



JOINT FAO/IAEA DIVISION

of Isotope and Radiation Applications
of Atomic Energy for Food and Agricultural Development



INTERNATIONAL ATOMIC ENERGY AGENCY
FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS

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INSECT & PEST CONTROL SECTION

NEWSLETTER

AND

INFORMATION CIRCULAR

ON

RADIATION TECHNIQUES AND THEIR

APPLICATION TO INSECT PESTS

No. 37

July 1986

SYMPOSIUM ANNOUNCEMENT

FAO/IAEA INTERNATIONAL SYMPOSIUM

ON

MODERN INSECT CONTROL : NUCLEAR TECHNIQUES AND BIOTECHNOLOGY

16 - 20 NOVEMBER 1987 : VIENNA

FORMAL NOTIFICATION TO BE PUBLISHED SHORTLY

IAEA TRAINEESHIP PROGRAMME

A 12-MONTH TRAINING PROGRAMME AT THE IAEA SECRETARIAT IS PLANNED TO START ON 1 JANUARY 1987.

PURPOSE : TO DEVELOP A CORE OF MANPOWER RESOURCES AND INCREASE THE NUMBER OF QUALIFIED CANDIDATES FROM DEVELOPING MEMBER STATES OF THE AGENCY FOR PROFESSIONAL POSITIONS.

APPLICATIONS MUST REACH IAEA THROUGH OFFICIAL GOVERNMENT CHANNELS BY 15 SEPTEMBER 1986.

MESSAGE

REPEAT

TO THE DIRECTOR

URGENT MESSAGE: PLEASE ACT NOW

Based on the response to the questionnaire which accompanied the 1971
issue of the Newsletter and information obtained from the 1971
survey, the following list needs revision.

Please note a few changes to complete the form below and mail it to us
immediately.

☐☐

DO YOU HAVE ANY OTHER INFORMATION ON THE NEWSLETTER
AND SURVEYING RESULTS?

YES

IF YES, PLEASE FILL IN THE SPACE BELOW IN BLOCK LETTERS
YOUR NAME AND ADDRESS IN FULL (PLEASE PRINT NAME).

NAME _____
ADDRESS _____

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WHEN RETURNING THIS FORM TO US, PLEASE ENCLOSE THE ALWAYS QUOTE YOUR
VEGETATION NUMBER WHICH APPEARS IN THE LIST OF THE NEWSLETTER FOR WHICH
YOU RECEIVE THIS FORM OF THE INFORMATION CIRCULAR. THIS RETURN
FORMERLY ASSISTED IN THE PROCESSING AND INFORMATION THROUGH THE
CIRCULAR.

PLEASE MAIL TO: Dr. R.A. Thompson

Head

Forest & Wetland Section

FAO

P.O. Box 100

6-1000 Vienna, AUSTRIA

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RESEARCH

PLEASE NOTE

The summaries of unpublished work often represent preliminary reports of investigations in progress and, therefore, such findings are subject to possible revision at a later date. The abstracts in this Information Circular should not be published or referred to in articles for publication without first obtaining permission from the authors.

Monthly Reports

Meetings

1. Advisory Group Meeting on "Institution and Data at Washington" at Washington at request of the Director, Forest Dept.

Public Meetings

Field Programs

Field

What's on in the Laboratory

ABSTRACTS OF RESEARCH PAPERS

Submitted Contributions

- A. Reproductive Biology
- B. Taxonomic Studies
- C. Insect Ecology and Behavior
- D. Pest Control

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I. INTRODUCTION

Publication Policy

The policy of the Joint FAO/IAEA Division in publishing the Information Circular is to emphasize the results of recent or on-going research on the use of radiation and radioisotopes in entomology. Therefore, emphasis is placed on unpublished data. For several reasons, we are unable to edit submitted contributions. These are reproduced by a photographic process, and therefore reflect faithfully, the author's care in preparing the material.

While emphasis is on unpublished data, we include, whenever possible, summaries of recently published papers. In that case, the material submitted should be no more than one page when typed double-spaced. (A form for submission of contributions is included in each distributed copy of the Information Circular; more can be provided on request).

The Newsletter is intended as a medium for informing our readers of "what is going on" and for providing an indication of "future plans". As far as possible, results or summaries of major activities during the preceding 6 months (e.g. field programmes, meetings, etc.) will be provided.

II. GENERAL INFORMATION

A. Professional Staff - Insect & Pest Control Section

Headquarters

D.A. Lindquist	Head, Insect & Pest Control Section
E.D. Offori	Technical Officer

Seibersdorf Laboratory

R.E. Gingrich	Head, Entomology Unit
A. Van der Vloedt	Tsetse Fly Investigations : Mass-Rearing
A. Economopoulos	Medfly Investigations : Rearing
J. Kabayo	Tsetse Investigations : Artificial Diets
E. Busch-Petersen	Genetic Sexing of Medflies
N. Bruzzone (FAO Associate Expert)	Medfly Rearing
T. El-Abbassi	Medfly Investigations : Diets

BICOT

M. Oladunmade	Project Leader and Officer-in-Charge, Field Operations
U. Feldmann	Technical Advisor (IAEA) and Team Leader, Laboratory Operations

PERU-MED

R. Rhode	Project Director
A. Perdomo	Medfly Ecologist

B. Entomology Laboratory

The IAEA has an international laboratory located at Seibersdorf, Austria, about 30 km from Vienna. A part of this laboratory, within the Agricultural Biotechnology Unit, is devoted to research involving the use of atomic energy in entomological research.

The primary objective of the entomology programme at the Agency's laboratory is to support and service the Joint FAO/IAEA Division's programmes on insect control. Thus, much of the research is concerned with problems that arise with field programmes.

The main thrust of research in Seibersdorf involves development of the Sterile Insect Technique (SIT) for pest control or eradication. Because of the dependence of this technique on efficient production of the target insect, much of the research at the laboratory involves development and improvement of mass-rearing techniques. Other major areas of activity include (1) development of methods of radiation sterilization for producing quality insects (in terms of sexual competitiveness and longevity); (2) investigation of handling techniques for large numbers of insects; and (3) supplying insects for field programmes.

In general, research is undertaken to:

1. develop and improve mass-rearing;
2. improve irradiation techniques;
3. develop methodology for "fail-safe" radiation sterilization;
4. develop methods for estimating "fitness" and sexual competitiveness of laboratory-reared, sterilized insects;
5. study possible genetic changes taking place during colonization and mass-rearing;
6. develop methods of shipping insects as pupae, either before or after sterilization;
7. develop release methods for large numbers of insects, both aerial and ground.

At the present time, the following species of insects are being reared at Seibersdorf:

1. Mediterranean fruit fly, Ceratitis capitata (Wied.).
2. Tsetse fly, Glossina palpalis palpalis.
3. Tsetse fly, Glossina pallidipes, Austen.

The entomology laboratory also assists entomologists in developing countries in planning or carrying out projects involving the use of the sterile insect technique (SIT). In addition, the laboratory serves as a training institution for entomologists from developing countries. These trainees are handled under the Agency's fellowship programme and usually spend from one to six months at Seibersdorf depending upon the needs of the country/institution requesting the assistance. In some cases, the fellows are supported to undertake scientific visits for up to 4 weeks.

Further information on this and other matters may be obtained by writing to:

Dr. D.A. Lindquist
Head
Insect & Pest Control Section
Joint FAO/IAEA Division
P.O. Box 100
A-1400 Vienna
AUSTRIA

C. Programmes of the Insect & Pest Control Section

1. Medfly

Among the most devastating pests of fruits in the world is the Mediterranean fruit fly, Ceratitis capitata. Research undertaken on this pest aims to:

- (a) develop less expensive larval and adult diets with particular emphasis on locally available ingredients (non-imported) from various parts of the world;
- (b) improve systems of rearing;
- (c) develop laboratory and field quality control techniques;
- (d) improve handling techniques for large numbers (100's of millions) of flies;
- (e) improve methods of releasing sterile flies in the field from aircraft;
- (f) provide emergency supplies of sterile medflies for field programmes;
- (g) develop genetic and mechanical sexing systems.

2. Tsetse Fly

The tsetse fly occurs only in Africa and is the sole transmitter of animal and human trypanosomiasis. The sterile insect technique which is currently being used to combat tsetse is supported by research to:

- (a) improve rearing technology with reduced handling of flies;
- (b) develop in vitro and in vivo feeding technology for mass-rearing;

- (c) develop methods for preserving blood (e.g. freeze-drying);
- (d) use blood additives for improving tsetse fly colony performance and offspring quality;
- (e) develop synthetic diet for tsetse fly rearing;
- (f) improve radiation sterilization techniques;
- (g) develop methods of estimating fitness of laboratory-reared, sterilized flies, study possible genetic and/or behavioural changes taking place during colonization and mass-rearing;
- (h) conduct cross-breeding experiments with morphological mutants;
- (i) develop laboratory and field quality control techniques.

D. Technical Co-operation and Assistance Programmes for which this Section has Responsibility

(a) <u>Medfly</u>	(b) <u>Tsetse</u>	(c) <u>Isotopes</u>	(d) <u>Others</u>
Peru	Nigeria	Kenya	Sri Lanka
Guatemala	Zambia		Iraq
Algeria	Ghana		Pakistan
Libya	Tanzania		Mauritius
Tunisia			

E. Experts and Consultants : January - June 1986

Name	Nationality	Location of Assignment	Dates and Task Performed
M. Hall	U.K.	Zanzibar (URT)	Ecological study of <u>G. austeni</u> (9 - 30 June)
P. Langley	U.K.	Vienna	Preparation of AGM Final Report (30 June - 4 July)

(Mr. P. Langley was a participant in the Advisory Group Meeting on "Nutrition and Diets of Haematophagous Arthropods with Emphasis on Tsetse Flies" held in Vienna from 23-27 June 1986.)

F. Trainees in Entomology : January - June 1986

Seibersdorf

(a) Tsetse Group

P.W. Mukiria	Kenya	85-09-16 to 86-01-13
O. Chalo	Tanzania	86-05-05 to 86-08-04
A. Pochet (cost-free)	Belgium	86-01-06 to 86-02-07

(b) Medfly Group

M.A. Ben Youssef	Libya	85-10-28 to 86-07-27
M.N. Elagal	Libya	85-10-28 to 86-08-27
E. Ahmed	Egypt	86-01-22 to 86-07-21
N. El-Badan	Egypt	86-01-22 to 86-07-21

III. NEWSLETTER

A. Special Features and Comments

Co-ordinated Research Programmes of the Insect and Pest Control Section

The Agency's Co-ordinated Research Programme (CRP) aims to bring together researchers in developed and the less developed countries, to solve well-defined problems, using nuclear techniques wherever applicable. Overall administration of the programme is the responsibility of the Agency's Contracts Administration Section. Institutions and/or individual researchers in Member States of the Agency are admitted upon application, provided that basic capabilities such as laboratory space and qualified staff exist at the institution for carrying out the research, and provided the work to be undertaken by the investigator is relevant to the Agency's overall programme. Institutions or researchers in developed countries normally participate in the programme on a cost-free agreement basis, while those from less developed countries are provided financial support on a cost-sharing basis, through a research contract. Research contracts are normally awarded for a period of one year, renewable for up to 3 years, or 5 years in exceptional cases.

In the area of insect and pest control, emphasis in recent years has been on developing and refining methodologies for applying the Sterile Insect Technique (SIT) as a major component of integrated pest management systems. Two economically important pest insects constitute the subject of study : the Mediterranean fruit fly (medfly), Ceratitidis capitata and the tsetse fly, Glossina spp.

Since ability to rear the target species in large numbers is one of the most critical requirements for the successful application of the SIT, research effort has been directed to improving methods of rearing the two insect pests. Through the combined efforts of researchers in various institutions around the world, it has become possible to rear the medfly in large numbers, so that rearing factories are able to produce millions of medflies per week. Considerable success has also been achieved with the rearing of at least 5 species or sub-species of tsetse flies. Procedures now exist for mass-rearing Glossina morsitans, G. m. submorsitans, G. tachinoides, G. p. palpalis and G. p. gambiensis, using animal hosts or artificial feeding systems.

Currently, the Insect and Pest Control Section has responsibility for the following Co-ordinated Research Programmes:

Tsetse

The programme title is "Development of Methodologies for the Application of the Sterile Insect Technique (SIT) for Tsetse Eradication or Control".

The objectives are to develop and improve technology for mass-rearing tsetse flies using in vivo and in vitro feeding methods including the use of artificial diets, and to develop procedures for ensuring production of good quality sterile males for release in the field.

Eight research contractors, including 7 from Africa and 5 agreement holders are participating in the programme. In addition to developing and testing diets for tsetse mass-rearing, participants in the programme undertake studies on such subjects as the effect of diet on trypanosome infection rates of laboratory-reared flies, the vectorial capacity of sterile males, ecology, mating behaviour, and survival of irradiated males. In the BICOT project area (see under Field Programmes), trypanosomiasis surveillance is undertaken to determine if there is a significant increase in trypanosomiasis in sentinel animals due to release of sterile males. A study is in progress on tsetse genetics, aimed at exploiting the merits of using hybrid sterility, and possibly conditional traits for tsetse population suppression. Considerable data have also accrued, during the past several years, on the use of traps, targets and other physical and chemical attractants and their effectiveness in modifying behaviour of tsetse in the field.

Medfly

The CRP on the "Development of Sexing Mechanisms in Fruit Flies through Manipulation of Radiation-Induced Conditional Lethals and Other Genetic Measures" aims to develop and refine procedures for eliminating females in the egg or early larval developmental stage so that males only are produced for sterilization and release in the field.

The advantages of such a sexing system are numerous. For example, the number of male insects mass-reared for release could be doubled at very little increase in cost. Alternatively, by eliminating the female neonate larvae, only half of the amount of larval diet would be required. Besides, substantial reductions in factory space and personnel, as well as disposal of waste larval medium, would result. A major advantage is that by not releasing females, damage from sterile stings to fruits would be eliminated, thus making the SIT attractive also to countries where such damage is unacceptable.

The procedure used by programme participants for developing a genetic sexing system is essentially a three-step process:

1. Selection of a conditional lethal mechanism;
2. Characterization of the mode of inheritance of this mechanism; and
3. Translocation of the dominant allele of this mechanism to the male determining chromosome.

Both physical and chemical agents are being tested for inducing chromosome and point mutations in medflies following similar studies of other dipterids. Thus, the investigations involve induction and isolation of temperature-sensitive, alcohol-sensitive and insecticide-sensitive lethal factors. Other studies aim at isolating morphological mutants (strains) based on pupal or eye colour. Investigations are in progress to develop female killing systems by use of conditional lethals linked to the male Y-chromosome so as to render the males immune and the females susceptible to environmental factors that could be applied early in the life cycle.

Medfly Trapping

A new programme has recently been initiated entitled "Standardization of Medfly Trapping in Relation to the Sterile Insect Technique (SIT)". To date, no standardized trapping procedures have been developed that are applicable under varying climatic conditions for use in medfly SIT programmes. Therefore the objective of the programme is to develop a standard trapping technique for use with the SIT for evaluating medfly densities by evaluating trimedlure-baited traps under various ecological conditions.

Participants in the programme will include researchers from representative areas around the world where the medfly constitutes a major problem. Institutes in Latin America, the Mediterranean basin, Middle East and Africa will be encouraged to participate. Using a common protocol, each investigator will conduct the same trapping experiments using the same trap design and attractants in medfly infested areas of his/her own country. The results will be assembled, computerized and analyzed statistically so that valid comparisons can be drawn between trap and lure efficiency in the different countries or areas.

B. Meetings

1. Advisory Group Meeting on "Nutrition and Diets of Haematophagous Arthropods with Emphasis on Tsetse Flies"

A 5-day meeting was organized in Vienna (23-27 June 1986) to advise the Joint FAO/IAEA Division on the future research relating to the nutrition and diets of blood-sucking insects, as related to the programme of the Joint FAO/IAEA Division.

The meeting was attended by 7 invited experts and staff members of the Insect and Pest Control Section and the Seibersdorf entomology laboratory. Participants identified problem areas with regard to the mass-rearing of blood-sucking arthropods, and in particular the tsetse fly which is the current subject of interest to the Joint FAO/IAEA Division.

In discussions following formal presentation of papers, potential areas of research were identified for emphasis in the future. The meeting stressed the need for continuing and expanding research on tsetse diets and nutrition aimed at producing good quality tsetse flies for use in sterile insect technique (SIT) programmes.

Recognizing the complexity and sophistication of the research approaches for developing artificial diets, the Advisory Group endorsed the active co-operation between researchers elsewhere and those of the Joint FAO/IAEA Division's tsetse laboratory in Seibersdorf.

Areas identified for research emphasis include investigation of essential components of tsetse diets, development of methods of incorporating these into the diets, identification of cheap alternative sources for compounding tsetse diets, and study of the effects of essential nutrients on ovarian growth, larval development and puparial viability.

The possibility was recognized of genetic changes occurring in laboratory colonies due to diets. Consequently, the group recommended that as soon as a satisfactory diet has been found for tsetse (or other species) a programme should be initiated to monitor and evaluate possible genetic changes in the insect that may have been induced by the diet.

A full report, with recommendations, is in preparation.

2. Future Meetings

The following Research Co-ordination Meetings are planned for 1986. The meetings are open to Co-ordinated Research Programme participants and invited guests/observers.

(a) Title: Development of Sexing Mechanisms in Fruit Flies through Manipulation of Radiation-Induced Conditional Lethals and Other Genetic Measures

Location: Crete, Greece

Dates: 16-21 September 1986

(b) Title: Development of Methodologies for the Application of the Sterile Insect Technique (SIT) for Tsetse Eradication or Control

Location: Vienna, Austria

Dates: 22-26 September 1986

C. Field Programmes

BICOT

The project, Biological Control of the Tsetse Fly by the Sterile Insect Technique (BICOT), was established to conduct the necessary research and development leading to the eradication of Glossina palpalis palpalis. The BICOT project area, covering 1,500 km² of agricultural land, is part of the Lafia Agricultural Development Project and contains approximately 450 linear km of riverine forest which constitutes the resting haunt and breeding site for the target tsetse species.

In a recent review of BICOT, the Project Advisory Committee (PAC) recorded excellent progress all round : large-scale rearing of G. p. palpalis has been accomplished using in vitro and in vivo feeding methods, so that the fly colony at the project headquarters in Vom now totals 120,000 females, giving at least 10,000 surplus males each week for sterilization and release. Additional flies are provided, as needed, from a 75,000 strong back-up colony maintained at Seibersdorf. Techniques have been developed for handling, sterilizing, packaging and transporting sterile male flies from the project headquarters to the target area approximately 200 km away. These techniques have been worked out jointly with staff of the FAO/IAEA tsetse laboratory in Seibersdorf near Vienna.

By the end of May 1986, more than 80% of the target area had been cleared of G. p. palpalis through a combination of trapping, use of insecticide-impregnated screens, and release of sterile males. Complete eradication from the entire area is expected by the end of 1986.

BICOT has been supported financially by Nigeria, Belgium, the Federal Republic of Germany, Italy, Sweden, the United Kingdom, IAEA and FAO.

The success achieved so far has encouraged the Nigerian government to request further assistance to develop a larger project (BICOT II) to include 12,000 km², primarily in Plateau State and partially in Benue State. The entire Lafia Agricultural Development Project area, including three cattle grazing preserves, is in the proposed tsetse eradication area. Included in BICOT II are plans for a training facility. If this becomes a reality, the training facility will serve as a major centre in West Africa for transferring the technology developed in the present project to scientists and technicians from other African countries.

D. What's on in Seibersdorf

Support for BICOT continued with maintenance of the back-up colony of Glossina palpalis palpalis at 75,000 producing females. Since January 1986, 388,000 puparia have been shipped to Nigeria where the excess males that emerge are sterilized and released in the test site. The females are used to supplement the breeding stock of the local colony. A member of the Seibersdorf laboratory staff served for 4 months at BICOT as an expert advising on the handling of puparia and assisting to set up a new microcomputer to handle the laboratory and field data. Imported supplies and equipment for BICOT were, as before, obtained and arranged for shipping by laboratory staff.

Experimental colonies of G. pallidipes, G. fuscipes and G. brevipalpis (all from Lambwe Valley origin) have been started at Seibersdorf. All colonies are being adapted to in vitro rearing and will be the source of materials for studying the laboratory aspects relating to the use of the SIT against the species.

Studies were continued to explore the use of trypanocides in the diet of tsetse flies fed in vitro for reducing the vectorial capacity of the flies. Samorin greatly reduced the rate of infection by Trypanosoma congolense amongst flies which, two weeks earlier when teneral, had been given an infested blood meal.

Investigations in the dietetics and nutrition of G. p. palpalis have revealed that:

1. dialysis of their diets greatly reduces their reproductive capacity;
2. irradiation up to 400 krad had no deleterious effect on the nutritional quality of fresh defibrinated blood;
3. chicken blood contains one or more factors which inhibit the tanning of puparia; and
4. the weight of puparia was positively related to the increased protein content of diets. Also, the nutritional value and relative extent of utilization of individual amino acids was determined.

Investigations were made to develop and improve mass-rearing systems for medflies. Cage netting with finer mesh (0.1 mm hole size) was found to stimulate greater oviposition, concentrations of some major ingredients in the finishing diet for rearing larvae were reduced without affecting production, and ingredients from Egypt were formulated into a satisfactory larval rearing medium.

Several million irradiated male pupae of a genetic sexing strain of medflies were produced at Seibersdorf and shipped to co-operators on the Italian island of Procida where the emergent adults were released. Effects of releasing sterile males only was compared with releasing sterile males and females to demonstrate the greater effectiveness of the former procedure for reducing fecundity in the wild female population. Experiments with packaging were done to optimize the procedures for shipping the medfly pupae.

Procedures were developed to quickly and effectively screen fermentation products of the bacterium Bacillus thuringiensis for lethal activity against adult medflies. The procedures were used to demonstrate that isolates from many varieties of the bacterium are active.

Experiments were continued to induce and isolate recessive genetic mutations that can be used to separate sexes of medflies. Characters were induced by radiation and isolated in strains that prevent recombination between an induced, potential genetic sexing allele and the morphological marker genes that are used to track the mutagenised chromosome. Of the several strains induced, the ones with the greatest viability were selected for further use.

Methods for using ethyl methanesulfonate to induce point mutations in medflies were developed. The treatment will be used to induce temperature-sensitive lethal factors in a breeding programme with the female recombination suppressor strain.

A.

1

JOHN G. RIEMANN

METABOLISM AND RADIATION RESEARCH LAB
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REPRODUCTIVE POTENTIAL AND OTHER ASPECTS OF THE
BIOLOGY OF THE SUNFLOWER MOTH, HOMOEOSOMA
ELECTELLUM (HULST) (LEPIDOPTERA: PYRALIDAE)

J. KANSAS ENTOMOL. SOC. 59: 32-36 (1986)

Over a two-week period, mated sunflower moths, Homoeosoma electellum (Hulst), of a laboratory colony deposited an average of 337 eggs per female. The reproductive potential of some females appeared to be substantially higher than the mean. Females from diapause larvae produced fewer eggs than those from nondiapausing larvae, while egg deposition by virgins was about one-fourth that of mated diapause females. The virgins produced fewer eggs in their ovaries and also retained mature eggs longer than mated females. All females did not have mature eggs at eclosion and some would not mate on the day following eclosion. At 27°C, egg deposition reached a peak at 3 days posteclosion, and some viable eggs continued to be deposited at the end of 13 subsequent days. Under laboratory conditions, with a 12:12 light-dark regimen, oviposition was initiated and peaked during the early part of the scotophase, while most mating occurred during the last 3 hours of the scotophase.

2

DEGRUGILLIER, M. E.

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IN VITRO RELEASE OF ACROSOMAL MATERIAL FROM
HOUSE FLY SPERM TREATED WITH MICROPYLAR CAP
SUBSTANCE

1985
Int. J. Insect Morphol. Embryol. 14: 381-391

Mature house fly, Musca domestica L. (Diptera: Muscidae), sperm were treated with female accessory gland secretion and micropyle cap substance to determine their effectiveness in eliciting an acrosome response. Scanning and transmission electron microscopy revealed that release of acrosomal material was achieved by treatment of the sperm with a combination of accessory gland secretion and micropyle cap substance but not by gland secretion alone. The gland secretion was utilized to dissolve the cap substance from mature ovarian eggs, and this combined solution was applied to sperm removed from the spermathecae of mated females. The acrosomes of several of the sperm were completely lacking, while others showed a partial effect of the treatment, i.e., extensive acrosomal membrane fragmentation and vesiculation. Most sperm that reacted to the treatment lacked a plasma membrane around the acrosome, suggesting that this membrane, along with the acrosomal membrane, is involved in formation of the vesicles alongside the acrosomal cavity.

A. E. HEACOX and R. A. LEOPOLD
 METABOLISM AND RADIATION RES. LAB
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 FARGO, NORTH DAKOTA 58105

OPTIMIZING CONDITIONS FOR CRYOPRESERVATION OF AN
 INSECT CELL LINE

Cryobiology 21, 435-442 (1984)

A cell line (UM-BGE-2) derived from embryos of the cockroach, Blattella germanica, was frozen to -196°C under a variety of conditions and cell viability was assayed after warming. It was found that cell viability was affected by the cooling rate, the warming rate, the controlled cooling end point temperature and the type and concentration of cryoprotectant. The best survival for cells suspended in Grace's tissue culture medium containing 1 M Me_2SO was obtained when cells were cooled at $1^{\circ}\text{C}/\text{min}$ to at least -90°C before being placed in liquid nitrogen and warmed at more than $900^{\circ}\text{C}/\text{min}$. Cultures initiated from these frozen cells produce typical growth curves and appear normal after several passages.

HEACOX, A. E., R. A. LEOPOLD, AND J. D. BRAMMER
 METABOLISM & RADIATION RESEARCH LABORATORY
 USDA/ARS
 FARGO, NORTH DAKOTA 58105

SURVIVAL OF HOUSEFLY EMBRYOS COOLED
 IN THE PRESENCE OF DIMETHYLSULFOXIDE

Cryo-Letters 6, 305-312 (1985)

Housefly embryos of various ages were cooled to -20° in the presence of dimethylsulfoxide as a cryoprotectant. Experiments using a variety of cooling and warming rates showed the best survival occurred in 7 to 10 hour old embryos cooled at 0.6 or $1.0^{\circ}\text{C}/\text{min}$. This broad age group represents embryos in the process of organogenesis. Factors affecting survival and some additional considerations are discussed.

R. A. LEOPOLD, E. P. MARKS, J. K. EATON, AND J. KNOPER
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ECDYSIAL FAILURES IN THE COTTON
BOLL WEEVIL: SYNERGISM OF
DIFLUBENZURON WITH JUVENILE
HORMONE

Pesticide Biochemistry and
Physiology 24, 267-283 (1985)

The frequency and etiology of ecdysial failures occurring during the pupal-adult transformation of the cotton boll weevil, Anthonomus grandis, was studied after treatment with juvenile hormone (JH) and/or diflubenzuron (DFB). Failure at emergence was the result of inability of the adult insects to escape from their pupal exuviae. Teratogenic effects in the form of pupal-adult intermediates or adults with pupal characters were not obtained with JH or DFB treatment after the larval-pupal ecdysis. Combining JH with the DFB treatment yielded a synergistic response that increased the frequency of ecdysial failures about 7-fold when applied in the later pupal stages. The fine structure of the abdominal cuticle of adults experiencing ecdysial failure after treatment with JH as white-eye pupae exhibited granular deposits within the lamellar region and interference with deposition of the non-lamellate endocuticle. DFB treatment of the earlier pupal stages interfered with deposition of the lamellate cuticle by eliminating or reducing the lamellar structure. It was concluded that the primary interaction of JH with DFB was that of extending or reinitiating DFB-sensitivity in the later pharate adult. A secondary interaction may involve inhibition of cuticle hardening as both JH and DFB appear to inhibit the tanning process of adults treated as white-eyed pupae.

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The reproductive capacity of moths of
Spodoptera littoralis fed with ascorbic
acid analogs.

Entomol. exp. appl. 41, in press

D-ascorbic acid, L-dehydroascorbic acid, 6-chloro-6-deoxy-L-ascorbic acid, 6-bromo-6-deoxy-L-ascorbic acid and ascorbyl-2-sulfate were bioassayed per os on moths of Spodoptera littoralis (Boisd.). The activity of each of the analogs was compared with that of vitamin C and sucrose controls. D-ascorbic acid caused males to produce malformed spermatophores with no reduction in mating capacity. Females mated to these males laid infertile eggs and fecundity was lower than in females fed other analogs, L-ascorbic acid or sucrose diets. 6-chloro-6-deoxy-L-ascorbic acid reduced fertility and fecundity of the females, although the analog did not affect the spermatophores. D-isoascorbic acid (=D-araboascorbic acid; D-erythorbic acid), D-glucoascorbic acid and all the other analogs were equally acquired by males microfed singly or at mating. The effect of D-optically active analogs on the spermatophore, and the applications of the analogs in pest control, are discussed.

B.

7.

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EFFECT OF BETA RADIATION ON
CORCYRA CEPHALONICA STANTON

Fifth instar larvae of *Corcyra cephalonica* Stainton were allowed to feed on crushed jawar treated with radioactive phosphorus and separated in to four groups according to the activity range (A; upto 2500; B: 2501-5000; C: 5001-7500 and D: 5001-10,000 counts per minute) to see the effect of beta radiation on further development of this insect pest. Studies revealed that there was no appreciable difference in per cent pupation and adult emergence between control and the highest activity range (5001-10,000 cpm). However, number of eggs laid per pair in the subsequent generation was significantly reduced at this level of activity.

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Sperm Transfer of Gamma-Irradiated Male
Fruit Fly, Dacus zonatus (Saunders)

Sperm transfer of male fruit flies, Dacus zonatus (Saunders) emerged from 8-day-old pupae irradiated at 80 Gray were determined from amount of sperm in female spermatheca. The results showed that the sperms of 10, 15 and 30-day-old sterilized males were not significant difference from normal males ($P > 0.05$). However, the sperms of 20 and 25-day-old sterilized males were less numerous than these normal males ($P < 0.05$).

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Quarantine Treatment on Fruit Fly,
Dacus zonatus (Saunders) in Fruits by
Gamma Irradiation.

The fruit fly, Dacus zonatus (Saunders) is a major pest of fruits in Thailand. The problem of ethylene dibromide (EDB) fumigant ban for disinfection of fruit flies in fresh fruits and injury to many fruits by cold storage have led to interest in the use of gamma irradiation for disinfecting export commodities. The purposes of this experiment are to determine the optimum dose of radiation for disinfection of fruit fly, D. zonatus eggs and larvae in Hom Tong banana, Nong Klangwan mango and papaya. The results of this experiment are as follows:- 1) The LD_{50} for 20-hour-old eggs in banana, mango and papaya at 3 days after irradiation were 0.17, 0.15 and 0.14 kGy respectively. The dose of 0.3 kGy caused 100% mortality of eggs and larvae hatching from eggs irradiated at 0.15 and 0.05 kGy were unable to pupate and emerge as adults respectively, 2) The LD_{50} for 1-2-day-old larvae in banana, mango and papaya at 3 days after irradiation were 0.64, 0.47 and 0.49 kGy respectively. The dose of 0.6 and 0.3 kGy were required to prevent pupation and emergence respectively, 3) The LD_{50} for 3-4-day-old larvae in banana, mango and papaya at 3 days after irradiation were 1.0, 1.0 and 0.77 kGy respectively. The dose of 1.2 and 0.3 kGy were required to prevent pupation and emergence respectively.

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Radiation sterilization and
cytoplasmic incompatibility
in a "tropicalized" strain
of the Culex pipiens
complex.

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In the Culex pipiens group of mosquitoes cytoplasmic incompatibility provides a "ready-made" source of sterile males for use in a SIT programme, but the inadvertent release of a few fertile females with the males could lead to replacement of one pest population by another, rather than pest suppression. This risk might be avoided by irradiation of pupae with a sufficient dose to ensure female sterility. This idea was studied with a synthetic "tropicalized" stock with cytoplasm of London origin but genome of Dar es Salaam origin, which had been introduced by backcrossing after breaking down the incompatibility barrier by tetracycline treatment of Dar es Salaam males. It was found that females of this synthetic strain could be rendered totally sterile by irradiation with 5 k rad on the second day of pupal life. Because of their cytoplasm of London origin, males of the synthetic strain were fully incompatible with Dar es Salaam females. Males of the synthetic strain irradiated with 5 k rad competed well in cages with Dar es Salaam males for Dar es Salaam females. By contrast, if Dar es Salaam males were irradiated at the pupal stage with 10 k rad to produce conventional radiation induced sterility, they competed poorly.

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LOPES.

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INFLUENCE OF TWO *Phaseolus vulgaris*
CULTIVARS ON THE DETERMINATION OF THE GAMMA
RADIATION STERILIZING DOSE IN IMAGOS OF
Acanthoscelides obtectus (SAY, 1831) (COL.,
BRUCHIDAE).

S U M M A R Y

The objective of the present work was to determine, using the weighing method, the ^{60}Co gamma radiation dose to sterilize imagos of *Acanthoscelides obtectus* (Say) in two *Phaseolus vulgaris* cultivars, Rosinha and Jalo. Insects from normal laboratory rearing were irradiated at 0 (control), 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150 and 200 Gy at a rate of 3.03 kGy/h. Each treatment consisted of five replications with 20 insects each. Glass flasks containing 100 g bean each were used in each replication. After irradiation the flasks were placed in a temperature controlled chamber, $27 \pm 2^\circ\text{C}$, and $70 \pm 5\%$ relative humidity, the weighing being done weekly.

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DETERMINATION OF STERILIZING DOSES OF GAMMA
RADIATION OF IMAGOS OF *Callosobruchus analis*
(FABR., 1755) AND *C. maculatus* (FABR., 1792)
IN *Cicer arietinum* CV. "GRÃO DE BICO" AND
Vigna sinensis CV. "SERIDÔ".

S U M M A R Y

The objective of the present work was to determine, using the weighing method, the sterilizing dose of ^{60}Co gamma radiation for imagos of *C. maculatus* (F.) and *C. analis* (F.) in seeds of "Grão de Bico" and "Seridô". To carry out this experiment imagos from normal laboratory rearing were irradiated at 0 (control), 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150 and 200 Gy at a rate of 3.03 kGy/h. Each dose consisted of five replications with 20 insects each. Glass flasks with approximately 100 g substrate were used as replications. After irradiation the flasks were placed in a temperature controlled chamber, $27^{\circ} \pm 2^{\circ}\text{C}$, with relative humidity $70 \pm 5\%$. Weighings were made weekly. It was concluded that the dose necessary to induce sterility in both species is 50 Gy.

C

13

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Population Fluctuation Studies of Oriental
Fruit Fly (*Dacus dorsalis* Hendel) and *Dacus*
zonatus (Saunders) and Microclimates at Royal
Any Khang Highland Station, Chiang Mai.

Studies on the population fluctuation of male fruit flies, *Dacus dorsalis* Hendel; and *Dacus zonatus* (Saunders) at Royal Ang Khang Highland Research Station Chiang Mai in 1983 were conducted. Population of both species started to increase in early of February. Population of *Dacus zonatus* increased rapidly and reached the peak, 103 males/trap/day, in the middle of May. In contrast, the population of *Dacus dorsalis* increased slowly following the population of *Dacus zonatus* until the beginning of June, after that, the population increased rapidly and reached the peak, 240 males/trap/day, in the middle of July. Later on they dropped down rapidly in the middle of August, then the population of the two species fluctuated together. Finally, the population declined near zero in November, December and next January. The population density of the two species adult flies were rather high from the end of winter to middle raining season and this period coincided with the time of fruit being developed in many introduced varieties (peach, plum, apple, pear and persimon).

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ECOLOGY OF GLOSSINA FUSCIPES FUSCIPES
(NEWSTEAD) IN RELATION TO THE
PREVALENCE OF BOVINE TRYPANOSOMIASIS

ABSTRACT

A study into the ecology of G.f. fuscipes was carried out in Kityerera subcounty, a small area with no observed epidemic but within the epidemic counties of Busoga. Glossina fuscipes fuscipes is the only tsetse species found in the area. 318 flies were caught during a dry period of January - February, 1984. No infection was found in the 224 flies dissected. 40 flies were crashed and their supernatant inoculated into laboratory mice but no parasitaemia was noticed after 25 days. Another 197 flies were caught during the beginning of the wet season (May/June). 116 flies were dissected and only one was found with gut-infection of trypanosomes giving an infection rate of 0.8%. Inoculation of these trypanosomes into a mouse did not give any parasitemia after 30 days. The infection rates reported for cattle in the same area is 14.4%.

The population of G.f.fuscipes caught by the bioconical traps declined steadily as the trapping over the study period continued. The ratio of male to female flies was 1:1 during the dry period and 2:1 in the wet period. These ratios did not change with subsequent trapping days in each season.

BEREGOVY, VLADIMIR

APPEARANCE OF FIRST GENERATION LARVAE OF THE SUNFLOWER MOTH
HOMOEOSOMA ELECTELLUM (HULST), (LEPIDOPTERA: PYRALIDAE) IN
THE CENTRAL UNITED STATES.METABOLISM AND RADIATION
RESEARCH LAB/USDA/ARS

FARGO, NORTH DAKOTA 58105

Jr. Kansas Entomol. Soc. 58:739-742. (1985)

The appearance of the first generation of larvae of Homoeosoma electellum (Hulst), was followed from May 6, 1983, in Texas, northward until August 9, 1983, in Nebraska and South Dakota. First generation sunflower moths utilized at least five host plant species in central United States: Gaillardia pulchella Fougereux, G. aestivalis (Walter), Coreopsis tinctoria Nuttall, C. linifolia Nuttall, and Helianthus annuus L. G. pulchella, C. tinctoria, and H. annuus were the most commonly used host plant species.

JOHN G. RIEMANN, V. BEREGOVY, and RITA L. RUUD

PREPUPAL WANDERING BEHAVIOR OF THE
SUNFLOWER MOTH (LEPIDOPTERA: PYRALIDAE)

METABOLISM AND RADIATION RESEARCH LABORATORY

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Ann. Entom. Soc. Am. 79:116-120 (1986)

In a 12:12 (L:D) photoperiod regimen, the sunflower moth, Homoeosoma electellum (Hulst), exhibited a well-defined prepupal wandering rhythm. Wandering occurred during the day; the timing depended somewhat on day length and temperature. Phase shifting mostly followed onset of darkness. Wandering starts were temperature compensated, but at lower temperatures larvae started to wander throughout the photophase. In constant darkness the wandering rhythm persisted with fairly normal timing for the first 2 days, but the numbers wandering dropped on the second day and, thereafter, wandering was essentially random in relation to time. Timing of gut purging and initiation of wandering were closely correlated. Larvae starting to wander were positively phototrophic. When larvae were ligated shortly after they started to wander, significantly more individuals pupated if they had been maintained in continuous darkness than if they had been kept in a light-dark regimen. Wandering during the middle portion of the day may have originated as a defense against predation.

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An Integrated Control Program for the
Mango Fruit Fly in the Philippines

ABSTRACT

This paper discusses the integrated mango fruit fly control program being planned in the Philippines based on the techniques developed and data collected for a period of 12 years. The methods used were:

1. Sex pheromone and naled baiting at 94:6 (v/v).
2. Sterile insect release, both male and female fruit flies were sterilized at 5 krad (0.05 kGy) and released in the field.
3. Mango disinfection by gamma radiation at 50 krad (0.5 kGy) for quarantine treatment.

In the course of the experiment, a cheap diet for mass rearing was developed from locally-available materials. Based on this diet a million pupae can be produced at US\$22 including labor.

The paper also described an insect trap developed from used plastic oil cans. This fly trap was found effective, cheap and easy to construct. Aside from lowering male fly population, when the field was saturated with fly traps, monitoring of peaks of fly populations in any given period of time was made possible. Field research also showed that fly populations are high even if mango fruits are not in production, indicating presence of other host plants of Dacus dorsalis.

Furthermore, the use of gamma radiation for disinfection of mangoes for export can be used as an alternative to ethylene dibromide for quarantine treatment of fruits.

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Screening of *Bacillus thuringiensis*
Berliner preparations for microbial control
of *Ostrinia nubilalis* Hubner in sweet corn

Phytoparasitica, 14 No. 2, (1986) in press

Bacillus thuringiensis (B.t.) preparations were bioassayed against the European
corn borer *Ostrinia nubilalis* Hubner larvae to determine their potency. B.t. serovar
3a3b *Kurstaki* (Dipel) was more potent than B.t. serovar 1, *thuringiensis*
(Bactospeine I) and 3a3b *kurstaki* (Toarrow CT), in which the spores are inactive
or serovar 7, *aizawai* (ABG 6104).

Another preparation of 3a3b *kurstaki* (Bactospeine III) at a concentration of 0.5
and 1% sprayed on sweet corn leaves of potted seedlings and plants in the field was
active against artificially-fed larva in the laboratory. The pH of the corn leaf
surface ranged 6.9 - 7.5, before and after application.

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ANTIBACTERIAL ACTIVITY DEMONSTRATED BY CULTURE
FILTRATES OF *PROTEUS MIRABILIS* ISOLATED FROM
SCREWORM (*COCHLIOMYIA HOMINIVORAX*) (DIPTERA:
CALLIPHORIDAE) LARVAE

J. Med. Entomol. 21:159-164. (1984)

Several strains of *Proteus mirabilis* isolated from screwworm (*Cochliomyia hominivorax*)
(Diptera: Calliphoridae) larvae were found to produce a bactericidal substance
(mirabilicide). The mirabilicide was extracted from culture filtrates at pH 2.9 with
chloroform. Both Gram-negative and Gram-positive bacteria were susceptible to mirabili-
cide. A coagulase-positive strain of *Staphylococcus aureus* (boil isolate) was killed in 4
min; however, a strain of *Providencia rettgeri* (formerly in the genus *Proteus*), isolated
from screwworm larvae, showed significant resistance to the mirabilicide. Ultrastructural
and morphological changes in *P. mirabilis* cells exposed to mirabilicide were limited to
cytoplasmic changes, while *Salmonella typhimurium* cells showed both cytoplasmic changes
and cell wall modifications, although the cells did not lyse. Mirabilicide may play a
significant role in the establishment of the nonpurulent, larval-infested wound.

SYED S.H. QADRI, M.K.J. SIDDIQUI, F. ANJUM and M.F. RAHMAN *In vitro* and *in vivo* evaluation of structure-toxicity relationship of newly

TOXICOLOGY UNIT, REGIONAL RESEARCH LAB- synthesised biphenyl derivatives of ORATORY, HYDERABAD-500 007, A.P., INDIA. carbamic and phosphoric esters against *Apis cerena indica*" M.M. SIDKY and F.H. OSMAN

PESTICIDE CHEMISTRY LABORATORY, NATIONAL RESEARCH CENTRE, DOKKI, CAIRO, EGYPT. Tropical Pest Management, Volume 32 (1986): In Press.

Abstract

The acute toxicity (LD_{50}) and anti-cholinesterase property of some newly synthesised derivatives of carbamic and phosphoric esters were determined against honey bee (*A. c. indica*). The LD_{50} for p-(4-nitrophenyl) phenyl-N-methylcarbamate (69.18 ± 0.059 ug/gm body weight) was significantly lower than p-phenyl-phenyl-N-methylcarbamate (173.78 ± 0.056) while a much higher value was found for p-(4-nitrophenyl) phenyl-N-ethylcarbamate (1023.29 ± 0.044).

The I_{50} values for nitromethyl and nitroethyl derivatives against honey bee head acetylcholinesterase (AChE) were found to be closer to N-methyl biphenyl carbamic ester. The LD_{50} values for biphenyl derivatives of methyl and ethyl parathion and paraoxon. Their I_{50} values were less than ethyl parathion and more than paraoxon against honey bee head AChE when tested in vitro. Based on LD_{50} and I_{50} values of these new analogues it can be concluded here that nitromethyl derivative was more potent than N-methyl biphenyl carbamic ester and two derivatives of paraoxon be structurally modified in order to prolong their biological half-life to avoid rapid detoxification and or to enhance the absorption.

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PARASITISM OF THE SUNFLOWER MOTH, *HOMOEOSOMA ELECTELLUM* (HULST) (LEPIDOPTERA: PYRALIDAE) IN THE CENTRAL UNITED STATES

J. Kansas Entomol. Soc. 58: 739-742 (1985)

Hymenopteran and Dipteran parasitoids, representing seventeen species in six families were reared from larvae of the sunflower moth, *Homoeosoma electellum* (Hulst) (Lepidoptera: Pyralidae) in 1982 and 1983. Parasitoids found and identified included: Braconidae, *Bracon mellitor* Say, *Chelonus altitudinus* Viereck, *Dolichogenidea homoeosomae* (Muesebeck), *Agathis buttricki* (Viereck) and *Macrocentrus ancyliivorus* Rohwer; Eulophidae, *Elasmus setioscutellatus* (Crawford); Bethyridae, *Goniozus floridanus* (Ashmead); Perilampidae, *Perilampus* sp.; Ichneumonidae, *Pristomerus austrinus* Townes & Townes, *Pristomerus* sp., *Diadegma* sp., *Temelucha* sp.; Tachinidae, *Clausicella opaca* (Coquillett), *Erynnia tortricis* (Coquillett), *Leskiomima tenera* (Wiedemann), *Pseudachaeta* sp., and one species of Blondeliini. *E. setioscutellatus*, *G. floridanus*, *Pseudachaeta* sp., and Blondeliini sp. have not been previously identified as parasitoids of the sunflower moth.

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Isolation of tsetse fly salivary gland
antigens by affinity chromatography on
purified IgG from exposed rabbits

Summary. As far as the control of hematophagous insects is concerned, the induced response of hosts causing either reduced fecundity and sterility or increased mortality represents a potential control method. Permanent immunological screening of laboratory animals is required in order to maintain the reproductive potential of the tsetse laboratory colonies at optimal productivity. However, the exact nature of tsetse salivary gland antigens (cf. Parker 1979 or Kaaya and Alemu 1984) and a simple method for their standard isolation have not been published yet. Moreover, further study of biological effects of host immunity on fly mortality and fecundity is not possible without purification and identification of salivary gland antigens.

Repeated feeding of tsetse flies (Glossina palpalis palpalis from Biotechnology Unit of Joint FAO/IAEA Division, Seibersdorf) on the same rabbit is accompanied by production of very high titres of specific antibodies against the saliva of this fly. The presence and titre of these antibodies are detected by counter current immunoelectrophoresis. Antibodies were precipitated by $(\text{NH}_4)_2\text{SO}_4$ and purified on DEAE-Sephacel. Antigens from tsetse salivary gland homogenates were isolated on purified antibodies coupled to CNBr-activated Sepharose 4B. Analysis of protein fractions separated by affinity chromatography by means of SDS electrophoresis provided evidence of two major fractions with molecular weights of 160 and 200 kilodaltons.

In view of the need for pure antigens, the method of affinity chromatography seem to be most profitable procedure mainly owing to minimal losses and high purity of isolated fractions. Application of these procedures enables us to isolate pure tsetse salivary gland antigens for the first time. Based on the comparison of results of affinity chromatography on Con A Sepharose 4B the isolated antigens are likely to be macromolecules of glycoprotein character. The exact chemical characteristics as well as their biological role are worthy of further study.

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A MEASURING GRID FOR THE ANALYSIS
OF ISOELECTRICALLY FOCUSED PROTEINS
IN POLYACRYLAMIDE GEL SLABS

Electrophoresis 6: 206-209 (1985)

An overlay grid was constructed to facilitate the measurement of densitometric scans of proteins in pH gradients formed by electrofocusing. The overlay procedure provides a rapid way for data collection. The relationship between pH and gel distance closely fit a straight line, ($R^2 > 0.99$). The reliability of the system was shown by the close agreement of the linear regressions from six different sets of data. Also, the feasibility of using a similar grid for a nonlinear system was established.

