain disappears, salinity increases, reducing mosquito populations and microsporidia infections. However, some microsporidia survive through the high salinity as indicated by isolated cases of infection found in 77% and 97% sea water.

Factors determining seasonal occurrence of microsporidia may affect deliberate use of the pathogen in mosquito control procedures. They therefore have to be determined in order to manipulate them in favour of highest mosquito mortalities. Although microsporidia have not been isolated from too many places along the coast, their presence indicates that with further studies, they could be manipulated in mosquito control operations against such important vectors as *A. merus* and *A. gambiae s. l.* from which they have been isolated.

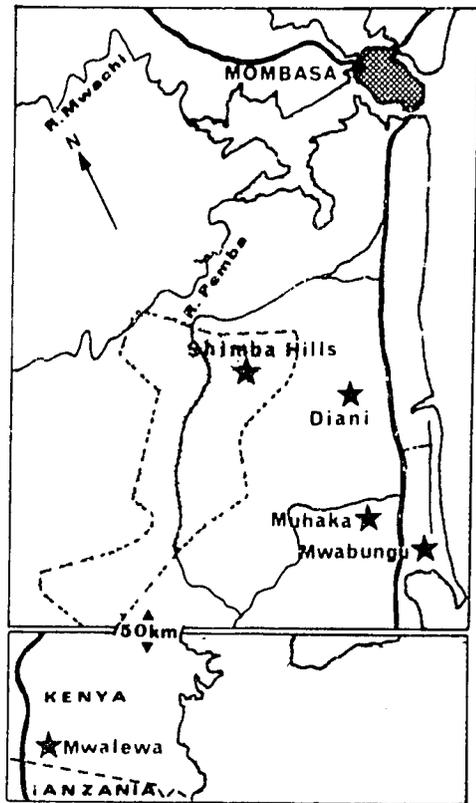
#### Incidence of salivary gland hypertrophy in field populations of the tsetse *Glossina pallidipes* on the south Kenya coast

The occurrence of hypertrophy in the salivary gland (HSG) in field populations of tsetse *Glossina* species was first reported from *G. pallidipes* trapped in Zululand. More recent work has traced the cause of cellular hypertrophy and hyperplasia resulting in the glandular enlargement to a large number of virus-like particles (VLPs) occurring in the nuclei and cytoplasm of infected salivary gland epithelial cells.

The present field investigations were planned to carry out a continuous study on wild *G. pallidipes*, *G. brevipalpis* and *G. austeni* from five sites of the Kenya coast in order to follow the course of VLP infection over 12 months. Virus-like particle infection has been associated with abnormality in both male and female reproductive systems, and could therefore contribute to the natural regulation of

tsetse numbers.

Tsetse were sampled in the field using the pale blue biconical traps. The five study areas were selected due to their diversity in vegetation, wildlife, and human activity. They were: Diani-Ukunda, Muhaka Forest, Mwabungu, Shimba Hills, National Hills, National Reserve and Mwalewa (Fig. 5).



**KEY**  
 ★ Study sites  
 Shimba Hills National Reserve  
 Indian Ocean  
 Mombasa Island

0 10  
 km

Fig. 5. Map showing study sites for incidence of salivary gland hypertrophy along the south Kenya Coast.

A sample of tsetse from each site were dissected each day and salivary glands examined for hypertrophy. The extent of hypertrophy was graded in nine categories, each category denoting the level of

enlargement of the salivary gland. Ages of female tsetse were determined by the ovarian method. Tsetse were also examined for any other pathological condition.

Infected species and symptoms of infection. Hypertrophy of salivary glands was recorded only in *G. pallidipes*. *Glossina brevipalpis* and *G. austeni* from the five trapping sites were also examined, but HSG was not recorded in these two species. Tsetse with hypertrophied salivary glands could not be distinguished from normal tsetse by external examination. In the teneral flies with infected glands however, the white enlarged salivary gland could be seen as a pale outline through the integument and it also formed irregular ridges on the soft and pliable integument. These two symptoms could not be relied on in older flies due to higher pigmentation, toughening of the cuticle, and enlargement of the abdomen by the developing egg and/or larva in the uterus of females. The activity of the flies bearing hypertrophied salivary glands were not impaired.

Hypertrophied salivary glands. The study was limited to observations on the gross pathology of the hypertrophied salivary glands. Hypertrophy was observed to start uniformly throughout the length of the distal part of salivary glands within the abdomen. Both salivary glands showed hypertrophy in all observed cases, and no instances of unilateral hypertrophy were seen (Fig. 6). Hypertrophied glands turned to intense blue when they came into contact with water. At the highest infection level the whole of the abdominal haemocoel was filled with the enlarged coiled salivary glands. Though hypertrophy occurred throughout the length of the salivary gland, the greatest enlargement occurred only in the area located in the abdomen. Some of the tsetse dissected for observations on the salivary gland had also enlarged midgut cells with symptoms of infection similar to those on the salivary glands.

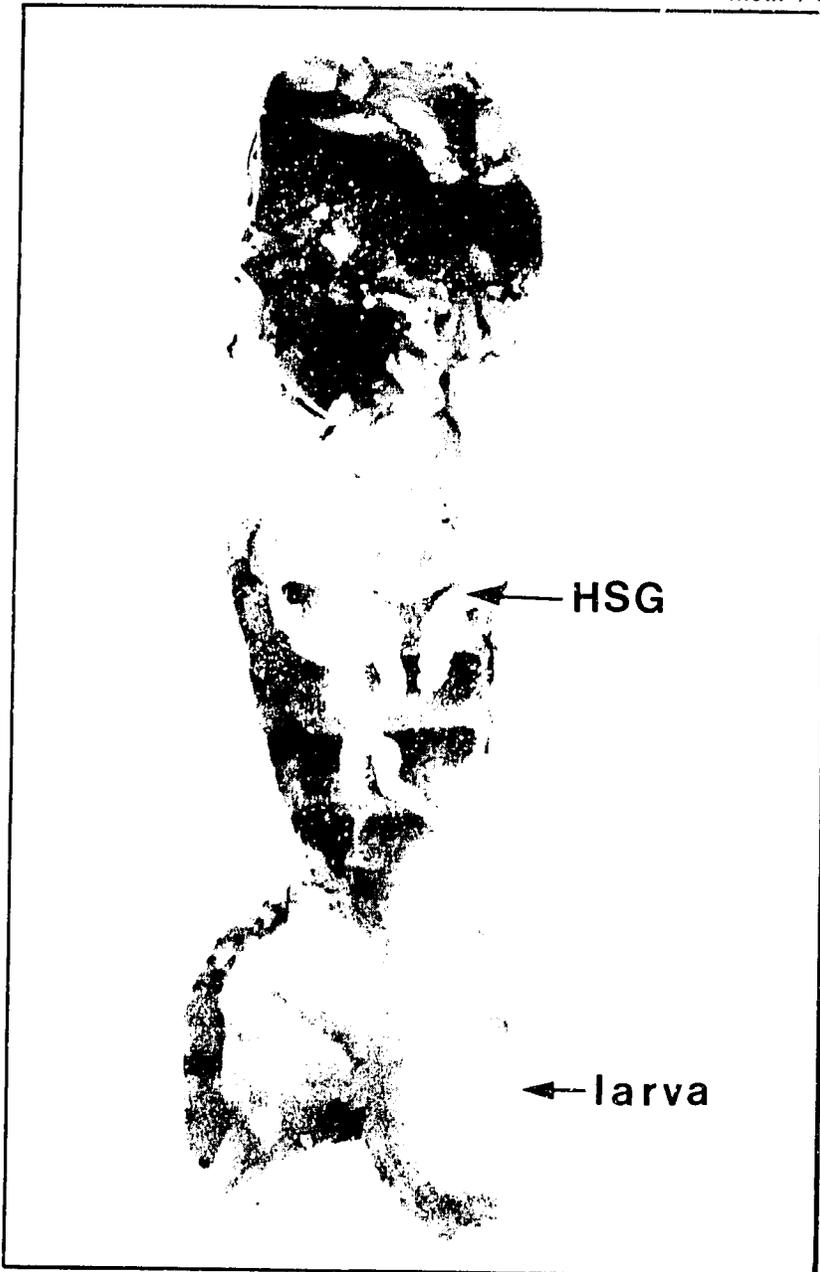


Fig 6. Hypertrophied salivary glands (HSG) in *Glossina pallidipes*. The uterus contains a full grown larva.

Field prevalence of hypertrophied salivary glands in the different trapping sites: Hypertrophied salivary glands were present in tsetse from all the study areas where the present investigations were carried out. However, the number of tsetse with HSG varied from one trapping site to the next, even in places of close proximity. Hence in July, Muhaka Forest had mean 2.7% infection, whereas 4 km to the east at Mwabungu, mean level of infection was 15.6%.

Relationships between level of infection and tsetse density: In Mwabungu, there was an inverse relationship between HSG incidence and tsetse density. Mean tsetse catches per trap per day showed a reduction in tsetse numbers during the months March and October when HSG level was highest. A comparison between tsetse population (mean tsetse catch/trap/day) and mean monthly per cent HSG level did not show an inverse relationship at Muhaka,

where there was a low HSG incidence.

The monthly observations on tsetse with hypertrophy of the salivary gland show variation in proportion of *Glossina* with salivary gland hypertrophy, although the pattern of infection does not correspond to any climatic variations, such as rainfall and temperature. Peaks of HSG prevalence are most likely to correspond to peaks in the source of infection.

It is not clear how infection gets to the tsetse in the wild. Two possible pathways have been suggested: a transovarial and/or transovum pathway in which the GLPs recirculate within the tsetse population, and a tsetse-animal host-tsetse pathway in which VLPs circulate through a vertebrate which acts as a reservoir for VLPs. It has been shown that teneral tsetse can be infected both by microinjection of membrane-filtered VLPs into tsetse haemocoel and by oral infection of tsetse, hence raising the possibility of oral infection through blood meals in the wild. Large numbers of the rod-shaped VLPs have been observed in the negatively-stained salivary gland secretions of HSG in the infected tsetse, further increasing the likelihood of tsetse-vertebrate host-tsetse passage of VLPs.

In the context of microbial control, VLP infection and the subsequent symptom of HSG have been associated with reproductive anomalies in both males and females. A high HSG incidence may have some effect on the reproductive capability of a tsetse population. The inverse HSG level/tsetse density relationship at Mwabungu may therefore be partly accounted for by VLP as a sterility factor in tsetse. In contrast a comparison between tsetse population (mean tsetse catch/trap/day) and mean monthly per cent HSG level did not show an inverse relationship at Muhaka, where there was a low HSG incidence.

### Development of immunological methods for the control of *Glossina*

The widespread use of chemical insecticides for the control of tsetse has been disappointing because it has led to considerable environmental pollution and development of insecticide resistance in insects. Furthermore, the effect of insecticides on insects is non-selective, thus causing the destruction of useful non-target insects as well. An immunological method of tsetse control using antigens derived from tsetse for vaccinating host animals, if successful, will overcome all the above mentioned problems. In our laboratory, different tsetse tissues have been used as sources of antigen for immunizing rabbits against tsetse and this paper reports briefly on our preliminary observations.

Groups of 50 female newly emerged *Glossina morsitans* were maintained for two successive generations ( $F_0$  and  $F_1$ ) on rabbits immunized with homogenates of whole crude tsetse (WCT), engorged tsetse guts (ETG), gravid tsetse uteri (GTU) or on untreated control rabbits. It was confirmed both by immunodiffusion and by immunoelectrophoresis that these tsetse-derived antigens provoked a strong antibody response in the inoculated rabbits. No mortality occurred in the flies maintained on the immunized rabbits but a decrease in fecundity and in the mean pupal weights, and a slight increase in pupal mortality were observed. For the two fly generations ( $F_0$  and  $F_1$ ), total fecundity decreases of approximately 31%, 36% and 47% were observed in the CWT, ETG and GTU groups, respectively when compared with the controls. The overall decreases in the mean pupal weights for  $F_0$  and  $F_1$  generations were approximately 12%, 9% and 12% for CWT, ETG and GTU groups respectively. No pupal mortality occurred in the pupae of  $F_0$  flies, but in the  $F_1$  generation alone, a pupal mortality of approximately 7%, 18% and 8% were observed in the CWT, ETG and GTU

groups, respectively.

These observations strongly suggest that this immunological technique, if improved, e.g. by using purified insect antigens may prove to be useful for tsetse control. This view is strengthened by the fact that different fly generations feeding on the same immunized animals are affected. This suggests that in an isolated tsetse-infested

area, if the animals favoured by the flies as the source of blood are immunized, the fly population will presumably decrease progressively. Lastly, it should be remembered that these data have been obtained from a laboratory maintained in almost ideal conditions and that in the field where adverse conditions exist, the pathological effects on both flies and pupae will be more pronounced.

# CHEMISTRY AND BIOCHEMISTRY RESEARCH UNIT

## Introduction

Some scientists were involved in synthesis of two components (13, 17 and 15, 19 — dimethylheptatriacontane) of the newly discovered sex-stimulant pheromone of *Glossina pallidipes*. This marked the beginning of synthetic chemistry for the ICIPE and the Unit hopes to equip itself to expand these activities even further. Professor Kenji Mori (Tokyo) and Michael Bentley (Maine) will assist us with pheromone chemistry in 1982.

The Finnigan mass spectrometer, now with capillary GC interface and a new electron multiplier, has improved sensitivity. We hope to expand our capabilities many fold when the data access and storage facility is acquired; manual acquisition and calculation limits the output of the Unit severely. Nevertheless,

besides identifying (with GC/MS) the branched alkanes mentioned above, the trail pheromone (Cembrene A) of one of the termites *Trinervitermes bettonianus* has been characterised and is being studied at Kajiado — another new development in the ICIPE!

Not to be outdone by the Chemists, the Biochemists have been hard at work proving for the first time that insects, like vertebrates, have inactive forms (zymogens) of their digestive enzymes and they have activators (enterokinases) to initiate digestion of the first meal. In tsetse, trypanosomes ingested with the first blood meal may delay activation of trypsin.

Continuing the vitellin research described in the last report, the ecdysteroid binding protein from *R. appendiculatus* (the first to be found in arthropod eggs)

Table 1: BIOLOGICALLY ACTIVE PHYTOCHEMICALS ISOLATED RECENTLY BY ICIPE CHEMISTS

Population no.	Plant name	Compound isolated	A c t i v i t i e s					
			Antifeeding	Antifungal	IC	IGR	IR	Other
1	<i>Plumbago capensis</i>	Plumbagin	++	--	--	--	--	+
		Juglone	+	+	--	--	--	+
2	<i>Rhabdosia spp.</i>	Diterpenoids	--	--	--	+	+	+
6	<i>Clausena anisata</i>	Imperatorin	--	--	--	--	--	+
		Xanthoxyletin	+	+	--	--	--	+
		Chalepin	--	+	--	--	--	+
		Xanthyletin	+	+	--	--	--	+
		Tecleantine	--	+	--	--	--	+
		Milicopicine	+	+	--	--	--	+
		Methyl tecleantine	--	+	--	--	--	+
14	<i>Milletia thonningii</i> <i>Crysanthemum cinerariaefolium</i>	Isoflavones 1-4	Not determined yet					
		Pyrethrin I	--	--	+	--	+	+
		II	--	--	+	--	+	+
		Jasmolin I	--	--	+	--	+	+
		II	--	--	+	--	+	+
		Cinerin I	--	--	+	--	+	+
		II	--	--	+	--	+	+
13	<i>Aspilia pluriseta</i>	(-)-kaur(16)en-(19)ic acid						
15	<i>Tovomita mangle</i>	Tovophenone A and B	No determined yet					

Key IC = insecticidal; IGR = insect growth regulator; IR = repellent; Other = bactericidal etc.

cross-reacted with antibodies from a rabbit previously immunised with whole engorged female homogenate. This rabbit was resistant to tick infestation in that 80% of the eggs from ticks which fed on the rabbit were non-viable. Ellie Osir, now with John Law in Tucson will be continuing research on the binding protein. Our consultant for 1982, Professor Jan Koolman is advising us on the latest affinity chromatographic methods for purifying the protein.

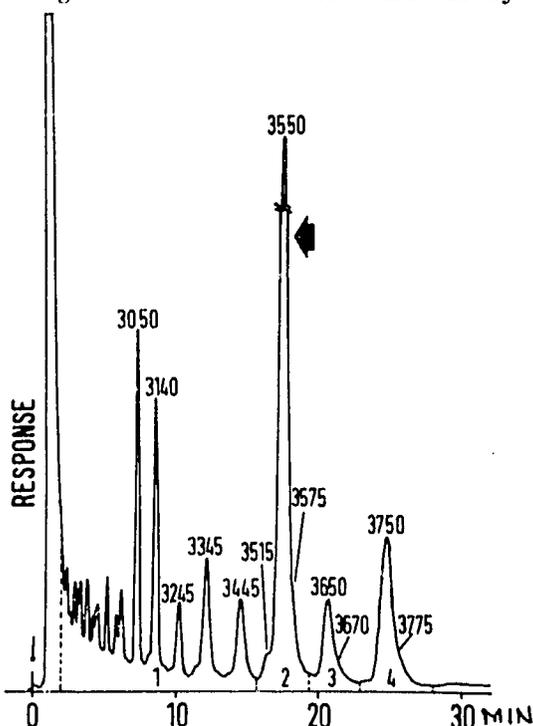
Ecdysteroid titres during the life-cycle of *Glossina morsitans morsitans* and the mode of action of sterol abortifacients fed on pregnant females were reported to the O.A.U/S.T.R.C. meeting in Arusha.

Wilber Lwande will soon be back from Rome to assist with the natural products chemistry of the unit. We continued to isolate and test new natural compounds for their effect on fungi and on behaviour and development of our target pests (see Table 1). In this context, collaboration with Dr. Ramesh Saxena at IRRI (Manila) continues, on phytochemicals which affect the brown planthopper. Optimisation of the known feeding deterrents for the African armyworm now occupies the minds of our chemists, but it is too soon to report on this aspect.

#### THE SEX-STIMULANT PHEROMONE TSETSE *GLOSSINA PALLIDIPES* (AUSTEN)

We have isolated the sex-stimulant pheromone(s) of *Glossina pallidipes* from the female cuticular waxes and identified it as a mixture of dimethylpentatriacontanes on the basis of gas chromatographic retention times and mass spectroscopic data. The pheromone was tentatively identified as a mixture of 13, 17- and 15, 19-dimethylpentatriacontanes, the former predominating. Figure 1 shows a gas chromatogram of the hydrocarbon fraction from mature females. These hydrocarbons, when deposited on dead male decoy flies washed free of their

own cuticular waxes, elicited sexual responses from live male *G. pallidipes* after coming into contact with the decoy.



Gas chromatogram of the cuticle hydrocarbons of female *G. pallidipes*. Column conditions: 5% OV-101 (3 m., 2mm. i.d.), isothermal at 320°C, carrier gas nitrogen (20 ml/min.) (fraction 2 active in sex-stimulant bioassay — see Insect Sci. Application 2, 181 (1981))

After fractionation of the hydrocarbons into four fractions, only one fraction containing dimethylpentatriacontanes elicited full sexual activity in the bioassay. The structures 13, 17- and 15, 19- dimethylpentatriacontanes were initially proposed from the mass spectroscopic data. These have been synthesised in five steps from octadecan-1-ol and hexadecan-1-ol respectively. Laboratory bioassay of these compounds indicated that both were active in stimulating mating responses from test males, the former isomer being more active at low doses. Comparison of the synthesised compounds with the natural compounds from fraction 2 showed that a third compound predominated in the natural fraction. Further mass spectral studies led to the proposal of the 13, 23-dimethylpentatriacontane isomer which is presently being synthesised for bioassay and chemical

comparison.

CHEMICAL IDENTIFICATION OF THE  
TRAIL PHEROMONE OF THE TERMITE  
*TRINERVITERMES BETTONIANUS*

Natural trails of worker *Trinervitermes bettonianus* laid on a filter paper substrate and extracted with hexane show trail following activity in a Figure 8 bioassay. Gas chromatograms of these extracts indicated a complex mixture of components largely of terpenoid composition. After fractionation only one fraction was found to be highly active at very high dilution.

The principal active component in this fraction was identified as a single peak. The quantities of material available for analysis by extraction of worker trails, however, were extremely small. Female alates, on the other hand, are known to contain larger quantities of trail active components (usually assumed to play an important role in mating behaviour, also). The hexane extract of female alate sternal glands was chromatographed. Again the same fraction as before was extremely active in the bioassay. The single component in this fraction also corresponds to the previous component from workers (by GC coinjection). In comparison, male alate sternal gland extracts contain very little of this component.

Similarly, whole body extracts of female alates contain large quantities of the active component from which milligram quantities of the compound were isolated for chemical identification. As expected, whole body extracts of male alates contained very little of the active compound. Soldiers, also, contained little of the trail compound.

Chemical and spectroscopic data indicated the diterpene hydrocarbon structure cembrene-A. The mass spectrum showed a molecular weight of 272, with a base peak ion at  $m/e$  68 which is characteristic of this structure. The hydrogenation pro-

ducts of the trail compound compare well with the hydrogenation products of the related compound, cembrene.

ECDYSTEROID TITRES DURING THE  
LIFE CYCLE OF *GLOSSINA*  
*MORSITANS MORSITANS* WESTWOOD

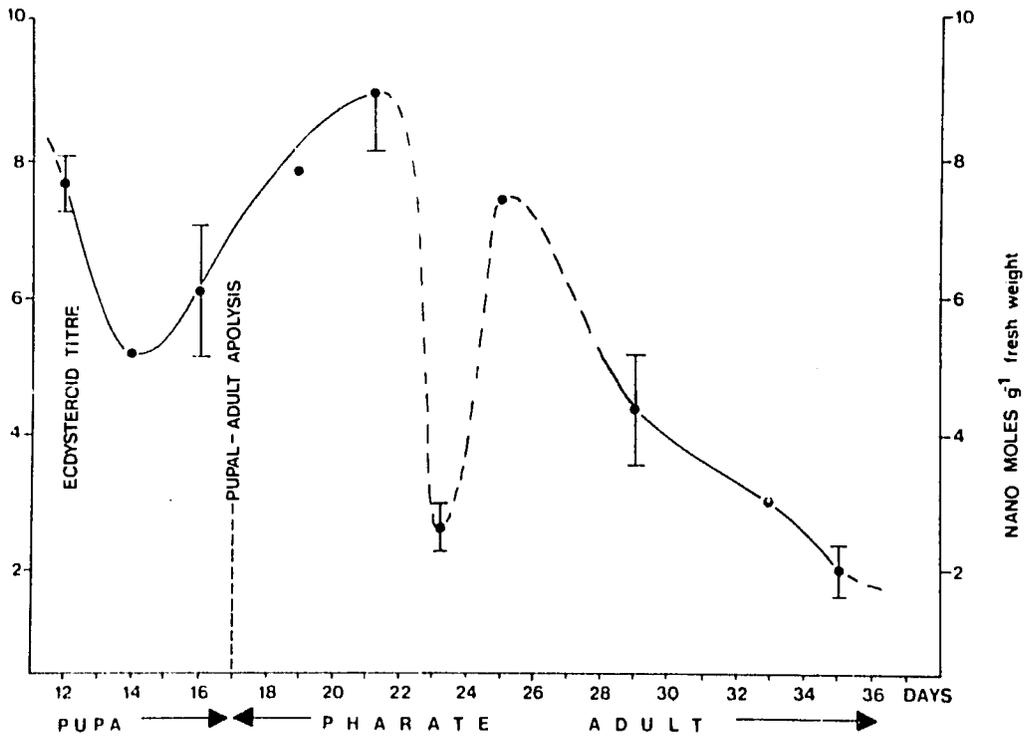
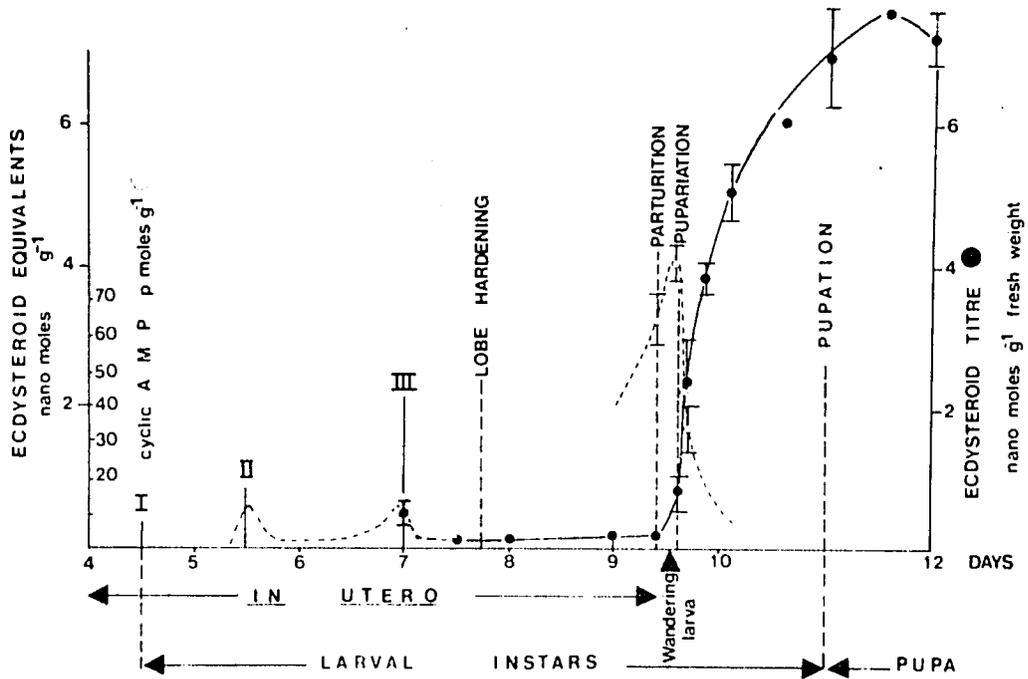
Ecdysteroids could not be detected in *G. m. morsitans* stage III larvae during the hardening of the polypneustic lobes which occurs 2 days before pupariation (Whitehead, 1976). Nevertheless, ecdysteroids were later detected by radioimmunoassay (RIA) in the bodies and haemolymph of teneral males of this species (Gee, Whitehead and Koolman, 1977). This report describes the use of RIA, gas chromatography with electron capture detector (GC/ECD) and high performance liquid chromatography (HPLC) to monitor ecdysteroids during the life cycle of *G. m. morsitans*.

The levels of free ecdysteroids present during development of *G. m. morsitans in utero* after parturition and pupal - adult apolysis, determined by RIA, are shown in Fig. 2. 4. 10 hr after pupariation the levels are  $4.8 \pm 0.6$  nmoles  $g^{-1}$ . Of this less than half is ecdystero ( $1.7$  nmoles  $g^{-1}$ , together as determined by GC/ECD.

The first peak (ca.  $7.6$  nmoles  $g^{-1}$ ) appears 40 - 44hr after larviposition. Initially there appears to be a rise in cyclic AMP titre after larviposition but this is followed by a drastic fall from 82 to 22 pmoles  $g^{-1}$  fresh weight of larva during the 2 to 3 hr when the ecdysteroid titre begins to rise.

A second maximum of  $8.9 \pm 0.8$  nmoles  $g^{-1}$  fresh weight is reached 10 days after pupariation. Preliminary HPLC analysis at this stage of development indicates that ecdystero ( $5.1$  nmoles  $g^{-1}$ ) and ecdysone ( $0.5$  nmoles  $g^{-1}$ ) are both present in pharate adults.

STAGES OF DEVELOPMENT OF *G. morsitans*



Ecdysteroid titres measured by R.I.A. (see text) throughout the major stages of development of *G. m. morsitans* reared at 25°C. (Vertical bars represent 2 x S.E.M. around the means while elsewhere the mean of two determinations is plotted)

The level of ecdysteroids is so low during the third larval instar that RIA cannot detect them except on the sixth to seventh day after ovulation (second cycle) when the titre was  $0.4 \pm 0.1$  nmoles g<sup>-1</sup>.

Topical application of 1.0 ug of Dimilin in 1 ul of acetone to the thorax of females after the first larva was deposited, resulted in the birth of live larvae which were unable to pupariate normally. Ecdysteroid

levels in these offspring, 10 hr after larviposition were  $3.9 + 1.1$  nmoles  $g^{-1}$ .

### FIRST REPORT OF AN ENTEROKINASE IN THE TSETSE GUT

As the concentration of enterokinase in adult flies is likely to be extremely low the enzyme was partially purified from 21 — day old puparia. Day 21 puparia were chosen because at this point we have shown trypsin-like activity is lowest. The activity increases again from day 22 onwards (see also Langley, 1967).

### RESULTS AND CONCLUSION

Three major protein peaks were eluted from the sepharose CL-6B column. Enterokinase-like activity was detected in the first peak together with AP. From the affinity column, enterokinase-like activity was eluted in the last peak. The eluted enzyme hydrolysed crystalline bovine trypsinogen to trypsin, as shown by the resulting hydrolysis of  $\alpha$ -benzoyl-DL-arginine paranitronilide hydrochloride (BAPNA). Boiling abolished the activity of the enzyme, but this appeared to be regained on standing at 4°C. If the presence of the enterokinase in teneral flies can be established, it will be of interest to see whether ingested trypanosomes can inhibit this enzyme and so prevent lysis of their own surface coat.

### ECDYSTEROID BINDING PROTEIN FROM RHIPICEPHALUS APPENDICULATUS

Research on proteins of ticks was begun because of their potential antigenic properties for inducing resistance in live-

stock against infestation by these vectors of disease. As will be seen from 1980, ICIPE Annual Report, the major proteins present in the eggs of the East Coast Fever Vector have been separated and partially purified by chromatography on Sepharose CL-6B. It was found that the Vitellin (M. Wt.  $\frac{1}{2}M$ ) is associated non-covalently with haeme derived from the host's haemoglobin. This report is concerned with a glycolipoprotein of M. Wt. 106,000 which has properties which should greatly enhance its antigenic value. Once the tick host has built up antibodies to it, the fecundity of females should be drastically reduced.

By electrofocussing we established (a) the isoelectric point ( $pI=8.2$ ) and (b) that engorged females possessed it in their haemolymph whereas males did not. Therefore, DEAE cellulose was used to adsorb the protein from the eggs of ticks previously injected with 23, 24- $^3H_4$ , ecdysone or 4- $^{14}C$ , cholesterol to show that it could carry steroid to be used by the developing embryo. It was soluble in methanol and in n-butanol used to extract ecdysteroids from *R. appendiculatus* eggs (see 1979 ICIPE Annual Report). When the complex of 4 or 5 proteins with  $pI$  8.0—8.4 was cross-reacted with antibodies from a rabbit previously immunized with whole engorged female homogenate, a strong precipitin line was seen. Only 15% of the eggs laid by ticks feeding on this rabbit developed. Therefore, these proteins are being separated by chromatofocusing with polybuffer to establish which of them binds ecdysteroid and which of them is most antigenic and effective in reducing infestation by ticks on the host.

# HISTOLOGY AND FINE STRUCTURE RESEARCH UNIT

## INTRODUCTION

The Histology and Fine Structure Research Unit, is contributing to the understanding of certain structure among the target insect pests and diseases. The Unit has aided the experimental biologists at the ICIPE in the determination of morphological changes in the secretory cells and target tissues brought about by drug or hormone treatment; cytochemical localization of substrates in important tissues, description of the morphology of the reproductive organs, morphology of sensory receptors which compliment electrophysiological and behavioural studies.

The range of subjects which have received histological and fine structural analyses over the last year include the studies on insect sensory systems. In insects, perception of environment involves the reception of external stimuli by specialized sensory receptors. Within the past year the SEM has been used appreciably in the study of sensillae types present on tsetse tibia. Studies on, their response to various types of stimuli are underway. Work is in progress on receptormapping to find out whether we can identify the olfactory and gustatory sensillae on several species of stemborers. Other studies were conducted to evaluate surface morphology of the various biotypes of the rice brown planthoppers. Emphasis in this work has

been on body parts possessing receptors.

A histochemical and ultrastructural study of the salivary glands of *Glossina morsitans* has been initiated as a contribution to a better understanding of the physiology of the salivary gland system.

Some evaluation on the topology of *Phlebotomus* sandfly eggs has been made to reveal details about the chorion.

## SPERMATHECAL ULTRASTRUCTURE OF THE INSECT *GLOSSINA* *MORSITANS MORSITANS*

In *Glossina morsitans morsitans* females, the spermatheca is characteristically an ectodermal derivative, lined with cuticle. The electron microscope reveals four distinctive zones in a representative section of a spermathecal duct: (1) a layer of muscle (Mc, Fig. 1), (2) a layer of columnar epithelium (Ep, Fig. 1), (3) a thick cuticle (C, Fig. 1) around the duct lumen.

The cells lining the spermathecal duct possess extensively folded apical plasma membrane. Laterally, cells interact by desmosomes. Bundles of microtubules run from the base to the apex. These are tied in with basal hemidesmosomes and the apical cuticle. The possible role of the spermathecal duct in sperm movement is being investigated in the light of epithelial ultrastructure.



# SENSORY PHYSIOLOGY RESEARCH UNIT

## INTRODUCTION

In insects, behaviour is to a large extent stereotyped and thus predictable. This predictability permits development of methods of insect pest control by manipulation of behaviour. Since behaviour begins with the reception of one or more signals from the environment, knowledge of the initiation of the various components of behaviour requires a study of the function of the various insect sensilla. Knowledge has to be gained of the way in which stimuli are detected and sensory responses initiated. Moreover, the patterns of sensory responses and their consequent behaviour have to be studied.

The Sensory Physiology Research Unit provides services to analyse the role which sensory organs play in behaviour and to understand the range of environmental factors which influence behaviour. Using electrophysiological and behavioural methods, investigations are undertaken in close collaboration with scientists of other Research Programmes working on target insects of the ICPE. Major attention is paid to chemocommunication, as chemical factors appear to be decisive in host and mate selection in most insects, but the Unit also investigates other senses when these are suspected to play an important role in a target insect's behaviour.

In 1981 two new electrophysiological set-ups in addition to the two already present, were built and experiments started with chemoreceptors of cropborers, stem-borers, African armyworm, and tsetse. The electrophysiological, behavioural and morphological studies on sensory organs in ixodid ticks and studies on acoustic communication in tsetse flies were continued. Morphological investigations have been initiated to elucidate the structure of the sensory organs on the mouthparts and antennae of stemborer larvae and adults,

and of the alleged pheromone receptors in male tsetse flies, which are supposed to be present on the tibiae.

## TICKS

It has been shown that on the first pair of legs of ixodid ticks different types of sensory organs are present. Thermoreceptors were found: a decrease in ambient temperature leads to increase in activity; gustatory receptors responding to sodium chloride; and to olfactory receptors. The latter have been studied most extensively using the 'tip-recording' technique, in which the tip of an olfactory hair is cut off with microknives to obtain electrical contact with the olfactory cells in the lumen of the hair.

The olfactory sensilla responded to the tick sex pheromone 2, 6-dichlorophenol and to the odour of washes from larvae and adult ticks. Subsequent behavioural tests indicated that the washes are repellent to adult ticks. In collaboration with the Chemistry and Biochemistry Research Unit investigations are in progress to elucidate the chemical composition of the deterrent.

## TSETSE

Sound production in *Glossina morsitans morsitans* shows a rhythm. Young flies in a 12 hours light: 12 hours dark cycle show a U-shaped diurnal pattern of singing with peaks in the morning and later afternoon and little singing around midday. In mature flies singing mainly takes place in the morning.

During feeding, sound production is negligible, but most flies sing immediately after feeding, up to about 30 min after engorgement. Semi-gorged flies seldom produce sound. During courtship, most flies only sing during the first 3 min of

mating and for about half an hour after separation. It is assumed that singing may attract other flies, and investigations to determine the behavioural responses of tsetse to the sounds produced by their conspecifics are currently being conducted.

Very recently investigations have started on the reception of sex pheromones and other chemical stimuli in *G. morsitans morsitans* and *G. pallidipes*. We have succeeded in recording olfactory responses (electroantennograms) from tsetse antennae. This offers a rapid method of screening odours for their activity at the receptor level. Experiments are being done to systematically screen a large number of synthetic and natural chemicals, taking into account the results obtained in field studies and the molecular structure of the substances.

In addition to the sensillae on the antennae, other possible sensory organs involved in the detection of chemicals are being investigated. Priority is paid to various receptors on the tarsi and to the alleged tibial receptors. The electrophysiology as well as the ultrastructure of these organs are being studied.

These studies are undertaken in co-operation with the Tsetse Research Programme, the Chemistry and Biochemistry Research Unit, and the Fine Structure Unit.

### CROPBORERS

In the sorghum shootfly, *Atherigona soccata* Rondani, there is evidence that a deterrent contact pheromone is associated with the glue with which the females attach their eggs to the leaves.

The effects of the deterrent on the cells in tarsal contact chemosensory hairs are being determined by electrophysiological recording from these hairs in gravid female shootflies.

It appeared that egg wash elicited action potentials from a separate type of cell. In addition, the deterrent appeared to have an excitatory effect on the salt sensitive cells (which generally mediate rejection behaviour) and it also seemed to inhibit the responses of the sugar sensitive cells (which mediate acceptable behaviour). 4-hydroxy benzoic acid and 4-hydroxy methylbenzoate, which originally were thought to be the active compounds in the deterrent, were found to be electrophysiologically inactive. These results were further strengthened by behavioural experiments, which proved that these substances did not have any deterring effect.

These studies are undertaken in co-operation with the Sorghum Shootfly Programme and the Chemistry and Biochemistry Research Unit.

### STEM BORERS AND AFRICAN WORM ARMYWORM

Studies have been initiated to identify the chemical factors which determine susceptibility of crops to stem borer and African Armyworm adults and larvae. Olfactory and gustatory responses of *Chilo partellus*, *Eldana saccharina*, and *Spodoptera exempta* are being recorded during stimulation with various plant substances. Moreover, scanning electron microscopical investigations are being done on the chemosensory sensilla on the mouthparts of larvae and on the antennae of adults.

It appeared that in the palps of *Chilo* and *Spodoptera* and in the lateral and medial sensilla styloconica of *Chilo*, *Eldana* and *Spodoptera*, specific salt and sugar cells are present, the latter being very sensitive to sucrose. All plant substances primarily stimulated the salt cells. In *Eldana*, no significant differences in the stimulatory effects occurred between the plant substances. This is in contrast to *Chilo* and *Spodoptera*, in which vanillic acid appeared to strongly stimulate the lateral sensillum styloconicum.

These studies occur in co-operation with  
the Bases of Plant Resistance Research

Programme and the Fine Structure Unit.

# BIOASSAY RESEARCH UNIT

## Services Provided by the Unit

The Unit has continued to provide bioassay services to many of the Programmes and Units in the Centre. Most of the services have been given mainly to three programmes, the Livestock Tick Research Programme (Moulting Hormone determinations), the Grassland Termite Programme (Juvenile Hormone assays) and the Chemistry and Biochemistry Research Unit, with which the Unit carries out a collaborative programme on the investigation of African plants for antifeeding, larvicidal, fungicidal, repellent and moulting hormone activities. The Unit has recently established a mosquito repellency test to augment the tick climbing test for the study of repellents from pyrethrum flowers. This bioassay is a standard test and involves taking 3 - 4 day old female mosquitoes (*Aedes aegypti*), which have been fed only on sugar solution since emergence, and offering cleaned human arm treated with a known concentration of the test material in 70% ethanol. The arm is held in the cage for 5 min and withdrawn. The mosquitoes are immediately removed and the number of engorged females recorded. An arm treated with only 70% ethanol and tested similarly serves as the control. The test is replicated four times and the results analysed.

## Investigation of Plants for Bioactive Compounds

The isolation and characterization of two bioactive coumarins, imperatorin and xanthoxyletin from the stem-bark of *Clausena anisata* (Rutaceae) was reported last year. Both compounds showed feeding deterrence activity against the African armyworm (*Spodoptera exempta*) larvae whereas xanthoxyletin also possessed fungicidal properties against *Cladosporium cucumericum*.

Further work on the same plant led to the isolation of two more bioactive coumarins, xanthyletin and chalepin. The compounds were characterized by their UV, IR, NMR and mass spectra. Xanthyletin showed both feeding deterrence (100 ppm) and fungicidal activities, whereas chalepin possessed weak fungicidal properties.

The methanol extract of the stem-bark of another Rutaceae, *Teclea trichocarpa* (Eng) Engl, was found to be moderately active as a fungicide and a feeding deterrent. Fractionation of the extract by column and thin layer chromatography on silica gel led to the isolation of three acridone alkaloids as active components. The compounds were identified as a melicopicine (feeding deterrent), teeleanthine (feeding deterrent and fungicide) and 6-methoxy-tecleanthine (fungicide from their spectral data.

# INSECT AND ANIMAL BREEDING SERVICES

## BREEDING OF TSETSE

The *Glossina morsitans morsitans* colony was expanded considerably in 1981. The colony was maintained at 25°C and 70% using rabbits as host animals.

Tsetse emergence per month ranged from 6831 00 and 7151 00 to 10,174 00 and 9566 00. The mean monthly emergence being 8228 00 and 8108 00. Mortality in teneral flies was negligible and in mated 00 did not exceed 0.9% per day (mean mortality was 0.75%).

The mean pupal production per female per month was 2.2 and samples of pupae weighed during the year gave a mean pupal weight of 29.4 µgm.

A total of 29,201 00; 30,035 00 and 5492 pupae were supplied for experimental use by the ICIPE scientists and other research organisations, an increase of more than 100% on the previous year.

## BREEDING OF *CHILO PARTELLUS*

The small *Chilo partellus* colony was maintained throughout the year, at the ICIPE, Chiromo on an artificial wheat germ based diet. The insects were supplied for sensory physiology work and used for experimentation with other artificial diets.

With the development of the ICIPE Mbita Point Field Station, there is now a demand for large numbers of crop boring insects for laboratory and field studies.

With the assistance of the Bases of Plant Resistance and Crop Borers Programmes, it is planned to establish insect mass rearing facilities at Mbita Point in 1982.

## BREEDING OF AFRICAN ARMYWORM, *SPODOPTERA EXEMPTA*

Demand for the African armyworm declined in the past year. The colony was therefore reduced and moved to smaller facilities. It continues to flourish on a natural maize leaf diet and the problems previously experienced from viral and bacterial infections were overcome this year (1981).

Insects amounting to about 400 per month are supplied to the Chemistry and Biochemistry and Sensory Physiology Research Units.

## ANIMAL BREEDING

### Rabbits

A breeding stock of 80 to 90 females has been maintained throughout 1981. Survival in young rabbits has remained at around 90% and no major disease problems have been encountered. Demand for rabbits has also increased and the unit supplies 90 to 100 per month for research purposes.

### Rodents

Demand for rodents has fluctuated throughout the year but the colonies are still geared to produce an excess of 75 rats and 100 mice per month.

## MAJOR SEMINARS GIVEN AT THE ICIPE DURING 1981

SPEAKERS	SUBJECT
1. Dr. Pritam Singh Entomology Division Depart- of Scientific and Industrial Research Auckland New Zealand	Insect Rearing, Nutrition and Management of Laboratory Reared Insects
2. Professor W. H. R. Lumsden Department of Protozoology London School of Hygiene and Tropical Medicine England	Diagnosis of Salivarian Trypanosome Infection
3. Dr. P. A. Langley Tsetse Research Laboratory University of Bristol Bristol	Effects of Mating on Receptivity and Ovulation in Female <i>Glossina morsitans</i>
4. Dr. K. C. Binnington Department of Zoology Cambridge	Ultrastructure of the Tick Neuroendocrine
5. Dr. J. Hardie Imperial College London	Endocrine Aspects of Aphid Polymorphism
6. Professor F. C. Kafatos Harvard University U.S.A.	Control of Gene Expression During Develop- ment: Advances in Understanding of Eggshell Formation in Insects
7. Dr. P. A. Lawrence Medical Research Council Laboratory of Molecular Biology Cambridge	Compartment in Insect Development
8. Professor M. Locke Department of Zoology University of Western Ontario London Canada	The Epidermal Feet in Insect Morphogenesis
9. Dr. S. Yagi Laboratory of Applied Ento- mology Tokyo University of Agriculture & Fuchu Tokyo	Diapause and Phase Variation in Some Lepidop- terous Insects
10. Professor W. S. Bowers New York State Agricultural Experiment Station Cornell University New York	Insect Growth Regulators — Practice and Promise

11. Dr. L. Strong  
Department of Zoology  
The University of Bristol England  
Hormones in Insect Embryos
12. Professor Baccio Baccetti  
Institute of Zoology  
University of Siena Siena Italy  
The Dynein Proteins
13. Dr. John B. Kaddu  
ICIPE  
Cryobiology (Basic and Applied) and its Applications to Leishmaniasis Research
14. Dr. Joseph O. Olobo  
ICIPE  
Antibodies to *Leishmania tropica* Promastigots During Infection in Mice of Various Genotypes
15. Dr. G. Ole Moiyo  
ILRAD  
Glandular kallikreins — their structure and function
16. Dr. M.B.A. Nyindo  
ICIPE  
Some Impressions of Australian Biological Research
17. Dr. A. Hassanali  
ICIPE  
Dehydroamino Acids and D — Amino Acids in Peptide Antibiotics
18. Professor Robert B. Stewart  
Department of Microbiology and Immunology Queens University Kingston Canada  
Regulation of interferon by AMP and effect of interferon on C type particles
19. Professor David Goldsmith  
Department of Chemistry Emory Atlanta Georgia USA  
Recent Progress in the Synthesis of Insect Antifeedants
20. Professor K. N. Saxena  
University of Delhi India  
Scope of Manipulation of Insect Behaviour for Pest Management Programme
21. Dr. R. D. Dransfield  
University of Jos Nigeria  
Trapping Strategy for Tsetse
22. Dr. P. G. McDowell  
ICIPE  
Chemical Communication in Termites and Tsetse
23. Dr. D. A. Otieno  
ICIPE  
Repellent Principles of Pyrethrum Extract
24. Miss Mary Sampson  
Legon University Ghana  
The Life History of *Eldana saccharina* Walker on Sugarcane in Ghana
25. Professor J. L. Auclair  
International Rice Research Institute Manila Philippines  
Biochemical Evidence for the Feeding Sites of the Green Leafhopper (*Nephotettix virescens*) within Susceptible and Resistant Rice Plants

26. Dr. T. Y. Kaufmann

The Behavioural Biology Feeding Habits and  
Ecology of 3 Major Maize Stem borers of  
Nigeria

27. Professor G. Dauben  
Department of Chemistry  
University of California  
Berkeley USA

Diterpene Cembrenes

## LIST OF PUBLICATIONS FOR 1981

1. Arshad M. A. (1981) Physical and chemical properties of termite mounds of two species of *Macrotermes* (Isoptera, Termitidae) and the surrounding soils of the semi-arid savanna of Kenya. *Soil Sci.* 132 (2), 161–174.
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