INFORMATION CIRCULAR

ON

RADIATION TECHNIQUES AND THEIR

APPLICATION TO INSECT PESTS

No. 31

December 1982
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## 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The summaries of unpublished work often represent preliminary reports of investigations in progress and, therefore, such findings are subject to possible revision at a later date. The contents of this Information Circular should not be published or referred to in articles for publication without obtaining permission from the authors first.
A. Publication Policy of Information Circular

The policy of the Joint FAO/IAEA Division in publishing this Information Circular is to emphasize the results of recent research on the use of radiation and radioisotopes in entomology. Therefore, emphasis is placed on unpublished data. Please bear in mind that we cannot edit your contributions and that these are reproduced by a photographic process. Therefore, their appearance and content in the Circular will faithfully reflect your own care. Some of you have sent in reprints of published papers or long articles as contributions to the Information Circular. We include summaries of recently published data only. The length should be no more than one typewritten page when double-spaced (a form for this purpose is included in this copy of the Information Circular).

B. Professional Staff

Headquarters
D.A. Lindquist  Head, Insect and Pest Control Section
E.D. Offori  Insect and Pest Control Section

BICOT (Biological Control of Tsetse by the SIT) - PO Box 76, Vom, Plateau State, Nigeria
R. Sarmiento  Project Director
M. Oladunmade  Project Co-Director
H-J. Hamann  Supervisor of Laboratory and Rearing Operations
T. Tenabe  Co-Supervisor of Laboratory and Rearing Operations
(not filled)  Supervisor of Field Operations
(not filled)  Co-Supervisor of Field Operations

Seibersdorf Laboratory
R.E. Gingrich  Head, Entomology Laboratory
D.J. Nadel  Mediterranean Fruit Fly Investigations (retired 1 March 1982)
U. Feldmann  Tsetse Fly Investigations (FAO Associate Expert)
A. Van der Vloedt  Tsetse Fly Investigations
E. Bush-Peterson  Research Entomologist
G. Kapatsa  Tsetse Fly Investigations (reported 18 December 1981)
P. Kaiser  Genetic Sexing Mechanisms
C. **Entomology Laboratory**

The IAEA has an international laboratory located at Seibersdorf, Austria, about 30 km. from Vienna. A part of this laboratory is devoted to the use of atomic energy in entomology.

The primary research objective of the entomological programme at the Agency's Seibersdorf Laboratory is to support and service the Joint FAO/IAEA Division's programme on insect control. This involves primarily the development of the Sterile Insect Technique (SIT) as a means of insect control or eradication. Because of the dependence of this technique on efficient, economical mass rearing of insects, much of the research at the laboratory involves rearing. Other major research areas include (1) methods of radiation sterilization for producing the best possible sterile insect (in terms of sexual competitiveness, longevity and quality), (2) handling techniques for large numbers of insects, and (3) field programme direction and/or supplying insects for field programmes. The laboratory's programmes are associated with existing field programmes and much of the research is concerned with the field problems that arise.

The general areas of research presently being pursued are:

1. Develop and improve mass rearing;
2. Improve radiation techniques;
3. Develop methodology for "fail-safe" radiation sterilization;
4. Develop laboratory 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 our laboratory:

1. Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann);
2. Tsetse fly, *Glossina palpalis palpalis* (Robineau-Desvoidy);
1. Medfly programme

(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 (100s 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 mechanisms.

2. Tsetse fly programme

(a) Improve rearing technology with reduced handling of flies.
(b) Develop in vitro and in vivo feeding technology for mass rearing methods.
(c) Develop methods for preserving blood (freeze-drying).
(d) Use of blood additives for improving tsetse fly 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.

3. Isotopes and Radiation in Integrated Pest Management programme

(a) Predator-prey and host-parasite relationships.
(b) Mass rearing.
(c) Selectivity of insecticides.
(d) Alternate hosts.
(e) Adult population estimates for forecasting.
(f) Pest dispersal.
(g) Training.
D. **Fellowships Awarded in Radiation Entomology (1980-82)**

Subject to quarantine regulations, availability of funds, etc., the laboratory can serve entomologists in developing countries planning or carrying out sterile insect projects. The laboratory also 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.

Fellowships may be awarded for a period of several months to a maximum of twelve months. In certain exceptional cases, extensions of up to twelve additional months may be granted. Fellowships can be awarded as part of a comprehensive project or on an individual basis as a direct contribution to projects in the country's atomic energy programme, and provide opportunities for training there.

Applications for fellowships must be made to the Agency exclusively through official channels, and priority is given to requests associated with projects of direct benefit to individual Member States.

<table>
<thead>
<tr>
<th>Country and Name of Fellow</th>
<th>Host Country</th>
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</thead>
<tbody>
<tr>
<td><strong>BANGLADESH</strong></td>
<td></td>
</tr>
<tr>
<td>Muhammad Husain</td>
<td>USA</td>
</tr>
<tr>
<td>Manjur A. Chowdhury</td>
<td>USA, Austria</td>
</tr>
<tr>
<td>A.K.M. Qudrat-e-Khuda</td>
<td>USA, Austria</td>
</tr>
<tr>
<td><strong>EGYPT</strong></td>
<td></td>
</tr>
<tr>
<td>Soher M. Riad Souka</td>
<td>UK</td>
</tr>
<tr>
<td>A. El-Zooka</td>
<td>Austria</td>
</tr>
<tr>
<td>T. El-Abassi</td>
<td>Austria</td>
</tr>
<tr>
<td><strong>GHANA</strong></td>
<td></td>
</tr>
<tr>
<td>Abdullah Essaka</td>
<td>Netherlands</td>
</tr>
<tr>
<td>Rajainder Kumar</td>
<td>USA</td>
</tr>
<tr>
<td>Henry Meier</td>
<td>USA</td>
</tr>
<tr>
<td>Kojo Cyapia Montford</td>
<td>USA</td>
</tr>
<tr>
<td>Jackson K. Akosmoah</td>
<td>Austria</td>
</tr>
<tr>
<td><strong>INDIA</strong></td>
<td></td>
</tr>
<tr>
<td>Amara Sambasiva Rao</td>
<td>USA</td>
</tr>
<tr>
<td>Obondiba H. Rananavare</td>
<td>USA</td>
</tr>
<tr>
<td><strong>KENYA</strong></td>
<td></td>
</tr>
<tr>
<td>Philip A. Onyango</td>
<td>Austria</td>
</tr>
<tr>
<td><strong>MEXICO</strong></td>
<td></td>
</tr>
<tr>
<td>Cecilia Garcia Viesca</td>
<td>Spain</td>
</tr>
<tr>
<td>J.L. Zavaia Lopez</td>
<td>Austria</td>
</tr>
<tr>
<td>D. Orozco</td>
<td>England</td>
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<td>P. Liedo</td>
<td>England</td>
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</table>

Scientific Visit
NIGERIA
Codis U. Okengwu
Emmanuel Ofodile
Timothy Tanko
Stephen O. Tenabe
Moses Oladummade

USA
Austria
Austria
Austria
Austria

PAKISTAN
Sana Ullah Khan Khattak

USA

SPAIN
M.E. Riva Francois

Netherlands

SRI LANKA
B.H. Rohitha

India

SUDAN
O.A.S. Mohammed
N. Sharaf El Din

USA
USA

THAILAND
Ratana Poramarcom
Pravat Kachoung

USA
USA, Austria

ZAMBIA
Blackwell Kafwimbiri
M. G. Bangwa
Geoffrey M. Kapatsa
S.M. Moobola
K.H. Chisanga

Austria, Nigeria
Austria
Austria
Austria
Austria
E. Technical Assistance Assignments, July - December 1982

<table>
<thead>
<tr>
<th>Name</th>
<th>Nationality</th>
<th>Location of Assignment</th>
<th>Major Responsibility</th>
</tr>
</thead>
<tbody>
<tr>
<td>R. Gingrich</td>
<td>USA</td>
<td>Mexico</td>
<td>To review and discuss quarantine programme against the Medfly in Mexico.</td>
</tr>
<tr>
<td>D.A. Lindquist</td>
<td>USA</td>
<td>Egypt</td>
<td>Prepare economic analysis of Medfly programme in Egypt.</td>
</tr>
<tr>
<td>G.C. Labrecque</td>
<td>USA</td>
<td>Czechoslovakia</td>
<td>Review of Training Film on &quot;use of SIT as applied to the Mediterranean fruit fly.</td>
</tr>
<tr>
<td>G.C. Labrecque</td>
<td>USA</td>
<td>Mexico/USA</td>
<td>To attend Regional Training Course on the Use of the Sterile Insect Technique for the Control of Fruit Flies and to meet with taxonomists to discuss Rhynchophorus ferrugineus problems in Sri Lanka.</td>
</tr>
<tr>
<td>D.A. Lindquist</td>
<td>USA</td>
<td>Egypt</td>
<td>Complete negotiations on the Agreement between the Agency and the Egyptian Government.</td>
</tr>
<tr>
<td>E.D. Offori</td>
<td>Ghana</td>
<td>Indonesia</td>
<td>To review UNDP/IAEA project INS/78/074 and to prepare a detailed plan of operations for the insect control component of the project.</td>
</tr>
</tbody>
</table>

F. Recent Publications by the Joint FAO/IAEA Division

Symposia


Technical Reports Series

S. Monica Asman, P.T. McDonald, W.K. Reisen  
M. M. Milby and W. C. Reeves  
Department of Entomology and Department of  
Biomedical and Environmental Health Sciences,  
University of California, Berkeley, CA. 94720  

A FIELD RELEASE OF RADIO-SterILIZED  
MALES TO SUPPRESS AN ISOLATED POPULATION  
OF Culex tarsalis

Two exploratory field release programs (1979 and 1980) using radiosterilized males  
from field-derived pupae indicated that released males were equally competitive with  
native unirradiated males for field females.

For the 1981 field trial a new colony of Culex tarsalis was established from the  
release site population during the late autumn of 1980. In the early spring of 1981  
the colony was mass-reared, males were radiosterilized and released into a native pop-
ulation at the onset of field emergence of this species in Kern County, California.  
Emphasis was placed on attaining a favorable (10 to 1 or higher) sterile to field male  
ratio. In total 84,652 sterilized males were released. A simulated parallel study  
was conducted for monitoring purposes in a large quonset-hut outdoor cage.

The released radio-sterilized males proved to be uncompetitive for field females  
against field females.

In summary, males from the laboratory colony initiated from the release site less  
than 6 months earlier were uncompetitive against field males. Radio-sterilization was  
not considered detrimental to any noticeable extent, since sterilized males derived from  
field-collected pupae were fully competitive under similar field conditions in 1980.  
The loss of competitiveness could not be attributed to loss of fitness, since the sur-
vivorship estimated for the released males in cages and in the field by mark-release-
recapture methods were the same as that of the males from the target population.

The loss of competitiveness was attributed to assortative mating which in turn was  
initiated with laboratory rearing and the process of colonization. The 2 types of females  
involved, field and laboratory-reared, mated more frequently with males of their own  
genotype. The preferential mating began with the initial rearing in the insectary, since  
the mating behavior data of female offspring from field-collected females were inter-
mediate between that of field and colonized females. Thus, females behave differently  
from their native parents after only 1 generation of insectary rearing.

In light of these conclusions the current research is oriented to the identification  
of laboratory environmental factors which cause the changes—genetic or conditional, and to  
eliminate or minimize them. Since selection changes are manifestations of more funda-
mental biochemical alterations, electrophoretic analysis will be used as one method of  
monitoring colonies stemming from field stock under various environmental regimes through  
subsequent generations in the laboratory. Additional types of monitoring will deal with  
measuring flight capability and mating competitiveness of laboratory males reared under  
various regimes.
The control or eradication of various insect species by the sterile-insect release technique requires the certain dose for sterilizing without effects on mating performance. This preliminary study was conducted to reveal some radiation effects of a certain sterilizing dose on mating propensity and competitiveness of flies.

Mediterranean fruit flies, *Ceratitis capitata* (Wiedeman) were irradiated with 14.5 krad of gamma radiation from a cobalt-60 source in a nitrogen atmosphere 2 days before eclosion. The procedures of tests were established by Boller in his International Course on Quality Control in *C. capitata* in 1979 Castellon, Spain.

The test of the mating propensity was conducted by using two plexiglas cages containing separately 25 pairs of irradiated and non-irradiated flies in each cage. The results indicated that there was no radiation effect on the mating propensity of 3-5-day-old flies (P = 0.05) except of 6-day-old flies. The mating propensity increased with age of both non-irradiated and irradiated flies.

In the test measured the mating performance of two strains, non-irradiated and irradiated flies, 20 pairs each in a competitive situation revealed that non-irradiated 3-6-day-old flies of both sexes had higher mating competitiveness than irradiated flies (P = 0.01) except at 4 days old. However, in this competitive situation, flies showed no difference in mating combination between the same and two different strains (P = 0.05).

Although this sterilizing dose at 14.5 krad affected the mating competitiveness of irradiated flies, there were no evident effects on mating propensity and mating combination.
ABSTRACT

The concept of chemosterilant technique lies in heavily outnumbering the natural pest population with release of sterile males so that the majority of matings with native females of the pest will be fruitless. Thus the progeny is controlled and ultimately the pest population reaches a zero-point. Often a dosage might produce sterility but the effect might be transitory and the insects might regain fertility in due course of time.

An attempt has been made in this study to know whether the chemosterilant-treated cotton bug *D. cingulatus* F. shows any recovery from the sterilant action.

It was evident from the study that a smaller dose can induce partial recovery from the sterilant action in both sexes. The gonial cells are quite sensitive to chemosterilants and doses that produce high frequencies of dominant lethal mutation in the mature germ cells are adequate to kill all the gonial cells. The partial recovery although observed in both sexes, was not of the same degree. The recovery in males were more rapid as compared to that in females.
ABSTRACT

Penfluron (A13-63223) a chitin synthesis inhibitor caused histopathological changes on Leptocoris coimbartensis (Hemiptera-pyrhocoridae). Topical application of 1% Penfluron induced 99% sterility in both the sexes, but gradual recovery of fertility was observed in males. Females failed to recover the fertility in their lifetime. Injection of higher doses induced gradual degeneration and resorption of oocytes in females. By the 7th day of treatment only a transparent tissue remained. At all concentration the mortality and mating competitiveness were unaffected.

EFFECT OF GAMMA IRRADIATION ON EGGS AND PUPAE OF THE FRUIT FLIES, Dacus zonatus (Saunders)

Effect of gamma irradiation on mortality of eggs and pupae of Dacus zonatus (Saunders) were investigated. The 20 ± 1-hour-old eggs and 1,3,6 and 8-day-old pupae were irradiated with gamma rays from Cobalt-60 at 0-400 and 0-1,000 Gray respectively. The results of this experiment are as follows:

(1) The LD_{50} and LD_{95} for 20 ± 1-hour-old eggs after 2 days were 169.3 and 303.2 Gray respectively. (2) The LD_{50} and LD_{95} for 1,3,6 and 8-day-old pupae after 10,8,5 and 3 days were 9.5, 19.50, 231.4, 470.7 and 15.6, 72.6, 409.2, 819.5 Gray respectively.
The experiment on insect disinfection in smoked fish by gamma irradiation was done by irradiating 2-day-old eggs, 28-day-old larvae, 5-day-old pupae, and 5-day-old adults, reared at 26 ± 1°C and 75-70% RH., with doses of 0.5, 0-20, 0-20, 0-20 Krad respectively for sterility dose and with doses of 0-2, 0-6, 0-60, 20-60 Krad respectively for LD_{50}. The results obtained from this experiment are as follows: (1) For the study of all stages of this insect sterility dose, there was no irradiated egg hatch in all doses and only irradiated larvae with 1-5 Krad reached pupal stage but emerged as malformed adults and died. Furthermore, the sterility doses for pupal and adult stage are both at 7.5 Krad. However the percent of egg hatch from 5 Krad pupae and 5 Krad adults are .18 and .4 respectively. (2) The LD_{50} checked at different times after irradiation in each stage for 2-day-old eggs after 2 days, for 28-day-old larvae after 7 days, 5-day-old pupae after 5 days, and for 5-day-old adults after 5 days are .48, 12.85, 18.5 and 29.78 Krad respectively.

**Summary**

Sterility was induced in the males of *Dysdercus cingulatus* F. (red cotton bug) when allowed to copulate with females injected with an aziridinyl alkylating chemosterilant, tepa. Higher doses of tepa treatment on the female induced a proportionately higher sterility in the copulated male. But the induced sterility was not of a permanent nature. A progressive regaining of fertility was observed in the contaminated male with passage of time.
Effects of three biologically-active substances in the diet on the development and reproduction of the leather beetle, *Dermestes maculatus* De Geer.

Department of Zoology, University of Ife, Ile-Ife, Nigeria.

ABSTRACT

The effects of three biologically-active substances namely, Sulfanilamide, Adrenaline and Beta Sitosterol, in the diet on the development and reproduction of the leather beetle, *Dermestes maculatus* have been studied at 70% relative humidity and variable temperatures. Adrenaline and Beta Sitosterol each at 1.0% w/w, 0.5% w/w, 0.1% w/w, concentrations in the diet respectively produced loss in weight and an increase in larval developmental period. Sulfanilamide at 0.1% w/w concentration in the diet stimulated growth and reduced larval developmental period. The highest survival of larvae to the adult stage was found among larvae fed on fish meal containing Sulfanilamide while the highest mortality occurred in the fish meal containing Adrenaline and Beta Sitosterol; 1.0% w/w Sulfanilamide in the diet produced a significant increase in the proportion of males to females.

The possible implications of these results to control measures for the leather beetle were discussed.

R.A. BALOIJON.

The control or suppression of the oriental fruit fly by the sterile-insect release method is potentially feasible if specific conditions are fulfilled, especially if the species can be mass cultured. The experiment on selection of 5 bulking materials as sawdust, corn cob, sugarcane bagasse, rice husk and milled paper which was mixed in rice bran medium containing sugar, brewer's yeast, preservatives and water to provide a suitable medium texture for larval development of the oriental fruit fly. Studies were conducted to compare the effectiveness of rice bran and bulking material media with wheat bran medium (same ingredients as rice bran medium except wheat bran in place of rice bran and bulking material) on pupal recovery, pupal weight and adult eclosion. The results of this experiment showed that there were no significant differences in percent pupal recovery (P=0.05) and adult eclosion between rice bran and corn cob medium and wheat bran medium. However, pupae reared from wheat bran medium were larger than those reared from all media.

The design for an expandable and convenient refrigerated table for immobilizing insects which accommodates two workers per unit is presented. The table allows multiple units to be connected in series and mechanical components to be isolated from the work area.
There are different measures which are used to describe the dynamics of laboratory populations. Apart from very simple ones they are in general derived from an age-specific life- and fertility schedule by means of either the continuous time Lotka model or the discrete time Leslie model. Both models are standard in demography as well as in ecology and have been used as a basis for different quantitative studies in the mass rearing of tsetse flies.

This investigation was initiated by the observation that the "weight structure" of the offspring (puparia) has an essential influence on growth parameters like the emergence rate or the sex ratio. In order to study the further consequences on expanding colonies (all puparia are returned to the colony) as well as on producing colonies (kept at a constant size by continuous removing of pupae) a growth model with age- and weight-specific parameters is formulated and used as a basis for different calculations concerning the growth and productivity of laboratory populations. The usefulness of this approach is demonstrated by considering some special questions like e.g.:

(a) How many cages are necessary in order to get a certain amount of males for monthly release and what is the corresponding colony size?

(b) What is the effect of removing pupae of weight class A (the lowest class) on the growth rates?

(c) What is the effect of using no B-males for release?

(d) What is the effect of increasing the initial sex ratio from e.g. 200:60 to 200:50?

A quick answer to these (and other) questions is possible by using a program-package, which enables the performance of organizational procedures (data manipulation) as well as computational procedures (calculations of statistics, growth rates, etc.)
Before investigating any effect of gamma radiation on the germ cell of Acrididae Species; it is thought necessary to study germ cell differentiation in two species. The species were Heteracris littoralis and Mioscirtus wagneri.

Both species underwent a fixed number of encysted spermatogonial divisions. Data showed clearly that the number of cells per spermatocyte cyst fall at $2^6$ for both species. Cell division and differentiation with each cyst is synchronous and clonal giving almost fixed cell number in cysts of one particular type. These results also predict that there would be 256 sperm per bundle (Table 1.).

Table 1:

Number of divisions in grasshopper spermatogenesis

<table>
<thead>
<tr>
<th>Species</th>
<th>No. of generations</th>
<th>Primary sperm per cyst</th>
<th>sperm per bundle</th>
</tr>
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<tbody>
<tr>
<td>Heteracris littoralis</td>
<td>6</td>
<td>64</td>
<td>256</td>
</tr>
<tr>
<td>Mioscirtus wagneri</td>
<td>6</td>
<td>64</td>
<td>256</td>
</tr>
</tbody>
</table>
Two crossing types have already been obtained in two *Ephesia cautella* populations at two different countries. The first one was imported from USA and was called strain A. The males of this strain were incompatible when crossed to females of strain B which was raised from adult moths collected from Baghdad date stores in Iraq. However, the incompatible cross (B x A) occasionally results in partial fertility which sometimes exceeds 4% fertile eggs. This incompatibility phenomenon was proved to be transmitted through maternal cytoplasm.

Simultaneously such two different crossing types have been obtained in strains established from adult moths caught in a single date store on the same day. Therefore, it was thought necessary to measure the percentages of the two crossing types in Iraq by analysing the cytoplasm of *E. cautella* females, caught in dry-food stores, for several years.

On testing for cytoplasmic incompatibility in 224 *Ephesia cautella* females (Table 1), collected from dry-food storage structures in Iraq during the past four years, two crossing types were also found. The percentage of baghdad (B) mating type was 93.50%, while American (A) mating type was 6.50%. However, approximately 61% of the females collected from stores containing imported dry products were of American (A) crossing type, and only 29% were of the Iraqi (B) type. On the other hand, dry-date stores contained predominantly (96%) individuals of the Iraq strain. Theoretically speaking, under conditions of panmixia where the 2 crossing types are equally viable, A crossing type will quickly eradicate B crossing type unless there is a very strong counter-selection for individuals of B crossing type. More females should be analyzed in future in order to have a clear picture of *E. cautella* situation.

Table 1. Testing for preliminary determination of the crossing types of *Ephesia cautella* collected from dry-product stores in Iraq:

<table>
<thead>
<tr>
<th>Year</th>
<th>No. of tested</th>
<th>No. of crossed (pair)</th>
<th>No. of eggs examined</th>
<th>B</th>
<th>A (%)</th>
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<td>1977-78</td>
<td>76</td>
<td>387</td>
<td>61033</td>
<td>3*</td>
<td>74*</td>
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<tr>
<td>1979</td>
<td>88</td>
<td>266</td>
<td>49378</td>
<td>6</td>
<td>82</td>
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<tr>
<td>1980</td>
<td>39</td>
<td>126</td>
<td>20438</td>
<td>7</td>
<td>32</td>
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<tr>
<td>1981</td>
<td>43</td>
<td>215</td>
<td>30186</td>
<td>0</td>
<td>43</td>
</tr>
<tr>
<td>Total</td>
<td>246</td>
<td>994</td>
<td>161035</td>
<td>16</td>
<td>231</td>
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</table>

*Progeny of one female mated apparently with both cytoplasmic types but were lost.*
Some advantages of Habrobracon in radiation genetic research include: (1) the elimination of lethals introduced by sexual reproduction due to their propagation by parthenogenesis and, (2) the seriation of eggs in known meiotic stages in the ovarioles at the time of exposure to various environmental conditions. Adult virgin females cultured at 30°C were exposed to 0, 2500, 3500, 4500 and 6500 R of Cobalt-60 gamma radiation (238 R/minute). Egg production (fecundity) and hatchability (fertility) were taken as the criteria of radiation damage to the germ cells. These parameters were recorded for 20 days. A total of 4,048 eggs were scored. The following were obtained:

<table>
<thead>
<tr>
<th>Gamma ray dose (R)</th>
<th>mean # eggs/female</th>
<th>fecundity</th>
<th>% fertility</th>
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<tr>
<td>0</td>
<td>6.71</td>
<td>1630</td>
<td>97</td>
</tr>
<tr>
<td>2500</td>
<td>4.96</td>
<td>1160</td>
<td>73</td>
</tr>
<tr>
<td>3500</td>
<td>3.12</td>
<td>536</td>
<td>64</td>
</tr>
<tr>
<td>4500</td>
<td>2.88</td>
<td>486</td>
<td>47</td>
</tr>
<tr>
<td>6500</td>
<td>1.73</td>
<td>236</td>
<td>20</td>
</tr>
</tbody>
</table>

The data indicated that fecundity and fertility progressively decreased with increasing radiation exposure except that no significant differences were observed in fecundity between controls and those exposed to 2500 R. Temporary sterility was observed on days 7-10 in those wasps receiving 4500 R. At 6500 R eggs were oviposited on days 1-5; thence permanent infecundity.

The fecundity and fertility results illustrated the differential radiation sensitivity of the various meiotic stages during oogenesis. In general, the radiation sensitivity progressively decreased with reference to these oogenic stages: zone of differentiation, oogonia, prophase I and metaphase I. The high sensitivity of the zone of differentiation indicates the vulnerability of cells undergoing active cell division. This finding is consistent with previous reports of radiation being more detrimental to dividing than non-dividing cells.
A recent breakthrough related to mass-production of laboratory reared Culex tarsalis for sterile-release experiments is our ability to separate the males and females by a genetic means. Such a system saves considerable time and space by the early elimination of unwanted females which are not used in the release. It also eliminates the tedious task of separating the sexes by hand or by mechanical means. The system is based on linking resistance of the insecticide, malathion, to the male-determining chromosome.

The sexing scheme involves a translocation which by alternate segregation gives viable sperm binding the male-determining chromosome bearing the M gene for maleness with the part of the autosome carrying the malathion-resistant gene, Mal. The close linking of the 2 genes with little if any recombination between them is inherited in subsequent generations by 99.8% of the males fathered by these translocation-carrying males. Thus, a 0.1 ppm discriminating dose to first-instar larvae is lethal to females and allows males to survive, eliminating the need for further sexing prior to radio-sterilization and the release of males.

We propose using this sex-sorting strain for future sterile-male releases once we can successfully select for competitiveness in laboratory rearing. Heterozygosity can be introduced into the strain by mating males with any malathion susceptible females.

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A complexity of abnormalities (duplication and deformation of elements) was found in eupyrene sperm cells in the progeny of Heliothis subflexa (Guenée) ♀ × H. virescens (F.) ♂ by using a rapid air-dry procedure for light microscopical study. The sperm cell acrosome, nucleus, neck region, and tail elements were clearly delineated in these preparations. Where sperm were scored in the parents, less than 2% were found to be abnormal. In contrast, more than 60% were abnormal in F₁ hybrids. Frequency of aberrations in BC generations gradually approached normal parental values. The presence of abnormal eupyrene spermatozoa might be used to identify field-released F₁ and early BC generation sterile males.
The permeability of the testis of *Anagasta kuehniella* has been studied by incubating the gonad in vitro with horseradish peroxidase as an electron-dense tracer. The exogenous protein freely penetrates across the complex and thick wall of the gonad at all stages of the postembryonic life (larval, nymphal, and adult stages). It impregnates the testicular fluid but is totally excluded from the interior of the cysts. Even the dividing gonia and the young spermatocytes appear isolated from the surrounding fluid. Consequently, and in contrast to other cases studied up to now, very few germ cells can be reached by exogenous macromolecules. The primary gonia which are not yet enclosed in the form of cysts and bathe directly in the testicular fluid are the only ones in contact with the tracer. The blood-testis barrier in *Anagasta* lies therefore exclusively in the cyst envelope. This fact, together with the permeability of the testis wall at all stages of the postembryonic life, is discussed in relation to the hypothetical action of a blood macromolecule on the spermatogenesis of some lepidopteran species.

Male Mediterranean flour moths, *Anagasta kuehniella* (Zeller), placed in continuous light (LL) from the early pupal stage on were partially sterile as adults. When old adult moths were transferred to alternating light and dark (LD), fertility was not recovered, but fertility was recovered when fairly young males were similarly transferred. Multiple matings of males indicated that recovery was correlated with sperm in the testes at the LL to LD transfer becoming available for ejaculation. Few morphological abnormalities were found in the sperm of LL males, but there was less activity than in the sperm of LD males. At 1 week after eclosion, the testes of LL males contained more eupyrene sperm bundles than did the testes of LD males, but the LL males had somewhat fewer bundles overall in their reproductive tracts. Disruption of the circadian rhythm regulating movement of sperm from the testes was suggested as the most probable cause of the sterility of LL males.
To test the hypothesis that a maternally transmitted cytoplasmic microorganism is involved in male hybrid sterility found in *H. subflexa* (Guenée) X *H. virescens* (F.) hybrid and backcross progeny, we reared *H. subflexa*, hybrids and backcross progeny on larval diets containing high concentrations of tetracycline, penicillin G, chloramphenicol, ampicillin, gentamicin, rifampin, and 5-iodo-2'-deoxyuridine. The insects tolerated relatively high concentrations (from 120 mg/liter to 6 g/liter) of antibiotics in the larval diet and showed virtually no changes in larval or pupal developmental time, adult lifespan, or fertility. Hybrid and backcross males reared on such adulterated diets were as sterile as those reared on conventional diets.

The effects of female accessory reproductive gland removal on egg hatchability and sperm penetration of the egg were determined in 3 dipteran species; the screwworm, *Cochliomyia hominivora* (Coquerel), stable fly, *Stomoxys calcitrans* (L.), and face fly, *Musca autumnalis* (DeGeer). None of the eggs from glandless screwworm and face fly females hatched, while only 1% of the stable fly eggs hatched. Cytological examination revealed that only about 1% of the screwworm eggs, 6% of the stable fly eggs, and 2% of the face fly eggs were fertilized. Eggs deposited by glandless screwworm females lacked the adhesive coating that attaches them to each other or to the oviposition substrate.
Karyotypic analyses of mitotic and meiotic chromosomes of the cotton boll weevil. *Anthonomus grandis* grandis Boheman, revealed the diploid number to be 42 (20 autosomes plus an Xy). The Xy has the typical parachute shape configuration as described for other related species. The evidence for supernumerary chromosomes is also present.

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Only a single locus (Tp1) is known in the *Drosophila melanogaster* genome that leads to early lethality when present as a heterozygous duplication (three doses) or deficiency (one dose). We report the recovery of third instar larvae (and of occasional adults) carrying a duplication for the triplo-lethal locus, Dp(Tp1). Karyotype analysis of the larvae showed that the individuals surviving were almost entirely 3X;2A metafemales. We examined the question of whether the entire X or a single X locus was a major factor permitting survival. X-Y translocations were used to produce females hyperploid for different portions of the X and carrying Dp(Tp1). Analysis of metaphase chromosomes by quinacrine fluorescence pattern indicates that the X chromosome region between 6D and 7DE must be present in an extra copy to enhance the survival of Tp1 duplication-bearing females. Another type of experiment suggests that it is the region between 7C and 7DE which is essential.
GENETICS OF DIABROTICA (COLEOPTERA: CHRYSDOMI
DAE): INHERITANCE OF XANTHINE DEHYDROGENASE,
HEXOKINASE, MALATE DEHYDROGENASE, AND ESTERASE
ALLOZYMES IN TWO SUBSPECIES OF D. LONGICORNS

The modes of inheritance for allozymes of xanthine dehydrogenase (Xdh), hexokinase (Hk),
malate dehydrogenase (Mdh), and an esterase (Est-m) were determined by electrophoretic
analyses of specimens from intra- and interspecific crosses of Diabrotica longicornis
(longicornis) (Say) and D. l. barbert Smith and Lawrence. Comparisons of $F_1$ and
the corresponding $F_1$ enzyme patterns provided evidence of co-dominant alleles segregating at
autosomal loci for Xdh, Hk, and Mdh. A sex-limited autosomal locus was implicated in the
expression of Est-m electromorphs. The data indicate that the various allozyme systems
are allelic in the two subspecies.

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RECIPROCAL TRANSLOCATIONS AND PARTIAL
CORRELATION OF CHROMOSOMES IN THE
STABLE FLY.

An initial investigation of the genetics of the stable fly, Stomoxys calcitrans (L.), is
described. Two recessive, autosomal mutants, carmine eye (ca) and rolled down wing (rd),
were assigned to chromosomes 2 and 4, respectively. Sex is apparently determined by a
locus on chromosome 1. Crossing over is restricted to females. Six reciprocal translocations
were induced with gamma radiation and used to assign ca and rd to their respective
chromosomes.

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HOUSE FLY GENETICS: FERTILITY
DIFFERENCES IN MATINGS OF MALE
TRANSLOCATION HETEROZYGOTES WITH
DIFFERENT "NORMAL" FEMALES

Fertility variations were frequently observed when heterozygous translocation-bearing male
house flies, Musca domestica L., from a number of strains were mated with non-
translocation-carrying females from two different sources. The data indicate a need to
test the fertilities of translocation-bearing insects against different physiological or
genetic backgrounds in selecting appropriate types for pest control programs.
A genetic selection experiment in the cotton boll weevil, *Anthonomus grandis* Boheman, was used to evaluate the degree to which variation in postirradiation survival may be under genetic control. Ten generations of family selection for 14-day postirradiation survival increased survival from 35 to almost 70% and increased life expectancy after irradiation by about 4 days. Estimates of realized heritability of half-sib family means of near 0.3 were required for this magnitude of response. The results indicate that selection for genetic improvement has merit in the development of a mass-rearing stock for release in a sterile insect program.

Female tobacco budworms, *Helothis virescens* (F.) (Lepidoptera: Noctuidae) that emerged in the laboratory contained no mature eggs, although oocyte development has proceeded as far as yolk deposition in the ultimate egg of each ovariole. After 18 h, ca. one mature egg per ovariole was present. In the females subsequently remained unmated, maturation of additional eggs was curvilinear with age and leveled off at ca. 500 eggs per 9 at 120 h. If the females mated, there was an inverse relationship between egg production and age mated; maximal production occurred when females mated the first night postemergence (ca. 30 h) and production then diminished until females mating at 5 days or more laid only slightly more eggs than virgin females. However, there was also a direct relationship between length of life and age of mating.

Females that mated at 1 or 2 days showed a significant correlation between numbers of eggs produced and longevity and between numbers of eggs and pupal weight. Virgin females and females that mated at 3 days showed a significant correlation between eggs and longevity but not between eggs and pupal weight. There was no significant correlation between eggs and pupal weight or longevity for females that mated at 5 days or later.