Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture and FAO/IAEA Agriculture and Biotechnology Laboratory, Seibersdorf

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TO THE READER

Letter from the Section Head

A European Union (DG-VI) sponsored Seminar on the "The Sterile Insect Technique as an Environment-Friendly and Effective Insect Control System" took place in November in Madeira, Portugal, with the participation of representatives from fruit industry, universities and research institutions of all countries of Southern Europe, and staff of the Insect & Pest Control Section as invited speakers. This was a major breakthrough for SIT, since the EU has until recently not supported, much less promoted the SIT. Except for the Madeira Mediterranean fruit fly control project initiated with EU and IAEA support in the second half of the 1990s, all operational SIT have been implemented in other parts of the world. A review of the world-wide distribution of sterile insect mass-rearing facilities indicates that over 60 % of these SIT facilities are located in the Western Hemisphere. where application of SIT technology is most advanced in Latin America, USA and Canada (followed by Asia and Africa).

There is increasing realization in the EU that the use of insecticides as now practised in Europe is clearly not sustainable in view of the major disadvantages for the environment, beneficial organisms and human health. Despite a number of successes in the implementation of an integrated pest management approach, the bulk of insecticide application in the world is still in the developed countries. The highest consumption is actually in Europe, reaching up to 21 kg of insecticides per hectare per year in the Netherlands.

The wide-spread application of insecticides broad-spectrum generally biological with activity has been causing outbreaks of secondary pests because of the inadvertent destruction of natural enemies, as well as wide-spread incidence of insecticide resistance in pests. Recently there has been increasing concern about the effects of pesticides on the endocrine system of both humans and wildlife. Greater public awareness and of the concern environmental community about harmful residues in food and negative effects on the

environment has already led to the reversal of misguided policies that subsidized the use of insecticides, and environmental appropriateness and food safety considerations are becoming the main criteria for the selection of acceptable methods for pest control.

In the Mediterranean region, the Mediterranean fruit fly causes devastating direct losses if not controlled, requiring up to 12 insecticide treatments in some regions to produce worm-free fruit. The resulting damage of this intensive insecticide use to non-target beneficial organisms, disruption of biologically based controls of other orchard pests, and general contamination of the environment and residues on fruit, are major forces driving the need for more environment-friendly methods such as SIT to control fruit flies. Some of the initial pilot SIT projects took place in the Mediterranean region in the 1960's and early 1970's. Nevertheless much less advances have been made in the application of SIT, even though environmental concerns due to intensive insecticide use against medfly and olive fly are increasingly of major importance, particularly in coastal areas where tourism and orchards coexist.

The main obstacles in the Mediterranean Basin for SIT implementation against medfly have traditionally been:

- a) the cosmetic damage some commercial citrus varieties suffer due to sterile female stings, limiting use of SIT largely to eradication campaigns;
- b) the difficulty to implement in the Mediterranean Basin quarantines to protect areas from which medfly would be eradicated; and
- c) the perceived high cost of SIT.

Advances made over the last decade in relation to all of these issues have led to a re-evaluation of the applicability of SIT. Viable male-only or genetic sexing strains, developed for medfly at the FAO/IAEA Agriculture and Biotechnology Laboratory, allow the production and release of almost exclusively male sterile insects. As a result, in the absence of sterile females and their fruit stings, the possibility now exist of using of SIT for routine fruit fly control rather than in the only for eradication programmes. past Consequently no quarantines need to be established and sterile male releases can now be used for regular control purposes, replacing aerial insecticide sprays with the environmentfriendly aerial release of sterile males. Control of medfly without insecticides also allows the implementation fully of integrated pest management (IPM) schemes, including the augmentative release of natural enemies against other fruit pests, which are normally not effective because they are eliminated by the insecticide applications against medfly.

The feasibility of the SIT control approach has been confirmed by a number of benefit-cost analyses, which indicate that the economics of an area-wide approach using SIT for medfly control are comparable with insecticide-based control, even without taking into account the resulting environmental costs from the insecticide use. These studies also indicate that the regular requirement of sterile males for medfly control will open the way for the commercial production and delivery of SIT. Currently the citrus industry in Israel spends an average of ca. US\$ 80/hectare/year for a very effective centrally-organized aerial bait-spray system for medfly control (not including the recurrent liabilities resulting from damages to the honeybee industry). The annual cost of areawide medfly control using SIT in Israel's Arava Valley is currently costing a comparable amount, even though expensive shipment of sterile males from Guatemala to Israel is In view of these favourable involved. economics, preparatory actions are in progress to expand SIT control areas in Israel and Jordan, and to include in the future also Gaza.

above developments have already The resulted in medfly SIT programmes in various stages of development in Madeira, Israel and Jordan, as well as feasibility studies in progress in North Africa, Spain, Portugal and Corsica. considerable activity indicates This increasing interest in the region in substituting medfly control based on insecticide sprays with environment-friendly medfly control based on The association of Mediterranean citrus SIT. producers (CLAM) in the Mediterranean Basin has welcomed the EU endorsement of the SIT

control approach, and also organized a conference on the use of SIT in conjunction with their recent General Assembly in Valencia, Spain (see report on page 20).

For medfly and other fruit flies, the worldwide weekly production capacity of sterile flies is reaching several billion sterile flies. However, in Europe with the exception of the Madeira mass rearing facility, the lack of a source of sterile medflies for use in SIT control programmes is presently a major limiting factor for the expanded application of this technology in the Mediterranean Basin. The establishment of large regional mass-rearing facilities, making use of economies of scale in the production of sterile males, is therefore of high priority to address the existing and potential sterile fly demand in the Mediterranean Basin. The control approach to SIT, involving routine SIT application for medfly management, will assure the long-term demand required for the commercialization of the supply of sterile males.

Finally, in relation to the above I would like to add that a Fruit Fly Thematic Planning meeting took place in Vienna to review the future involvement of FAO/IAEA in the field of fruit fly management. The major recommendation by the outside consultants from fruit industry and various governments was that fruit industry and pest management the infrastructure is sufficiently advanced in three main regions where utilization of SIT for practical fruit fly control should focus: Central America, the Southern Cone of South America and the Mediterranean Basin.

As 1999 draws to a close, I would like to send seasonal greetings on behalf of everyone in the Insect Pest Control Sub-programme and wish you all the best for the new millennium.

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Jorge Hendrichs Head, Insect Pest Control Section

B. STAFF

The Subprogramme staff, consisting of those in the Joint FAO/IAEA Division located in the Vienna International Centre, those in the FAO/IAEA Agricultural and Biotechnology Laboratory in Seibersdorf Laboratory and field experts, are listed below.

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FORTHCOMING EVENTS

Research Co-ordination Meetings (RCM)

"Improved Attractants for Enhancing the Efficiency of Tsetse Fly Suppression Operations and Barrier Systems used in Tsetse Control/Eradication Campaigns", 21 - 25 February 2000, Bamako, Mali. 3rd RCM.

"Enhancement of the Sterile Male Technique Through Genetic Transformation Using Nuclear Techniques", 14 - 18 August 2000, Sao Paulo, Brazil, 3rd RCM.

"Development of Improved Attractants and their Integration into SIT Fruit Fly Management Programmes", 14 - 18 August 2000, Brazil, 1st RCM. "Automation in Tsetse Mass-rearing for Use in Sterile Insect Technique Programmes", February 2001, Burkina Faso, 4th and final RCM.

"Evaluating the Use of Nuclear Techniques for the Colonization and Production of Natural Enemies", March 2001, Guatemala, 2nd RCM.

"Quality Assurance of Mass Produced and Released Fruit Flies", April 2001, Hawaii, USA, 2nd RCM.

Consultants Meetings

"Development of cost effective diets for mass production of tsetse flies", 15 - 19 May 2000, IAEA, Vienna, Austria.

"Genetic Sexing and Population Genetics of Screwworm", 29 May - 2 June 2000, IAEA, Vienna, Austria. "Thematic planning for tsetse fly intervention strategies and related research", 11 - 15 September 2000, IAEA, Vienna, Austria.

FAO/IAEA Training Courses

FAO/IAEA Regional Training Course on "The Sterile Insect Technique as a Component for Integrated Area-wide Tsetse and Trypanosomosis Management", 20 March to 14 April 2000, Tanga, Tanzania. Applications for this course close on 15 January 2000. "Revision and Reinforcement of Technical Criteria and Control and Detection Methods of the Binational Programme Chile - Peru for the Eradication of Mediterranean fruit fly", February 2000, Peru.

IV.Other meetings

Regional Forum for Africa on Tsetse SIT, 2 - 5 May 2000, Addis Ababa, Ethiopia.

Workshop to review the Agricultural Research Service (ARS) program on exotic pests, fruit flies and quarantine, 24 - 26 January 2000, Honolulu, , Hawaii, USA.



Research Co-ordination Meetings (RCM)

"Automation in Tsetse Mass Rearing for Use in Sterile Insect Technique Programmes", 12-16 April 1999, Vienna, Austria, 3rd RCM.

"Medfly Mating Behaviour Studies Under Field Cage Conditions", 29 June - 3 July, 1999, Antigua, Guatemala. 4th and final RCM.

"Molecular and Genetic Approach to Develop Sexing Strains for Field Applications in Fruit Fly Sterile Insect Technique Programmes", 12 - 16 July 1999, Tapachula, Mexico, 3rd RCM.

"Quality Assurance of Mass Produced and Released Fruit Flies", 27 September - 1 October 1999, Vienna, Austria. 1st RCM.

Consultants and Other Meetings

"Formulating an Approach for Integrated Area-Wide Tsetse / Trypanosomosis Intervention in the Lambwe Valley, Kenya", 6 and 7 April 1999, Vienna, Austria.

"Alternative Tsetse Suppression Techniques for Use before Sterile Male Releases", 8 and 9 April 1999, Vienna, Austria. "Genetics Application to Improve the SIT for Tsetse Control/Eradication including Population Genetics", 3 - 7 October 1999, Mombassa, Kenya. 2nd RCM.

"Evaluating the Use of Nuclear Techniques for the Colonisation and Production of Natural Enemies", 18 - 22 October 1999, IAEA, Vienna, Austria. 1st RCM.

Proceedings are available on request at the Insect & Pest Control Section's office.

"Thematic Planning for Fruit Fly Control Activities", 15 - 19 November 1999, Vienna Austria.

Proceedings are available on request from the Insect & Pest Control Section's office.

FAO/IAEA Training Courses

Second Workshop on Quarantine Procedures needed for the Creation of a Fruit Fly Free Zone in Tacna and Moquegua, Moquegua, Peru, 6 - 9 April 1999

The Interregional Training Course on the "Use of the Sterile Insect and Related Techniques for the Area-Wide Management of Insect Pests", University of Florida, Gainesville, Florida, USA, 14 April - 19 May 1999. (Cofunded by the US Government, FAO and IAEA.) IAEA/AOAD Regional Training Course on "Techniques Used for Area-Wide Control/Eradication of the Old World Screwworm Fly", Screwworm Fly Laboratory, Institut Haiwan, Malaysia, 31 May to 12 June 1999

Second National Training Course on Integrated Fruit Fly Control, November 1999, Arequipa, Peru.

Other meetings

Third meeting of the Working Group on Fruit Flies of the Western Hemisphere, Guatemala City, Guatemala, July 4-9,1999 which was cofunded by the Joint FAO/IAEA Division.

The XIVth International Plant Protection Congress including a symposium on "The Sterile Insect Technique, Past, Present and Future", July 25-30, 1999, Jerusalem, Israel.

International Scientific Council for Trypanosomiasis Research and Control 25th meeting, 27 September - 1 October 1999, Mombasa, Kenya.

Regional Screwworm Coordinators Meeting, October 1999, Managua, Nicaragua.

Comité de Liaison de L'Agrumiculture Méditerranéenne (CLAM) Annual Meeting, including a special conference on the "Use of SIT to Control and Eradicate the Mediterranean Fruit Fly Within the Mediterranean Region", 13-15 October 1999, Valencia, Spain.

IOBC Symposium on "Evaluating Indirect Ecological Effects of Biological Control", 17 -20 October 1999, Montpellier, France.

EU Seminar on "The Sterile Insect Technique as an Environmentally Friendly and Effective Insect Control System", November 1999, Madeira, Portugal.

Programme Against African Animal Trypanosomosis (PAAT) Committee Meeting, 22 - 23 November, Rome, Italy.



TECHNICAL CO-OPERATION PROJECTS

Over the last four years, the Section has had technical responsibility for over 35 technical co-operation projects. They fall under four major areas, namely:

Current Operational Projects (1999-2000) are:

- ALG/5/017 Control of the Date Moth using SIT/F-1 Sterility Principle
- ETH/5/012 Integrating SIT for Tsetse Eradication
- INT/5/145 Promotion and transfer of Sterile Insect Technology
- IRQ/5/014 Field Monitoring and Laboratory Rearing of Old World Screwworm
- ISR/5/009 Feasibility Study of SIT for Medfly Eradication
- JAM/5/006 Eradication of the New World Screwworm: Preparatory Phase
- JAM/5/007 New World Screw-worm Eradication
- JOR/5/007 Feasibility of Area-wide Control of Medfly by SIT
- LEB/5/013 Feasibility of Integrated Control of Medfly Using SIT
- MAL/5/023 Feasibility of Old World Screwworm Control through SIT
- MAR/5/009 Control of Diamondback Moth by Sterile Insect Technique
- PAL/5/002 Area-wide Application of SIT for Medfly Control
- PHI/5/026 Integrated Control of Oriental Fruit Fly on Guimaras Island
- POR/5/005 Mediterranean Fruit Fly Programme on Madeira
- RAF/5/040 SIT for Tsetse and Trypanosomosis Management in Africa

- Tsetse
- Fruit Flies
- F-1 Sterility for the Control of Lepidopteran Pests
- Screwworm and Others
- RAW/5/008 Preparing to Combat the Old World Screwworm in West Asia.
- RLA/5/039 Bi-national Project Chile-Peru: Eradication of the Fruit Fly in Southern Peru
- RLA/5/044 Preparing Caribbean Eradication of New World Screwworm.
- SAF/5/002 Feasibility Assessment for Fruit Fly Eradication Using SIT
- SLR/5/002 Feasibility Study for a Mass Rearing Insect Facility
- SYR/5/019 Controlling Codling Moth for Apple Crop Using SIT.
- THA/5/044 Extension of Areas Under Integrated Fruit Fly Control
- TUN/5/019 Control of the Date Moth using Radiation Sterilisation.
- UGA/5/021 Integrated Tsetse Control in Buvuma Island Phase II
- URT/5/018 Post Eradication Entomological and Veterinary Monitoring on Zanzibar
- URT/5/019 Support to National Tsetse and Trypanosomosis Management

In keeping with our policy to highlight activities in a few of our Technical Cooperation projects in each Newsletter the following projects are discussed in this issue:

Feasibility study for an insect mass rearing facility in Slovakia (SLR5002)

The increasing interest in the SIT as a means to control or eradicate several pest populations, particularly the Mediterranean Fruit Fly, has lead to an increased demand for sterile insects. This demand is however unmet due to the lack of rearing facilities, and there is a clear opportunity for a commercial rearing facility.

A survey of possible sites identified Eastern Europe as having an ideal combination of skilled labour, low labour costs and ready access to the major Mediterranean basin market for medfly. The Slovak Republic took up the idea, and with assistance from the Agency initiated a feasibility study. Slovakia has the additional advantage of proximity to the IAEA headquarters and the Seibersdorf laboratories.

In order to make a commercial success from the proposal the study will examine the possibility of a multi-species rearing facility, to combine medfly with tsetse and lepidoptera. Individual modules within the factory will be design for easy conversion from one group to the other, so that production can be tailored to the current demand.

The study has been set up as a contract to the Forest Research Institute, Zvolen, Slovakia, with the IAEA providing expert services to cover fields where expertise is lacking in Slovakia. A meeting was held in Vienna in October between the project leader and his assistant, and experts in the fields of medfly, lepidoptera and tsetse rearing. From this meeting a basic module design was derived that could be used for medfly or tsetse, but the problems of scales in lepidoptera rearing mean that conversion from medfly to lepidoptera may not be economic.

The study will investigate all factors, including environmental, social and legal, that could influence the facility. The study is due to be completed by July 2000.

Interest has already been shown in this project by Israel, and it is anticipated that a ready source of sterile flies in the region will further stimulate SIT activity.

International Symposium on "The Sterile Insect Technique, Past, Present and Future"

XIV International Plant Protection Congress (IPPC) held in Jerusalem from 25-30 July 1999

As part of activities under TC project INT/5/145 "Promotion and Transfer of Sterile Technology" Insect an International Symposium on "The Sterile Insect Technique, Past, Present and Future" was organised and chaired during the XIV International Plant Protection Congress (IPPC) held in Jerusalem from 25-30 July 1999. The IPPC was attended by ca. 2000 participants from close to one hundred countries. During the opening Plenary of the IPPC, the plant protection authorities of Israel showed a movie to all delegates which prominently included the joint SIT activities with Jordan against medfly under TC project ISR/5/009. The SIT symposium, which involved a whole one morning session of the IPPC had been co-organized over the last year with Dr. Yoram Rossler from Israel. Among the speakers was also Dr. John Mumford. The Symposium was well attended, reflecting a growing interest in area-wide and more environment-friendly approaches to insect pest management as a response to the growing trends against conventional large scale pesticide applications.

During a visit to the SIT pilot area in the Arava Valley region where project activities under TC Project ISR/5/009, "Feasibility Study of SIT for Medfly Eradication" are in progress, discussions were held with various members of the project team in the Sapir Center and a visit was made to the Arava Medfly Eradication Project (AMEP) medfly emergence centre and some of the sterile fly release areas. The QC data of the flies that have undergone long distance shipment of pupae from Guatemala to Israel is remarkably This reflects very well on the good. management at the El Pino Facility in Guatemala and emphasizes the potential that long distance shipments of sterile medflies will have on the eventual commercialization of medfly production. An essential contractual component for the implementation of the project in the Arava is the release of sterile males only. The percentage of sterile female flies that are received in the shipments from Guatemala for aerial release over the Arava valley is well below 1%, confirming the excellent genetic sexing characteristics of the The regular shipment of sterile strain. medflies from Guatemala to this Centre is well established and it has enabled the project to proceed as planned.

The current programme in the Arava releases sterile males in both Jordan and Israel. The logistics of carrying out an aerial release over these areas is a sensitive issue but has been satisfactorily solved.

The next phase of the programme will be the extension of the releases to the Negev. This expansion will require an extra 4 million males/week and considerably more flying time. The base-line data collection for the area is in process so that releases can begin as soon as the extra funds from MERC (USAID) are available. Further expansion of the project into other areas in Israel and Gaza will probably require a re-evaluation of the supply of flies. For the proposed large scale regional programme a production facility in closer proximity will be required.

In terms of impact, there are very encouraging figures so early in the project, confirming the significant economic benefit of the establishment of fly free areas using SIT.

Fly Free Areas	Date
Paran	1994
Sapir	1997
Ein Yahav	1998
Hazeva	1999
Neot H'kikar	1999
Ein Tamar	1999

The impact can be measured both in terms of agricultural areas which have now been designated as fly free together with increase in exports to the USA from those areas. Below are given some data on these impact factors, showing a 50-fold increase in the economic value of vegetables exports from the Arava:

Vegetable Export to	Date
USA (US\$1000,s)	
112	1994
415	1995
1,899	1996
2,713	1997
5,077	1998



EXISTING AND PLANNED CO-ORDINATED RESEARCH PROJECTS (CRP)

Enhancement of the Sterile Insect Technique (SIT) through Genetic Transformation Using Nuclear Techniques (D4. 10.12)

The second RCM was held in Penang, Malaysia, 26-27 May 1998, in conjunction with the FAO/IAEA International Conference on "Area-wide control of insect pests integrating the sterile insect technique and related nuclear and other techniques". The participants reviewed the current status in the field of transformation of non-drosophilid insects and recommended research strategies for the remaining period of the CRP.

Since the initiation of this CRP in 1996, dramatic advances have been made in our ability to introduce genes into insects. Specifically, there are now multiple strategies available to routinely generate transgenic insects. So far, three different mobile element systems were used successfully to transform the medfly. Participating laboratories of the current CRP are therefore now in a unique position to incorporate this technology into the SIT and they will continue to develop vectors and identify strategic gene systems for use in the SIT.

It was proposed to hold the next RCM in 2000 in conjunction with the Entomology Congress in Brazil. Furthermore, the CRP was extended into 2001 and, consequently, there will be four RCMs.

Expected duration: 6 years (1995-2001)

Contract Holders (2) from Greece and New Zealand

Agreement Holders (8) from Australia, United Kingdom, United States (3) and Italy (3).

Medfly Mating Behaviour Studies under Field Cage Conditions (D4.10.14)

The final Research Co-ordination Meeting was held from 29 June - 4 July in Antigua, Guatemala with 25 behaviourists and quality control experts participating (see page 31). The peer-review and editing of final papers to be published in the Florida Entomologist will be coordinated by J. P. Cayol.

Slow motion video-recordings of the sexual behaviour of wild and/or mass-produced flies have been collected from Argentina, Costa Rica, Greece, Guatemala, Israel, Kenya, Madeira, Mexico and Reunion for centralised analysis. A quantitative analysis (still in process) has shown that no consistent qualitative difference can be found between the courtship behaviour of males from the different wild populations.

Wild female flies exerting mate choice in field cage tests with host trees have been found to be the most reliable tool available to assess mating performance and sexual compatibility of mass-reared medfly males when competing with wild males for wild females. There is clear evidence from the tests and field assessment studies that some quantitative differences in terms of mating performance and sexual activity between mass-reared and wild flies can be detected.

Among the most important outcomes of the CRP it was shown that, for the countries represented in the CRP with the exception of some populations in Kauai, Hawaii, and Madeira, Portugal, no sexual incompatibility was encountered between wild medfly populations from different geographic origins.

Duration: 5 years (1994 - 99)

Contract Holders (8) from Argentina, Costa Rica, Greece, Guatemala, Israel, Mexico, Reunion and Kenya. Agreement Holders (1) from the United States.

A Molecular and Genetic Approach to Develop Sexing Strains for Field Application in Fruit Fly SIT Programmes (D4.10.15)

The third RCM was held in Tapachula, Mexico from 12-16 July, 1999 (see page 36).

During the course of the CRP genetic sexing strains have been introduced into mass rearing facilities in Argentina, Guatemala, Chile, Madeira and Crete. In addition Hawaii, South Africa, Peru and Western Australia are preparing to rear genetic sexing strains. The expertise developed in the CRP has been essential in order that this technology transfer meets the needs of the customer. In other fruit fly species, progress towards the development of genetic sexing strains has been made in the areas of polytene chromosome analysis and the isolation of genetic markers. There is also an increasing emphasis on the use of molecular techniques targeted to the cloning of sex determination genes.

The CRP has been extended for a further year into 2001.

Expected duration: 5 years (1995-2001)

Contract Holders (6) from Argentina, Bangladesh, Brazil, Greece, Guatemala, and the Philippines.

Agreement Holders (3) from Australia, Italy and the United States.

Genetic Applications to Improve the SIT for Tsetse Control/Eradication including Population Genetics (D4.20.05)

The second RCM was held from 3 - 7 October 1999, in Mombasa, Kenya in conjunction with the 25th OAU/STRC ISCTRC Meeting (see page 35).

The CRP focuses research on the population genetics of tsetse as a tool in the management of tsetse SIT programmes, using a range of modern techniques, with emphasis on the requirements of the tsetse eradication project in the Southern Rift Valley of Ethiopia. Expected duration: 5 years (1997-02)

Contract Holders (3) from Greece, Kenya and Burkina Faso.

Agreement Holders (6) from Greece, Kenya, Belgium, Canada, United States (2) and Italy

Automation in Tsetse Fly Mass-rearing for Use in Sterile Insect Technique Programmes (D4.20.06)

Several stages in the mass production of tsetse have been addressed so far. Progress has been good in the automated stocking of production cages, where it is now possible to emerge flies under controlled conditions into production cages to give the desired female to male ratio of 4:1 with less than 0.5% females remaining in the un-emerged pupae, *for G. austeni, G. palpalis gambiensis* and *G. pallidipes.* The necessary conditions for other species remain to be determined. The next stage in this work is to institute large scale evaluation of the procedures, and to introduce the technique into the routine running of all the Seibersdorf colonies and then to other colonies.

After emergence of the females, the remaining male pupae have to be handled. Work is now underway on controlling the emergence of these males by manipulating the holding temperature to allow synchronous emergence, and on chill holding of the adult males in preparation for release.

The automated recognition of males and females has given mixed results. A transport system has been devised that moves the flies one at a time at emergence past a camera, where high quality images can be captured. So far though the variation in light intensity and orientation of the flies is too great for the image recognition software to reliably separate males and females.

Work on an improved system to handle cages for feeding is progressing well. A first fully automated prototype proved to be too complicated and a second prototype is now undergoing trials and shows good promise of reducing the effort of cage handling by approximately ten fold. The system holds 63 large cages on a single trolley that can be moved to feed all the cages simultaneously and then returned to the larval collecting unit. Prototypes of the new system will be transferred this year to TTRI, CIRDES, KETRI and Addis for field evaluation using existing cages, and the locally available tsetse species. The system is also being further modified to incorporate recommendations resulting from the recent 3rd RCM held in Vienna in April 1999.

Other work has looked at the handling factors affecting flight ability of irradiated males, increasing cage holding density by the use of inserts, energy saving and blood decontamination. This last is a very important factor in the running of large colonies, and the possibility of using pasteurization or UHT sterilization is being investigated.

The 4th and final RCM is scheduled for February 2001 in Burkina Faso.

Expected duration: 6 years (1995-01)

Contract Holders: (5) from Austria, Czech Republic, Burkina Faso, Tanzania and Kenya.

Improved Attractants for Enhancing the Efficiency of Tsetse Fly Suppression Operations and Barrier Systems Used in Tsetse Control/ Eradication Campaigns (D4.20.08)

This CRP was set up to address the shortcomings in attractants for a number of important tsetse species where the standard odours used for *G. morsitans* and *G. pallidipes* are poor or ineffective, and in general to try to improve attractant effectiveness and reduce cost.

So far the Nile monitor lizard has been confirmed to be the principal host of *G*. *palpalis gambiensis* and *G*. *f. fuscipes* by blood meal analysis, but odour experiments using live lizards have been inconclusive.

A number of natural kairomones and their analogues have been synthesised, and tested in laboratory experiments or in the field. Decylaldehyde increased caches of *G. austeni*, and linoleic acid, a possible precursor of 1octan-3-ol also proved active.

Molecular modelling of the structure of known attractants has indicated possible new synthetic attractants, and several of these have proved active in laboratory tests. This approach could lead to the identification of not only more attractive and cheaper compounds, but also attractants for the current "difficult" tsetse species. Field investigations on these new potentially attractant compounds are underway.

Work is also going on to characterise the cuticular hydrocarbon sex pheromones of tsetse, for use in assessing population differences. Comparisons of conspecific populations have shown that the hydrocarbons mixtures are usually very similar.

The 3rd RCM will be held in Bamako, Mali, 21-25 February 2000.

Expected duration: 5 years (1995-2001)

Contract Holders (7) from Mali, Burkina Faso, Kenya, Uganda, Tanzania and Hungary.

Agreement Holders (2) from Switzerland and the United States.

Quality Assurance of Mass Produced and Released Fruit Flies (D4.10.16)

The first Research Co-ordination Meeting, to plan and co-ordinate the research, was held 1 - 5 November 1999 in the IAEA, Vienna, Austria (see page 37). Proceedings of this meeting are available from the Section or via our web site.

The objective of the CRP is to improve and standardise international quality control procedures for mass produced fruit flies. There are now over ten fruit fly mass rearing facilities in the world that produce sterile flies for SIT programmes. With international trade in sterile insects becoming a reality, it is important that producers and users apply standard international quality control procedures. A CRP involving behaviourists, physiologists and mass rearing specialists will allow fine-tuning of the internationally accepted standards and procedures as well as developing new tests measuring more representative parameters. Α Consultants Group Meeting International on the Standardization of Quality Control Procedures for Mass Reared and Released Fruit Flies was

held in May 1997 in Vienna. It produced an updated international manual of standard QC procedures (available for downloading from www.iaea.org/ the internet at http:// nafa/ d4/ public/ programmes/ recommended d4_pbl_5_1.html) and implementing this CRP to address those technical issues that require fine-tuning and those that could not be resolved and therefore require a co-ordinated R&D approach to develop new or better OC tests.

The 2nd RCM will be held in Hawaii in April 2001.

Expected duration: 5 years (1999-04)

Contract Holders (12) from Argentina (2), Chile, Costa Rica, Guatemala, Israel, Lebanon, Mexico (2), Peru, Philippines and Portugal.

Agreement Holders (4) from Australia, France, Japan and the United States.

Evaluating the Use of Nuclear Techniques for the Colonisation and Production of Natural Enemies (D4.30.02)

The first Research Co-ordination Meeting, to plan and co-ordinate the research, was held 18 - 22 October 1999 in the IAEA, Vienna, Austria (see page 29).

Nuclear techniques have considerable potential for various uses in biological control. These applications should provide significant benefits to producing biological control agents and for using them to manage pests, facilitate trade, and protect the environment. The First Co-ordination Meeting focused on developing a research plan for the following potential applications of nuclear techniques in biological control:

a) to provide a non-destructive means for pasteurization/ sterilization of artificial diets. Using ionizing radiation to destroy micro-organisms in artificial media provides a viable method to sterilize media without the damaging effects associated with heat treatment, and allows sterilization to be accomplished after diet dispensing and packaging ("terminal sterilization").

- b) to provide non-reproductive supplemental hosts/prey for parasitoids and predator to build-up naturally occurring or augmentatively released natural enemies early in the season when pest populations are low. Non-parasitized hosts would be sterile, even further contributing to suppress the pest population.
- c) to provide sterile pests/hosts as food during commercial shipment of entomophagous insects/mites, thereby assuring quality during transport and that no new pest or pest race is introduced into the regions or countries of customers. Irradiation would also help to fulfill quarantine regulations by avoiding the transport of other hitchhiking pests.
- d) to improve the suitability of natural or factitious hosts/prey for use in parasitoid/predator mass rearing, by helping

for example to overcome host resistance such as encapsulation of parasitoids. Radiation of hosts during mass rearing would also avoid the emergence of fertile adults of the pest, or the need for costly procedures to separate parasitized from non-parasitized insects.

e) to reproductively sterilize exotic beneficial insects that are promising candidates for classical biological control, thus enabling safe field testing of their host or prey specificity on weeds or insect pests. In view that there are many reported cases of natural enemies becoming pests, and the fact that promising natural enemies are eventually not released because doubts persist as to their specificity after detailed assessments under quarantine conditions, safe field testing of specificity is a major use of ionizing radiation not exploited to date.

The 2^{nd} RCM will be held in Guatemala in March 2001.

Expected duration: 5 years (1999 - 04)

Contract Holders (13) from Argentina, Bangladesh, Bulgaria, China, Guatemala, India, Mexico, Pakistan, Poland, Slovak Republic, Syria and Turkey (2).

Agreement Holders (3) from Austria and the United States (2).



TSETSE

Mass Rearing Design Developments

The new tsetse production Unit (TPU2) was evaluated. The Unit holds many cages which can be brought to a blood feeding table together and from which pupae can be collected easily. Experiments were completed in new cages to determine optimal fly densities. The large cages caused aggregations of flies in certain areas leading to high mortality levels. Feeding was excellent and pupal collection efficient. Based on this evaluation some problems in certain components were identified. Cage design is still not optimal and pupal collection procedures can be improved. These changes have been incorporated into a modified design and 3 units have been constructed and will shortly be shipped to laboratories in Africa for field evaluation

New Fly Handling Procedures

Self Stocking of Production Cages (SSPC) This system has been developed to enable flies to be introduced into cages at the right density and sex ratio without the need for handling the flies. From July onwards the system was integrated into the routine maintenance of a large *G. pallidipes* colony. Production in the colony has been assessed using experimental cages and very good results have been obtained. Introduction of the system

Handling of Male Pupae and Adults

The use of the SSPC system to deliver males and females to production cages produces, as a very important by-product, male pupae. Experiments have been carried out to develop new procedures for handling these pupae and the emerging adults. Control and synchronization of male emergence would enable flexible sterilization and release strategies to be developed. Initial experiments

The principle of TPU2 involves the movement of cages containing flies to a stationary blood source. Consideration has now been given to reversing this procedure and assessing if a system can be developed in which the blood is moved to the flies. With outside consultancy a mobile blood feeding table has been designed and built. The constraints on this type of system include the necessity of a maintaining a accurate horizontal movement and the prevention of spillage of blood during movement. A system will be placed in Seibersdorf before the end of the year for evaluation. The proposed design of the large rearing facility in Ethiopia was evaluated.

has required that pupal collections be made 7 days a week. The data from this large scale trial have indicated that providing good pupal incubation conditions can be achieved in a facility then this procedure is suitable for technology transfer to operational tsetse SIT programmes.

This study has been supported by an in depth analysis of the effects of adult age on the mating behaviour of this species.

focused on cooling the mature pupae to different temperatures for different amounts of time and monitoring the effects on several quality parameters of the emerged flies. Initial results are encouraging. Adult male emergence can be manipulated by temperature without affecting some important fitness parameters such as survival and mating. This work has been continued to look at effect of low temperature on the adult males. In a large field programme aerial releases of sterile males will be made using a chilled adult release system. The flies are remarkably resistant to low temperature and can be cooled to 4oC for extended periods without apparent ill effects. *Other R and D News*

• Several new colonies have been established and are flourishing. They include a *G. swynnertoni* colony kindly provided by R. Gooding in Canada, colonization of *G. m. centralis* from a field population in the Ocavanga Delta in Botswana and the establishment of a *G. m. centralis* colony from Ketri, Kenya. • Establishment of a *G. pallidipes* colony from Ethiopia has been problematical due to low survival of flies emerging from the very large number of pupae produced in Arba Minch. The cause of this has now been identified and action taken.

• Hybridisation experiments have been initiated in the *G. morsitans* group of flies. The aim of this work is to see if natural systems of sterility induction could be integrated with radiation induced sterility. Certain crosses conform to previously published results but anomalous results have also been obtained.

• At the recent ISCTRC Meeting in Mombasa, 3 papers were presented covering work carried out in the Unit (see reports starting on page 24)

MEDFLY

Synthesis of a New Genetic Sexing Strain, Vienna 7/Mix-99

A new genetic sexing strain was developed to address some of the problems that have arisen with the previous GSS. Two problems were targetted i.e., improving stability and quality control evaluation. To increase the genetic diversity in this new strain GSS VIENNA 7, the laboratory strain was outcrossed with genetic material from the following seven different populations (origin/year of colonization):

1.) Tunisia (wild, 12/94)
2.) Madeira (wild, 12/96)
3.) Kenya (wild, 1/98)
4.) South Africa (wild, 2/98)
5.) Australia (wild, 8/97)
6.) Reunion (wild, 12/97)
7.) Chile (lab, 93)

The original GSS was constructed in a background originating from genetic а population in Egypt which was colonized in 1983. Material from the field populations was maintained on grapes. Males from each of the seven populations were mated in parallel with Egypt females carrying the *tsl* mutation. Egypt females were used as they carry a specific genetic marker and they are adapted to oviposition in the laboratory. The progeny from these crosses was inbred to produce a homozygous tsl line with the mixed genetic background, Mix-99. The chosen translocation for Vienna 7, (T(Y;5)3-129), was also outcrossed for several generations and then combined with Mix-99 to produce the new strain.

Evaluation of a new Genetic Sexing Strain VIENNA 7-99 Mix

This strain has been constructed using a different translocation to that used in VIENNA 6. The "Mix" in the title refers to the genetic background introduced into the strain and it includes a genetic material from at least 6 different geographic origins. The strain has been evaluated for both its mass rearing profile and its mating competitiveness.

It has been mass reared for 7 generations and shown much better stability than previous strains. The level of recombinants is negligible and the quality control parameters especially that for pupal emergence are very encouraging. The mating competitiveness profile of the strain is comparable with other mass reared bisexual and GS strains.

Provision of GSS to Counterparts.

An important task of the Unit is to provide improved strains to rearing facilities in different parts of the world. This transfer is only done following the type of evaluations described above. Facilities in South Africa and Australia have recently been supplied with the

Other R and D News

• In preparation for developing programmes on fruit flies other than medfly, for which quarantine regulations exist in Europe, the rearing facility is being upgraded to meet regulatory standards.

• In collaboration with colleagues in the US and Greece a detailed molecular and cytological analysis of medfly transgenic lines is being carried out.

VIENNA 7-99Mix for use in their SIT programmes. Staff from these facilities have been trained in the rearing of this strain at Seibersdorf. Colonies for experimental work have been delivered to Crete, Greece and Madeira, Protugal.

• The analysis of inversions is continuing as is the search for the male determining gene. Both these projects are long term and aim at improving genetic sexing methods using both conventional and molecular approaches. At the international meeting on fruit flies of the Western Hemisphere in Guatemala in June, 3 posters were presented of medfly studies carried out in the Unit



5th PAAT Committee Meeting, FAO Rome, 23-23 November 1999

The fifth meeting of the Programme Against African Trypanosomiasis (PAAT) Committee was held in Rome on 22-23 November 1999. The meeting was attended by representatives of International Organisations, donors, scientists and technicians as well as the affected countries in Africa. Great concern was expressed over the increasing incidence of sleeping sickness now affecting many areas of In many foci the disease the continent. situation has returned to the epidemic levels last recorded in the 1930's with over half of local populations infected. At the same time the animal form of the disease severely constrains food production and is now accepted as a major cause of poverty and suffering in many of the poorest rural regions on the continent. In discussing this issue the committee recognised the need to raise the awareness of both African Governments and the international agencies to this deteriorating The Committee urged that the situation. PAAT secretariat explore ways to improve the dissemination of information at all levels in order to secure the appropriate long - term commitments needed to address the problem.

Mr. Leonard T. Budd, an economist contracted by the Department for International Development (DfID) of the UK presented the findings of his general assessment on the benefits accruing from investments into tsetse/trypanosomosis research and control. The results strongly suggest that, in order to effect a substantial economic difference in the tsetse/trypanosomosis affected livestock and agricultural sector of sub-Saharan Africa, large scale (>100,000 km²) control / eradication projects should be implemented. According to the study, the investment of US\$ 20 billion $(US\$ 1,950 \text{ per } \text{km}^2)$ will be needed to eliminate tsetse and the trypanosomosis problem from the African continent. The accruing benefits in the first 20 years will amount to US\$ 50 billion. This is a benefit : cost ratio of 2.6 : 1 over the first 20 years, which will gradually increase to 5.2 : 1 thereafter. In comparison, small scale (1 - 10 km^2), farmer based projects require the investment of US\$ 4,300 - 5,600 US\$ per km², achieving a benefit : cost ratio over a 20 year period of 2.1 - 2.4 : 1, but without further increasing benefits thereafter, because of the need to pursue the control efforts and respective investments.

The intention to transfer the Policy Planning and Implementation (PPI) module of PAAT to OAU/IBAR in Nairobi was reconfirmed and the elaboration of reviewed terms of reference for this and other functional bodies of PAAT was suggested.

25th Meeting of the ISCTRC, Mombasa, Kenya, 27 September -2 October, 1999

At 25th biennual and golden jubilee Meeting of the ISCTRC (International Scientific Council for Trypanosomosis Research and Control) it became apparent that the work of IAEA to assist Member States in fighting the tsetse and trypanosomosis problem is receiving increasing recognition. At the opening session of the ISCTRC the Vice-President of the Republic of Kenya, Hon. George Saitoti, the Assistant Secretary General of OAU, Dr. Hamat Habib Doutoum, and the Director of OAU/IBAR, Dr. Walter Masiga, specifically highlighted the contributions of IAEA over the past years to alleviating the tsetse and trypanosomosis problem.

Several papers presented in the vector control session of the ISCTRC meeting confirmed the increased international acceptance of tsetse SIT: A majority of the papers either highlighted the usefulness of certain research or control components in support of SIT operations or actually were presentations on completed or progressing SIT projects. A highlight was the paper by Khalfan M. Saleh *et. al.* on "Eradication of Glossina austeni from the island of Unguja confirmed: Results of 2 years of post-eradication monitoring activities", in which Mr. Saleh underlined that the tsetse eradication resulted in the elimination of disease transmission. Furthermore, biting flies, for example the widespread stable fly population on the island, failed to maintain the transmission of nagana trypanosomes on Zanzibar, suggesting that the potential of mechanical transmission of these blood parasites is of no epidemiological importance.

There apparently still is an insufficiently clear understanding regarding the meaning/ definition of "area-wide". Many colleagues in the "tsetse/tryps community" confuse it with large scale operations; instead it is a harmonised, concerted approach of total insect population management.

SIT presentation to the CLAM meeting, Valencia, Spain, 13 - 15 October 1999

The Comité de Liaison de L'Agrumiculture Méditerranéenne (CLAM) invited the IAEA to send a representative to Valencia, Spain to promote SIT through a special conference on the "Use of SIT to Control and Eradicate the Mediterranean Fruit Fly Within the Mediterranean Region" sponsored by CLAM from 13-15 October 1999. Pat Gomes represented FAO and IAEA at the meeting.

CLAM is an important professional nongovernmental organisation formed in 1953 that plays a central co-ordinating role among Mediterranean citrus producers in terms of statistical reporting, market promotion, and advising clients on the use of technically sound approaches to production, pest control, etc. CLAM often convenes special meetings to discuss topics that affect the Mediterranean citrus industry as a whole. The close liaison maintained with National authorities in each of the Mediterranean countries and the EU makes CLAM a key group for communicating the advances and benefits of SIT to the industry as a whole. Several resolutions favouring the use of area-wide SIT for Medfly control were passed by the CLAM General Assembly.

The Special Conference took place at the Instituto Valenciano Investigaciones Agrarias (IVIA) on Wednesday, 13 October 1999.

Pat Gomes made a presentation of approximately 1 hour highlighting the positive aspects of area-wide SIT for control and eradication. This presentation highlighted benefits of SIT over conventional pesticide approaches, recent advances and developments in Medfly SIT, present and future applications, and the potential for commercialization of SIT. CLAM members were particularly interested in the cost of applying this technology, so several slides showing the results of the economic analysis conducted by the Imperial College for the proposed area-wide SIT project in the Near East were presented. It was pointed out that building a facility in the region would address one of the major constraints to using SIT for either control or eradication purposes. Control, eradication or establishment of pest free areas depends upon the overall market strategy that will be pursued by the industry. Commercialization of SIT, from production through release, will deliver the technology to clients at a lower overall cost in the future.

Pat Gomes also highlighted the mission findings by experts who recently returned from Egypt where they were asked to evaluate the Peach fruit fly, Bactrocera zonata (Saunders), a new fruit fly pest recently introduced into CLAM members were surprised to Egypt. learn of its presence and the threat it poses to neighbouring countries within the Mediterranean, particularly to citrus producers. Copies of the pest biology, description, list of hosts, and an action plan for detection and eradication of this pest developed by the US Department of Agriculture were distributed.

Jonathan Knight, Imperial College of Science, Technology and Medicine, Centre for Environmental Technology described the benefits of using SIT for control purposes versus eradication and presented various options for funding SIT. Brian Barnes made a presentation on the SIT project underway in the Hex River Valley in South Africa aimed at control of the Medfly. Rui Pereira provided the group with an update on Medfly control activities in Madeira while Antonio Guerreiro described the aims and prospects for implementing area-wide control using SIT in the Algarve region of Portugal.

Additional presentations were made by a representative for the Ministry of Agriculture for the Valencia Community, Sr. Jose Moner, and Dr. J. Pedro Ros, a research entomologist with the Instituto Nacional de Investigaciones Agrícolas. Sr. Moner described the current control measures for Medfly in Spain that depend entirely upon the application of malathion bait sprays applied from the air by the local governments and ground sprays by the farmers. He also reported on some laboratory studies using lufenuron, an insect growth regulator that inhibits the production of chitin in insects and other arthropods. No field trials have been conducted yet. Lufenuron is a Novartis product that is sold under the brand name Program ® or Match ®. It acts as an ovicide and larvacide. Dr. Ros reported on studies using mass-trapping for Medfly control. They currently place from 50 to 80 traps per hectare in cherimoya orchards baited with the female attractant that was developed jointly by IAEA and USDA under a CRP (IAEA TEC-DOC-1099, available from the Section or via our web pages, see page 24).

Mr. Len Beans made a presentation describing the aims and interests of Slovakia in possibly building a commercial facility in the future to produce and sell sterile Medflies, tsetse and codling moth to interested clients. Mr. Beans recently conducted an expert mission on behalf of the IAEA to Slovakia to assist them with their feasibility study (see page 8).

During the official dinner of the CLAM Assembly sponsored General by the d'Agricultura, Conselleria Peixca V Alimentasio de la Generalitat Valenciana (Regional Council of Agriculture, Fisheries, and Food of the Valencia Community), Ms. Regional Maria Angeles Ramón-Llin. Minister, gave a speech in which she endorsed the need for additional biofactories within the Mediterranean to address the needs of CLAM members and expressed interest in having one constructed in Valencia. She also made a call for co-ordinating research efforts among CLAM countries to seek better methods of Medfly control in the future. Mena Davidson, Chairperson for the Agro-Technical Committee of CLAM and General Manager for the Citrus Marketing Board of Israel, expressed his keen interest in pursuing SIT for control purposes rather than eradication in Israel. If he can achieve similar control with SIT as with bait sprays, then he is willing to implement a larger scale "experiment" using SIT. He is willing to accept some additional damage, but did not say exactly how much would be tolerated by the industry. He wants to demonstrate that effective control can be obtained in the coming months.

Conclusions and Recommendations

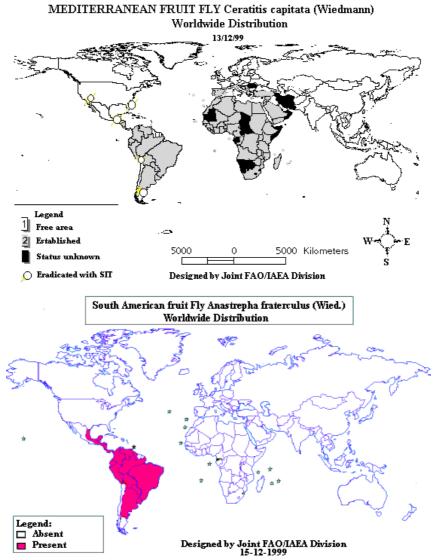
- CLAM agreed to conduct an industry-wide survey to assess the economic impact of the Medfly. This would include identifying existing control measures and costs, type and quantity of pesticide applied, percent fruit infestation or damage caused by fruit flies, etc. CLAM will seek technical input and advise on developing the questionnaire to be completed by the **CLAM** representatives in each of the countries.
- The CLAM members endorsed the use of area-wide control of Medfly using SIT as a viable pest control approach for achieving effective suppression.
- There is support amongst CLAM members for the establishment of biofactories for the production and distribution of sterile insects.
 - The representative from Italy expressed interest to have a facility located in Sicily.
 - The Minister of Agriculture for the Valencia Community in Spain expressed interest to have a facility located in Valencia.
- Various CLAM members expressed their interest to use area-wide SIT for control of the Medfly in the future.
- CLAM members were alerted to the presence of the Peach fruit fly, *Bactrocera*

zonata, in Egypt and encouraged to strengthen their exclusion and detection measures for this pest.

- CLAM members expressed their concern about the presence of the Peach Fruit Fly in Egypt, and called for detection efforts throughout the Mediterranean.
- One way of gaining grower and consumer acceptance of SIT would be through brand name association where it is an integral part of an **integrated production** system. As the SIT is an environmentally-friendly technology for pest control, growers would receive credit for using it for pest control.

Section adopts GPPIS pages for medfly and Anastrepha fraterculus

FAO created the first electronic, interactive, multimedia compendium of plant protection information in 1987. The current Global Plant & Pest Information System (GPPIS), is an Internet/WWW based version of FAO's earlier work. GPPIS is a global implementation and extension of the Pacific Plant Protection Information System (PPPIS) developed by FAO and the South Pacific Commission (SPC). GPPIS replaces PPPIS and fulfills FAO's constitutional mandate in the area of nutrition, food and agriculture.



GPPIS is a dynamic framework for collective knowledge processing that can be

implemented in a variety of cross-platform environments on the Internet, in local Intranets

and as a stand-alone database on CD-ROM available free of charge from FAO.

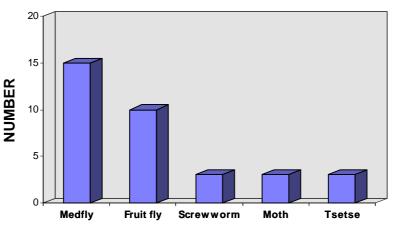
GPPIS data and source code are in the public domain because they are created and maintained by the community of individuals who choose to participate in the evolution of GPPIS. The Insect and Pest Control Section of the Joint FAO/IAEA Division has adopted and maintains two insect species, Medfly *Ceratitis capitata* and the South American Fruit Fly *Anastrepha fraterculus*. Data include Taxonomy/ Description, Management, Ecology, Dispersion and an updated World Distribution Map.

The GPPIS database is currently available at http://pppis.fao.org/. The site is being moved to the Ecoport, hosted by the University of Floida jointly with the FAO and the Smithsonian Institute. Public access to Ecoport will be available from February at http://www.ecoport.org/

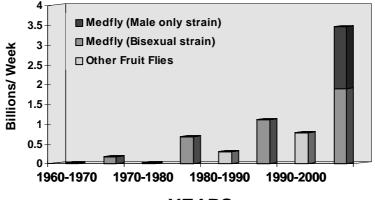
Directory of Mass Production Facilities of Sterile Insects

A Directory of Mass Production Facilities of Sterile Insect was developed by the Insect and Pest Control Section. The Directory contains information regarding locality of the facilities in the world, the starting date of operation, the strain produced, the production capacity and the dose of radiation for sterilization, F1-sterility in case of Lepidoptera and larval sterilization for parasitoid mass production.





WORLDWIDE PRODUCTION CAPACITY OF STERILE FRUIT FLIES



YEARS

The data is compiled in 7 tables: medfly, other fruit flies, Moths, Tsetse and Screwworm, parasitoids and medfly strains in Seibersdorf. The directory includes references regarding the project leader and complete contact address.

This directory is an attempt to provide all the necessary information regarding the past, present and future SIT project facilities. The directory is available on request from the Section, and will be placed on the section's web site shortly.

Screwworm officially declared eradicated from Nicaragua

In a meeting of regional co-ordinators held in Managua, Nicaragua in October 1999, New World Screwworm was officially declared eradicated from Nicaragua. Eradication is also well under way in Cost Rica and Panama, which together with Cuba, Jamaica and Hispaniola are the only countries north of the Darian Gap still infested with screwworm.

IAEA-TECDOC-1099 available "Development of Female Medfly Attractant Systems for Trapping and Sterility Assessment"

The proceedings of the final Research Coordination Meeting of the CRP "Development of Female Medfly Attractant Systems for Trapping and Sterility Assessment" is now available as a TECDOC. Copies are available from the Insect & Pest Control Section, from our web site in .pdf format. For details of how to obtain the document in electronic format or on microfilm contact the INIS clearinghouse (see the INIS web page at http:// www.iaea.org/ programmes/ inis/ inis.htm or e-mail chouse@iaea.org).

Effect of low temperature treatment on Glossina pallidipes pupae

The success of insect control programmes integrating the sterile insect technique (SIT) depends on the ability of the sterile males to seek out wild females and compete with wild males for mating opportunities. Since only male insects require to be released, sex separation at some stage of development can be very important. Formerly, sex separation for tsetse was only possible in the adult stage where flies are immobilised by chilling and individually sexed. With the development of an automated sexing technique based on differences in emergence pattern of females and males and the direct stocking of fly production cages for Glossina austeni and G. pallidipes, it is now possible to obtain pupae which are better than 99% male.

With some species of tsetse e.g. *G. pallidipes*, it is important that males attain sexual maturity before they are released and therefore a planned and synchronised emergence of males would be beneficial. A

protocol for controlled emergence based on low temperature treatment of mature male pupae is under development.

A majority of mature male pupae, obtained after females and a proportion of males have been introduced into production cages, were incubated at different temperatures between 15-20°C for 24-72 hours and thereafter allowed to emerge at either 23-24°C or 26.5°C. Emergence rate, adult male survival without blood (stress test) and male mating behaviour were monitored.

There was no significant difference due to low temperature treatment on emergence rate (F = 0.02, df = 7,74, P > 0.05), survival without blood and insemination capacity. Low temperature treatment delayed eclosion of *G*. *pallidipes* by 2-3 days and this has potential for use in operational SIT programmes.

(Paper presented at the 25th ISCTRC Meeting, Mombasa, Kenya by Opiyo. E., Mutika G. and Robinson, A.)

Initial studies on RAPD polymorphism in Glossina fuscipes fuscipes

In order to successfully implement the Sterile Insect Technique (SIT) for the eradication of tsetse populations, it is essential to determine the degree of isolation of the target population. A population of *G. f. fuscipes* on Buvuma Islands might be targeted for eradication and it is important to know how related this population is to the populations of the Uganda mainland. This study makes use of RAPD primers to analyse the genome of *G. f. fuscipes* populations. 100 RAPD oligonucleotide primers (10 mers each) have been tested for their ability to amplify *G. f. fuscipes* DNA. The primers which amplified the DNA were screened for their sex specificity and polymorphic nature by testing each primer on a set of 16 DNA samples comprising 8 males and 8 females. One male specific primer (no. 77) with the sequence GAG GAC GAG and 7 polymorphic primers with sequences CCT GGG CCT A (no.6), CCT GGC GGT A (no. 8), CCC GCC TTC C (no. 23), TTA AGG

GGG C (no. 46), TTG GCC GAG C (no. 60), AGG GGC GGG A (no. 65) and GAG CAG GGG G (no. 81) have been selected and used in preliminary comparisons of DNA of *G. f. fuscipes* from the Seibersdorf colony and two populations from Uganda. The male and species specificity of primer 77 has been confirmed by testing it with *G. brevipalpis*, *G. pallidipes*, *G. austeni*, *G. morsitans morsitans*, *G. palpalis palpalis* and *G. tachinoides*.

(Paper presented at the 25th ISCTRC Meeting, Mombasa, Kenya by A.M. Gidudu, A. S Robinson, A. Stamenova)

Glossina pallidipes Austen (Diptera: Glossinidae). Effects of Age at mating on receptivity, on mating duration, insemination and fecundity.

Glossina pallidipes is an important vector of trypanosomosis in the East Africa region. Major constraints to SIT application to eradicate the species are lack of knowledge of the mating behaviour and labour intensive rearing. Experiments on some of these aspects were carried out at the tsetse mass rearing facilities of the FAO/IAEA laboratories, Seibersdorf, Austria. They are part of a series of tests aimed at improving methods to optimise the mass rearing technique and also to provide high quality males for SIT release programs. We studied reproductive parameters considered useful for successful colonisation and SIT targeting G. pallidipes. The optimum age of mating G. pallidipes in the laboratory is 9-13 days when they are highly receptive. Mating duration does not depend on age and receptivity of the females. Females mated with 7-15 days old males showed no difference in insemination rate an indication that age of males after day 7 has no effect on the amount of sperm transferred. There was no difference in pupal production when females aged 6-12 days were mated in different age combinations with males aged 6-12 days. No difference was found in pupal production between these groups and the group that was mated at emergence without separation. The optimum age at mating males is days 11-13 which is the peak of sexual activity. The age of males does not affect the duration of mating for males older than 5 days. Colonisation of G. pallidipes can be successful accomplished by mating females when they are 8-13 days old. Though males from day 7 and above can successfully inseminate females, male ages 11-13 can be used when high numbers of pairings are required within a short duration. There is no need to age flies before mating as long as males and females are left together in the same cage where several matings can take place. Though males are at peak activity from 11-13 days, it would be advisable to release males when they are 9-10 days old so that they have the highest chance of finding a wild female. The optimum age of mating females and males should be standardized for different G. *pallidipes* populations.

(Paper presented at the 25th ISCTRC Meeting, Mombasa, Kenya by P.A. Olet, A. S. Robinson, E. Opiyo.)

Co-operative Medfly Preventive Release Program in the Los Angeles Basin

The Co-operative Medfly Preventive Release Program (PRP) in the Los Angeles Basin of southern California is in its third year of operation. The program, funded jointly by the United States Department of Agriculture and the State of California Department of Food and Agriculture, is releasing sterile medflies year round as a tool to prevent the establishment of Medfly in California.

The PRP uses chilled adult release technology to release the sterile medflies from aircraft. Close to 300,000,000 sterile medflies

are released every week over the 2,155 square mile program area. The program releases throughout the entire area twice each week at three to four day intervals, in order to ensure a well distributed population of healthy sterile flies. The combined weekly release rate is a minimum of 125,000 per square mile per week, based on the bisexual strains.

a SIT During eradication campaign completed in the past months for medfly and Mexfly in the San Diego area, the program experimented with combined release flights. Both species of sterile fruit flies were carried on the same flight by loading a release container of medfly on top of the Mexfly release container. The "double box" system allowed for the aircraft to start releasing medfly at the end of the Mexfly release simply by pulling the slide at the bottom of the medfly container without having to make a fortyminute return flight to change release containers. In the future it is planned to have two release machines in the plane, so that the release of more than one species can be achieved simultaneously as required, or in the case of high density release rates, twice the number of sterile flies can be carried on a flight.

At the present time, 85% of the sterile flies released are from bisexual strains, and 15% are from the TSL strain. The PRP receives sterilized bisexual pupae from the USDA Rearing Facility in Waimamalo, Hawaii, (300 million per week. Maui-93 strain) and the USDA International Service's El Pino Production Facility in El Pino, Guatemala (120 million per week, Antigua strain). Sterile TSL pupae (49 million per week, Toliman TSL strain) are also received from the production facility in El Pino, Guatemala. The California Agriculture's Department of Food and production facility in Waimanalo, Hawaii began an extensive renovation plan in September 1999. Following completion of the renovations, the facility will begin rearing a TSL colony.

A trapping network of Jackson traps baited with Trimedlure and McPhail traps baited with torula yeast is operational year-round for the detection of Medfly within California. Approximately 7,000 Jackson and 7,000 McPhail traps are located within the preventive release program area. Trap density is 5 per square mile of each trap type. The traps are inspected biweekly and weekly, respectively. All specimens are collected and submitted to the Identification workforce, where the specimens are screened under UV light for dye. The Identification unit screens over 350,000 specimen each week. If dye is not visible, an Insect Biosystematist will perform a dissection on the specimen to determine sterility.

In the past two years, there have been only three separate trap collections, each with a single wild medfly, within the preventive release area. In November 1998, a single wild female was trapped in a McPhail trap in Santa Ana. In April 1999, a single wild female was trapped in a McPhail trap in El Monte, over twenty miles north of Santa Ana. The third collection occurred in August 1999, when a wild male was trapped in a McPhail trap in South Gate, ten miles west of El Monte and twenty miles northwest of Santa Ana. Intensified delimitation surveys, including trapping and fruit sampling, were conducted around each fly find. The delimitation surveys have resulted in no further fly finds and no larval discoveries in all three locations.

The PRP is posting results from the trapping program and the Quality Control test performed on each shipment of sterile pupae sent to the project on the Internet. The address is http://pi.cdfa.ca.gov/prp. Trap recovery results are updated every two weeks. Quality control reports are updated weekly. The PRP is also developing a QC database that will be accessible through the Internet. An Internet accessed database will allow for remote data entry from production facilities and end users in a standardized format. Real-time updates will be just one of the many benefits for production facility directors and project managers.

The following data reflects some of the many things the staff of the PRP has accomplished since January 1999.

- Released over 14 billion sterile flies
- Cooked over 95,000 gallons of agar for the adult diet

- Flown over 575,000 linear miles during 2,500 flight missions
- UV light screening of 20 million trapped sterile flies
- Provided training and/or tours to visitors from 9 countries

The program has recently been reviewed in California Agriculture (DOWELL, R.V., SIDDIQUI, I. A., MEYER, F., SPAUGY, E.L., Early results suggest sterile flies may protect S. California from medfly, California Agriculture **53** 2 (1999) 28-32).

Review of Area-wide Control of the Codling Moth in Apples and Pears in the Western USA

A 5-year project in the states of Washington, Oregon and California tested the efficacy of **area-wide pheromone-based insect pest management (IPM) systems**. The project was sponsored by the USDA-ARS, and directed by Dr. Carrol Calkins. A final review of the project was held on 18 and 19 November 1999.

The codling moth is the key pest of apple and pear crops. The project was a successful demonstration of codling moth control using sex pheromone mating disruption (MD) and a organophosphate (OP) insecticide few applications, usually only one per year. The main achievement was to reduce the number of insecticide treatments for the codling moth from 3 or more to 1 or sometimes none. A few insecticide treatments are still needed since mating disruption does not work well under all circumstances and in all locations (uneven terrain, windy area, on orchard borders, very small orchard blocks and infested sites located nearby). Growers are assisted in appropriate timing of sprays by consulting a web page on the Internet that provides information about spray timing and degree-day models.

A total of 3,000 acres of apples and pears in 5 sites was included in the first year of the project, and this increased to 21,000 acres (and 466 growers) in 1999, the last year of the project. Grower acceptance of mating disruption was often slow since growers were reluctant to risk using a new technology, but most participating orchardists now say that they will continue to use mating disruption as the primary method of codling moth control even though the subsidy provided by the project has ended. The growers want to use biological control techniques rather than insecticides. The growers have realized that they benefit from co-operating together in an

areawide approach to pest control rather than each orchardist making decisions independent of what neighbours are doing. Also labour efficiency has improved since orchard workers can carry out normal operations all of the time in MD orchards, but in OP-sprayed orchards there is a 14-day waiting period after each spray during which, for safety reasons, no one should enter the orchard. A new regulation in the USA, the Food Quality Protection Act, placed increased emphasis on avoiding 'hard' pesticides, and this encouraged growers to participate in the project. Also there was a real threat from insecticide resistance developing in the codling moth populations.

The use of MD is expanding each year, especially in pear orchards where codling moth control with MD seems to be especially In 1999 in Washington State, appropriate. including non-project sites, MD was applied in a total of 60,000 acres This is expected to increase in the future. The cost of using MD decreased from US\$135/acre in 1995 (\$120 for materials, \$15 for application) to \$105/acre in 1999. Usually this is still higher than the cost of the standard OP treatment programme for the codling moth, and applying the MD release devices is labour intensive, but the comparison gives a different result if the whole IPM system is considered. At first the cost of pheromone-based IPM was higher than using insecticide-based IPM, but because of fewer insect problems resulting from greatly reduced OP treatments, especially in pears (75% reduction), the total cost of protection against insect pests using MD technology is now lower than when using insecticide technology. When the codling moth population density is reduced to a low level. MD is more effective. and also the rate of MD chemical used can be reduced, for example, to half the rate. The

economics of pest control is critical in the orchard industry since currently fruit production is often not economically viable.

Secondary insect pests (especially leafrollers) appear, in some cases, to increase in number and cause more damage when the number of OP insecticide treatments is reduced, and research to find biological methods of controlling these secondary pests is research underway at several centers. Currently most secondary pests are controlled 'soft' materials, such as Bacillus with thuringiensis, insect growth regulators (IGRs) and oils, or chemicals are applied prior to fruit formation. Additional control methods under research for leafrollers are MD and 'attract and Fruit production, from petal-fall to kill'. harvest, is now largely an organic (non chemical) operation, and this might enable growers to sell their produce at a higher price due to the value-added quality of 'eco-fruit'. Already there are environmental benefits and increased flexibility in pest control options due to a reduction in 'hard' pesticide applications. However biologically based control systems increase the risk that quarantine pests, for which there is virtually no tolerance, will reduce export potential. Pest monitoring is essential in IPM systems where routine insecticide treatments are replaced with 'need based' treatments, and monitoring costs money.

The northern-most project site was located adjacent to the Washington/Canadian border. Some of the orchards at this site (total of 480 acres) used BOTH mating disruption and the Sterile Insect Technique (SIT), and achieved extremely good control of the codling moth; no applications of OP insecticides were made in these orchards during the past 3 years.

New technologies using sex pheromones were announced by representatives of commercial companies. MD must be used areawide, is a rather 'fragile' technology, and its effectiveness needs to be improved.

A. Programmable "Puffer" machine (aerosol dispenser) produces bursts of pheromone only at the time of day when it is needed. Two machines are placed in each acre of orchard.

- B. Sprayable micro capsules. This type of formulation, first tested some years ago, is now being evaluated again.
- C. "Last Call CM" is an 'attract and kill' or male annihilation technology that involves placing (by hand) drops of a mixture of pheromone, insecticide and a UV absorbent on to tree trunks. This product will be commercially available next year. The cost is US\$45/acre per application, and several applications per year are needed.
- D. Improved lures in traps: A new lure technology (bubble cap) produces a constant release rate and lasts 60 days, while the standard rubber septa have a declining release rate. Another company sells lures that last 70 days. A third company is adding a wax matrix to the lures to increase the duration of attractiveness.

Research into alternative chemicals to control the codling moth indicates that IGRs could replace OP insecticides. After many years of testing, IGRs for the codling moth are expected to be available commercially next year.

A report on the Canadian programme to control/eradicate the codling moth in the Okanagan region of British Columbia was presented. Control in the southern portion of the programme area was the best since the first sterile moths were released in 1994, with most commercial orchards evidently undamaged by the codling moth. The use of OP insecticides (primarily azinphosmethyl) for codling moth control has been reduced by one half from what it used to be. The pest population is also decreasing in urban areas. The programme is now aiming at areawide codling moth control, not necessarily eradication, in orchards using an SIT-based IPM strategy. Besides the SIT, MD, insecticides and tree-banding to destroy mature larvae are used in the IPM system. Economic problems in the fruit industry and difficulties in obtaining financial support (presently largely derived from grower and community taxation) for the programme have resulted in an uncertainty about future use of the SIT.

Evaluating the use of nuclear techniques for the colonization and production of natural enemies of agricultural insect pests

First RCM, Vienna, Austria, 18 - 22 October 1999

Participants involved in research related to this CRP reviewed all potential applications of nuclear techniques in biological control and agreed to focus their research on the following ones:

A. REARING

1. <u>Artificial Diet Sterilization: Use, Storage</u> and Table Life Extension

Gamma radiation, as well as X-rays, can provide a non-destructive means of killing microbial contaminants in artificial diets that may degrade the diet and/or impair insect growth and development.

2. Suppression of Host Immune Reactions

Exposure to radiation has been shown to suppress host immune system responses. This may make irradiated larvae of older instars more suitable for parasitoid development and thus increase rearing efficiency and parasitoid quality.

3. Storage Extension of Hosts or Prey

Use of radiation may be used to arrest insect development and thus facilitate the development of procedures that would allow for storage and stockpiling of hosts or prey.

4. Host Suitability Extension

Use of radiation may be used to delay normal insect development and thus extend the time interval when a particular host stage is available for use by the parasitoid or help to regulate (slow-down or synchronize) parasitoid development within the irradiated host.

5. Use of Sub-Products of SIT Mass-Rearing

Sub-products of insect mass-rearing programs (e.g., excess or "off" season production or products that do not meet minimum quality standards) may be irradiated and used to support the production of natural enemies and thus improve the overall (cost) efficiency of a mass-rearing (SIT) system.

6. <u>Stimulation Effects of Very Low Doses of</u> <u>Radiation to Natural Enemies</u>

Very low doses of radiation may stimulate a variety of physiological (e.g., pesticide

tolerance) and behavioural (e.g., increased longevity and searching ability) processes in insects that may be beneficial. As such, very low doses of radiation may be useful in improving field performance of laboratoryreared parasitoids/predators.

7. <u>Use of Radiation as a Tool to Study Host-</u> <u>Natural Enemy Interactions</u>

Use of nuclear techniques may be used to selectively modify certain physiological processes in the host (e.g., host "odours") thereby facilitating the study of particular hostparasitoid interactions. Nuclear techniques may be also used to modify or terminate certain parasitoid processes that affect host physiology and behaviour (e.g., sterilizing the parasitoid egg).

B. HANDLING-SHIPMENT-RELEASE AND TRADE

1. <u>Reproductive Sterilization of Host /</u> <u>Factitious Hosts/ Prey</u>

Radiation can be used to reproductively sterilize hosts, factitious host or prey, thereby inhibiting further development and preventing the emergence of unused individuals. This application of nuclear techniques would:

a) allow for the earlier shipping of hosts together with natural enemies without the need to wait for emergence unused hosts;

b) reduce the handling procedures required during rearing of natural enemies, thereby increasing the cost effectiveness of the rearing process and the quality of the natural enemy product;

c) facilitate the preservation of purity of host, prey and/or natural enemy strains;

d) provide a cleaner product for customers purchasing/using natural enemies produced in this fashion.

3. <u>Provisioning Natural Enemy Shipments</u> with Sterilized Artificial Diets

Radiation can be used to preserve the quality of the diet by delaying the process of diet

degradation and, thereby, extending the acceptability and suitability of the artificial diet to the natural enemies. In this way, the system also becomes more cost effective, as diets have longer table-life.

4. <u>Provisioning Natural Enemy Shipments</u> with Sterilized Host/Prey

Use of radiation has the potential of:

a) killing or reproductively sterilizing host/prey to provide the required quarantine security to overcome regulatory barriers, and thereby facilitating and encouraging national and international trade;

b) reducing the risk of inadvertently shipping hitch-hiking arthropods with host/prey;

c) extending the period of suitability of host or prey (as food) during shipping;

d) allowing the addition of safe, nutritional supplements (in the form of host/prey) to shipments of natural enemies that will improve/maintain their quality;

e) allowing more timely delivery of natural enemies by eliminating the time required to allow nonparasitized hosts to emerge, and eliminating the need to separate emerged adult host from parasitized hosts

f) allowing the customers more flexibility in release timing of natural enemies

5. <u>Shipping of Sterilized Pests/Factitious</u> <u>Hosts or Prey (in the Absence of Natural</u> <u>Enemies):</u>

The use of radiation will allow for the commercial shipment of sterilized host individuals, from one laboratory or insectary to another, both within and between countries, Thus, commercial laboratories will be able to rear the same strain of natural enemy using the same host material, insuring a standarized quality of the product for the customer.

C. SIT/ F1 STERILITY + BIOLOGICAL CONTROL

<u>1. Combination of Augmentative Releases :</u> <u>SIT/F1+ Natural Enemies</u>

Nuclear techniques (SIT/F1 sterility) and augmentative releases of natural enemies

(parasitoids, predators, nematodes, and insect pathogens) can significantly reduce insect pests populations. Combining these tactics can yield both additive and synergistic effects. These combined augmentative releases would be compatible with traditional IPM programs which could include resistant plant varieties, biopesticides, cultural practicies, and mating disruption.

2. Supplement Hosts for Natural Enemies Prior to Pest Population Outbreak

An increase in the number of host insects available for natural enemies can increase the population density of natural enemies before a cyclic pest outbreak begins. In this way, an optimum level of natural enemies can be available to prevent expected pest outbreaks. Nuclear technique can be used to produce sterile host insects or host insects with inherited sterility (F1). Releasing sterile insects as hosts for the natural enemies can increase the number of host insects available for natural enemies without increasing the risk that the released insect pest will cause economic damage in the future.

Detail objectives:

a. Suitability of irradiated host for parasitoids [lab and field tests]

b. Optimum methods for releasing the sterile hosts

c. Optimum methods for measuring the effects the natural enemies following the releases of sterile hosts.

<u>3. Supplemental Hosts for Seasonal</u> <u>Maintenance of Natural Enemies</u>

The use of SIT or F1 sterility could provide supplemental hosts in the form of sterile eggs, larvae, and pupae. These supplemental hosts could sustain higher population levels of natural enemies so that future pest population increases would be moderated. An example of this application would be the release of an egg parasitoid during a SIT program so that sterile eggs deposited by irradiated insects could be used by the parasitoid.

4. SIT Against Natural Enemy Pest

In this relationship, the natural enemies could be sterilized with nuclear techniques and released in an SIT program to reduce the detrimental effects of the natural enemies on the useful insect.

D. FACILITATION OF CLASSICAL BIOLOGICAL CONTROL

1. <u>Use of Reproductively Inactivated Agents</u> for Final Confirmation of Host Specificity of Potential Biological Control Agents

Radiation may be used to reproductively inactivate natural enemies so that they can be released and studied under actual field conditions without the risk of establishing breeding populations. The use of reproductively inactivated forms would allow one to further assess and confirm oviposition behaviours and host (acceptability) associations. The use of F1 sterile larvae of herbivores being considered for release against plant pests also would allow one to field test larval feeding preferences and the ability of these larvae to develop and survive on related plants that are of concern.

2. <u>Use of Sterilized Hosts for Exploration of</u> <u>New Natural Enemies and for Monitoring</u> <u>Natural Enemy Field Populations</u>

Reproductively inactivated host insects may be placed in the field in strategic locations as sentinels to aid in the exploration and collection of new natural enemies. These sentinels may also be used to monitor natural enemy populations. Furthermore, the use of live but reproductively inactivated hosts will eliminate the risk of increasing pest populations.

Medfly mating behaviour studies under field cage conditions

Final RCM held in Antigua, Guatemala, June 29-July 3 1999.

The final RCM of this CRP evaluated the project. Several findings and protocols developed during the implementation of this CRP have now been incorporated into operational projects and the internationally recognized "A Manual of Quality Control for Fruit Flies" and are now a standard component of QC assessment of mass reared medflies. Further updates to the manual were also discussed.

Sessions held at the end of the meeting to assess the CRP confirmed its impact on the implementation of medfly SIT programmes and on directing future research needs to be approached during the upcoming CRP on fruit fly quality control.

It was decided that the results of CRP will be published as a special issue in the Florida Entomologist, and a list of papers was approved and a timetable for submission and peer-review agreed on. The final paper in the issue will be an overview of the CRP and its relevance to the implementation of SIT and will be co-authored by Alan Robinson, J.P. Cayol and Jorge Hendrichs. Below are the major conclusions and recommendations:

1. Courtship behaviour Conclusions

- Wild and mass reared flies from different origins exhibit similar courtship behaviour patterns.
- A high variability is noticed in the quantitative aspect of the behaviour of mass reared and wild flies.
- Mechanisms determining female acceptance of a mate are complex and not yet fully understood.
- Mass rearing conditions do affect the quantitative aspects of the courtship behaviour.
- Irradiation has a negative effect on the behaviour of mass-reared males.

Recommendations

• Mass rearing protocols should be considered carefully in order to avoid selection of unsuitable behavioural traits.

- If any mass reared strain shows strong reduction of sexual competitiveness with one or several wild populations, then the strain needs to be deeply analysed to identify potential keys of mating failure.
- The effects of irradiation on the details of the behaviour of mass-reared males needs to be measured on a regular basis.
- Although the differences found in analysis of video-recording could not yet be directly related with differences in male competitiveness, mass rearing strains should be periodically checked to determine if the mass rearing conditions affect their courtship pattern.
- Potential cues for wild female acceptance should be studied experimentally.

2. Lekking behaviour Conclusions

- Compared to other stages in the reproductive sequence of the medfly, leks are poorly understood.
- Lek behavior is common to all medfly populations studied to date.
- Mass reared, sterile males appear to have maintained the ability to find, join and participate in leks.
- Leks appear at 2 different scales: the large scale, which focuses on the individual tree and the small scale, which analyses leks at the microhabitat level within a tree canopy.
- Of the mechanisms driving and regulating lek behaviour, predation and female preference appear to be of primary importance.
- Small laboratory cages are unsuitable for studying any aspect of lek behaviour.

Recommendations

- Large scale spatial studies should be carried out in view of optimising sterile male release.
- Studies of lek dynamics should be undertaken focusing on:

- \checkmark chemical ecology
- ✓ predation
- ✓ microclimate
- ✓ energetics
- ✓ factors affecting variations in individual behaviour, such as size, nutrition, age and experience.
- The importance of alternative mating tactics in the medfly mating system should be addressed.
- In general, the impact of predation on the SIT should be quantified.
- •

3. Comparative approaches of sexual behaviour

Conclusions

- Courtship behaviour and morphology studies in related Tephritid species have provided valuable information to understand medfly courtship behaviour.
- The phylogenetic relationships of members of the Ceratitini tribe is poorly understood at the present time.
- Physical, especially visual, properties of medfly courtship behaviour have been recorded in detail, yet the chemical and acoustic properties of such behaviour are poorly understood.
- Basic components and structure of medfly courtship behaviour are similar in all geographic strains studied to date.

Recommendations

- Compare aspects of courtship behaviour and morphology, primarily in closely related members of the Ceratitini tribe, especially *C. rosa*, and also in other more distantly related Tephritids.
- Expand intensive studies of chemical aspects, especially pheromones, of medfly courtship behaviour.
- Continue comparative studies of courtship behaviour in medfly populations worldwide, emphasizing areas of present or future SIT programmes.

4. Morphometrical studies Conclusions

- Multiple measures of body size are more reliable than single measures to determine levels of mating compatibility and competitiveness.
- There are contradictory data on the importance of male body size on mating success.
- The absence or abnormalities of SFO bristles can have a negative effect on male mating success.
- Occasionally mass-reared males have been observed to lack SFO bristles.

Recommendations

- Studies in which the effects of male body size are checked should include multiple measurements of body size.
- Body size should not be used as an indicator of male quality unless further studies demonstrate its relevance.
- Mass-reared strains should be checked periodically for the lack of SFO bristles.
- Measurements to be used in studies of body size need to be standardised.
- The allometric scaling of body traits should be studied to clarify their relative importance in mating success.

5. Nutritional aspects in relation to sexual behaviour Conclusions

- Studies on various fruit flies indicate that ingestion of specific precursors affects
- pheromone production and male reproductive success.
- Post teneral nutrition enhances reproductive success of male medflies by affecting:
 - \checkmark the ability to emit pheromone in leks,
 - \checkmark copulatory success,
 - \checkmark the renewal of female receptivity.

Recommendations

- The feasibility of providing sterile males with a protein meal prior to release should be considered.
- The relationship between larval and post teneral diet on mating competitiveness and survival of irradiated males should be studied.
- Studies should be continued to reveal the effects of larval nutrition (both larval hosts and artificial diets) on teneral nutritional reserves.
- The relative importance of natural food sources (e.g., bacteria, fruit juices and faeces), for male reproductive success should be established.
- The feeding behaviour of irradiated males needs to be studied.
- The nutritional status of sterile males used in competitiveness studies must be standardized to conform with the status of males released in SIT operations.

6. Remating

Conclusions

- Contrary to previous beliefs, remating in wild medfly females is common in nature.
- Sterile males are less able than wild males to suppress remating in wild females. This effect is increased in males that have been colonised for a longer time.
- Mass reared males have reduced copulation times when compared with wild males. These reduced copulation times appear to be associated with increased female tendency to remate.
- There is a strong evidence that male accessory gland fluids influence female receptivity and olfactory behaviour. However, the mechanisms and variables involved are not well understood.

Recommendations

• A remating component with a standard protocol should be added to field cage quality control tests.

- The significance of differential remating inhibition induced by wild and mass reared irradiated males should be ascertained for SIT programmes.
- Further studies should be carried out to determine the mechanisms and variables which influence or modulate the change from mating to oviposition behaviour in females.
- Studies on the characterisation, biosynthesis and biological activity of accessory gland fluids should be carried out.
- Both male and female effects on the amounts of sperm and accessory gland fluid transfer in one or more matings by the same male should be studied.
- The costs and benefits of female remating should be studied both under laboratory conditions and in the field.
- Operational factors such as
 - ✓ Shortened male sexual maturation period,
 - ✓ Increased survivorship of sterile males,
 - ✓ Delayed release of flies,

which influence sterile to wild ratios should be studied to compensate for any detrimental effect of remating.

7. Compatibility and isolation studies Conclusions

- Wild strains from different geographic origins show a high degree of compatibility.
- Mass reared strains originating from any wild population can potentially be used worldwide.
- There is evidence that some mass reared strains of medfly exhibit a degree of incompatibility when tested with different wild populations.
- Regular field cage monitoring of mass reared strains is required to assess the compatibility with the local target population.

• In case of incompatibility, the role of visual, sexual pheromone and acoustic signaling is unknown.

Recommendations

- Experimental evidence should be gathered to verify the possible existence of genetic basis for behavioural resistance.
- Field cage evaluation must be a regular component of strain evaluation for field operations.
- The role of visual, sexual pheromone and acoustic signaling in close range courtship should be investigated.
- In view of the need for replacement of mass reared strains, a programme of developing and maintaining backup strains is essential.

8. Field cage tests Conclusions

- Field cage tests with host trees are the best compromise between laboratory conditions and costly and impractical field observations to assess medfly mating behaviour under semi-natural conditions.
- Results of field cage studies are still not fully comparable due to insufficient standardisation of test protocols.
- The nutritional status, sex ratio and density of flies in relation to available canopy surface in the field cage influences test results.
- Adequate environmental conditions within a field cage are reflected by wild flies remaining on the tree canopy.

Recommendations

- Field cage tests should be further standardised as follows:
 - ✓ Fly nutritional status = fed water and sugar/protein (3:1) before testing,
 - ✓ Overall fly sex ratio = male-biased (at least 3 males to 1 female),

- ✓ Fly density/canopy available = not less than 20 medium sized (citrus, guava ...) leaves per fly,
- ✓ Fruit availability = at least one medium sized ripe fruit/artificial oviposition substrates for every 20 females,
- ✓ Water availability = at least 4 water sites per tree.
- Even though marking of flies with waterbased paint appears to have no effect on field cage test results, colours used for marking strains should be randomized among replicates.
- Introduction of test flies into the field cage should be done the day of experimentation, at sunrise, releasing the males at least 15 minutes before the females.
- Census frequency should be at least every 30 minutes
- Test duration should cover most of the known sexual activity period.
- Field cage tests should always include wild flies as a control.
- Starting times of field cage testing should be adjusted according to environmental conditions and fly activity (i.e. early start on hot days).
- Test replicates with more than c.a. 25% of wild flies on the cage screen during calling periods reflect inadequate environmental conditions (such as lack of shade, water, light, etc.) and should therefore be repeated.
- Test replicates with less than 20% of potential matings should be repeated.
- Analysis of test results should involve the use of the same indices (RSI, RII, ISI, FRPI, MRPI) and non parametric statistics.

• Fly activities occurring away from the host tree should not be included in the data analysis.

9. Field evaluation Conclusions

- Field cages are the closest approximation to open field evaluation. However, results obtained under these conditions may not always fully represent field performance.
- Given the variability in control programme environment, field evaluation becomes a necessity for major field programmes.
- Currently field evaluations are based primarily on trapping, fruit infestation, and dispersal and survival studies.
- Field evaluations are used to determine effectiveness of mass-reared strains in inducing sterility in target populations.

Recommendations

- Field cage evaluations should be carried out prior to large releases. When possible this should be done at the release sites.
- Induced sterility checks based on egg hatch should be carried out as part of any release program on a regular basis. Therefore, efforts to develop a practical (close to natural) egg laying substrate should be encouraged.
- Strains should be periodically evaluated to ensure compatibility to wild flies and to determine effective survival in the field.
- Post-release fly longevity should be assessed as a supportive measure to strain performance in SIT.

Genetic Applications to Improve the SIT for Tsetse Control/Eradication including Population Genetics

Second RCM held in Mombasa, Kenya, 3-7 October, 1999

There is a need for effective tools to delineate tsetse populations in order that rational decisions can be made for SIT intervention. Populations are isolated when there is no genetic exchange ie gene flow. Powerful new tools are available to analyse gene flow in tsetse and they are being developed and applied in this CRP. In the current meeting several new techniques were described and their use illustrated with the analysis of flies from the morsitans and palpalis groups of tsetse.

The analysis revealed that there is a surprisingly high level of population structuring in tsetse which supports the use of SIT in these areas. However these studies are very preliminary and much more genetic and ecological data are required before SIT intervention is planned. The power and applicability of these techniques will provide a new level of confidence in the complete description of potential target tsetse populations.

New analytical procedures have been developed and many more tsetse populations have been analysed since the last RCM. Polytene chromosome maps are now available for *G. austeni* and *G. pallidipes* and continued studies on the tsetse symbionts have emphasised the role they might play in developing tsetse that are refractory to infection or in the development of natural sterility mechanisms.

As well as the current level of support being provided to the SIT programme in the Southern Rift valley in Ethiopia, considerable discussion took place on the need for population genetic analysis to support proposed SIT projects in Kenya and Mali. The tools to carry out these analyses were described in detail and preliminary results were presented. There is no doubt that if an appropriate survey was carried out in and around the projected target populations, then definitive answers could be given regarding the level of population isolation.

To strengthen the capacity of local counterparts to carry out genetic analysis in Africa, there is a need for training and local infrastructure. During the meeting, plans were made to encourage the training of two fellows in these techniques by enrolling them for PhD programmes in the USA. Concurrently, with the support of RAF/5/040 a small molecular biology laboratory will be set up in Entebbe, Uganda. This laboratory will enable the local counterpart to make good quality DNA preparations from field collected tsetse.

The recommendations included the request that a multi-disciplinary approach be taken to analyse tsetse population genetics and that studies on the bacterial symbionts harboured by tsetse should be expanded.

Molecular and Genetic Approaches to Develop Sexing Strains for Field Applications in Fruit Fly Sterile Insect Technique Programmes

Third RCM held in Tapachula, Mexico, 12 - 16 July 1999

This report summarizes the third Research Co-ordination Meeting (RCM) of the Coordinated Research Programme (CRP) entitled "A Molecular and Genetic Approach to Develop Sexing Strains for Field Application in Fruit Fly Sterile Insect Technique Programmes", the objectives of which are:

- optimization of genetic sexing strains as regards their genetic composition, productivity and application.
- development of third generation genetic sexing strains using nuclear and molecular methods.
- development of genetic sexing strains in fruit flies other than medfly.

Results presented at the meeting demonstrated clearly that for the current

genetic sexing strains (GSS) the first objective has been achieved. Progress to achieve the second objective has been very positively influenced by advances made in a related CRP (D4.10.15) where genetic transformation of several fruit flies has now been demonstrated. Some of this work has been carried out in the Entomology Unit at Seibersdorf and it represents a major breakthrough in the development of third generation GSS. Development of GSS in other fruit fly species remains slow.

The meeting was held in Tapachula, Mexico in order to inform staff at the Moscamed fruit fly facilities of progress in this area. The medfly facility in Tapachula remains the only rearing facility which does not yet use a GSS. The holding of this and the previous RCM's at centres where fruit fly mass rearing is being carried out enables the participants to see the constraints that successful GSSs have to face.

The management of GSSs under massrearing conditions has been significantly improved since the last RCM by the introduction of 1) the filter rearing system (FRS) and 2) the use of better GSS. The results of the use of both these improvements was presented in detail. It is unlikely that using the current technology major improvements will be made in the mass rearing efficiency of the strains. However new data on the isolation of new inversions in medflv will manv subsequently enable the introduction of new genetic material into GSS to be carried out very efficiently.

New data was presented on the use of molecular markers in *Bactrocera tryoni* to understand the distribution and relationships of populations of this species in Australia. These data can be of major importance to plan and implement an SIT programme for this species. Progress in the development of sexing strains in *Bactrocera* species has, in general, been slow with the exception of the work carried out by the Australian group. This is of some concern and during the meeting many suggestions were discussed as to how to improve the situation. One key aspect would be the transfer of marker strains to the relevant laboratories.

The CRP consists of 9 research teams and representatives of all were present at the current meeting together with a number of observers. The first two days of the meeting were devoted to the presentation of reports, followed by two days of deliberations in working groups during which reports were made of the current status, future research goals and a set of recommendations was produced. The meeting concluded with a plenary session at which the list of recommendations was finalized. The meeting agenda and the breakdown of the working groups are given in Annex 3.

Many colleagues from fruit fly programmes in Mexico and Chile were present as observers and they all presented a short overview of the programmes in which they are involved. The colleague from the medfly programme in Chile expressed great interest in the introduction of the *tsl* GSS into the rearing facility in Arica and he will visit Vienna in the fall to further discuss this subject.

Quality Assurance of Mass-Reared & Released Fruit Flies

First RCM held in Vienna, Austria, 1-5 November 1999.

A total of 16 scientific investigators from 14 different countries participated in the identification of research priorities and tasks to be completed over the next 18 months. The group identified 6 key areas where they believe significant progress can be made to improve the production and efficient use of sterile insect during the next 5 years. These areas include:

- 1.) Strain colonisation and colony maintenance,
- 2.) Methods of assessing mating behaviour and competitiveness between facilityreared and wild-collected flies under field cage conditions,
- 3.) Orientation of fruit flies to pheromones, host odours and other scents,
- 4.) Determining what constitutes effective copulation in terms of sperm and

accessory gland fluid transfer and how the incidence of remating be minimised or reduced among once-mated females,

- 5.) Improved and simplified dosimetry methods to measure absorbed doses of radiation and better methods of longdistance shipping of fruit fly pupae, and
- 6.) Examination of survival and dispersal of sterile fruit flies in relation to larval and adult nutrition.

Co-ordination and co-operation between rearing facilities and end-users was encouraged. All participants agreed to input results of their standard emergence and flight ability tests into a shared global database on a weekly basis using a standard input format. This data will include the dosage used to sterilise the insect, name of the production facility, percentages of unemerged, halfemerged, deformed, and normal appearing flies incapable of flight, percentage of emergence and flight ability. Sharing this data will allow end-users to compare their quality control test results with those of the production facility supplying the insects. It also will allow all users and other interested parties to compare the results across different facilities. Lastly, it will promote the universal acceptance and use of standard testing procedures. Transmission of the data will be accomplished via the Internet which is available at most locations with few exceptions.

Consensus was reached in declaring the overall objective of the Co-ordinated Research Project (CRP) to assist fruit fly facilities to increase efficiency of production and end-users in the effective use of sterile fruit flies through standardisation and improvement of quality assurance. More specifically, this will be achieved by harmonising and improving OC/OA tests. values. processes. data management and information exchange on a global level. One of the key outputs will be the publication and distribution of an updated OC Manual in several languages containing new and/or improved procedures, methods, and tests for assurance of quality in the product, mass-rearing production processes. and Emphasis will be given to devising simple yet meaningful ways of assessing quality while trying to avoid additional burdens on facilities and end-users in terms of time, manpower or Automation other resources. and computerisation of testing methods will be encouraged as a means of addressing this need. Equally important to the development of new methodologies will be the new knowledge gained through intensive research into fruit fly behaviour, biology, ecology, and nutrition along with the open exchange of new findings in these key areas.

I. ANNOUNCEMENTS

Call for Collaborators in Molecular Tsetse Population Genetics

Molecular genetic tools based on PCR can now be used to analyse in detail the tsetse genome and that of its symbionts. This analytical approach will enable us to develop a better understanding of intra- and inter-species characteristics of population structure, including isolation, and will ultimately also provide additional epidemiological information, e.g on vectorial capacity.

Symbiotic micro-organisms inhabiting the gut and reproductive tissue in tsetse flies are intimately associated with the fly but very little is known about their genetic variation and distribution within the same or different species of the insect. For instance, the role of *Wolbachia* in intra-population or inter-species sterility is unknown.

There is need to develop appropriate genetic tools with which to analyse tsetse population dynamics and determine gene-flow between neighbouring fly populations.

The recent PAAT Committee Meeting in Rome, 22-23 November 1999, welcomed the relevance of such new technologies for:

- a) the identification of isolated tsetse populations and of confinable "peninsulas" and
- b) a better understanding of the paths that tsetse populations once took or are taking to invade new areas.

Information on the genomes of the tsetse and its symbionts in the field will help in the development of relevant dendogrammes, in the construction of tsetse population-genetic maps and in the design of better strategies for tsetse control / eradication in the future.

Kingsley Fisher leaves Seibersdorf unit

Kingsley Fisher has left the Entomology Unit, Seibersdorf after more than four years. During this time he has helped many facilities with implementing rearing of temperature sensitive lethal strains (*tsl*) of medfly. He has taken up a new position with the USDA and CDFA in Hawaii to work with the upgrading The Joint FAO/IAEA Division intends to facilitate the establishment of a network of scientists in Africa and their collaborators elsewhere in the world who are doing research on the genetic analysis of tsetse populations and their symbionts and can make their results available for use in the development of tsetse population-genetic maps.

This initiative does not aim at supporting general research on tsetse molecular genetics but specifically targets at the design of dendogrammes and population genetic maps, initially focusing on identified priority areas for population-wide tsetse intervention in Eastand West-Africa. Additional intervention areas in other sub-regions will be covered in a subsequent phase.

Scientists, particularly from Africa, are invited to indicate their interest in participating in the establishment of such a network by contacting:

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In your letter briefly outline your relevant experience in the field or laboratory work.

of *tsl* rearing there and the complete refurbishment of the CDFA factory.

We look forward to continuing to work closely with him to further develop the mass rearing of *tsl* strains.

Marc Vreysen leaves Addis Ababa

Marc Vreysen, Regional Expert under the project RAF/5/040, SIT for Tsetse and Trypanosomosis Management in Africa, is leaving Addis Ababa, Ethiopia, after two years. Marc moved to Addis after the completion of the Zanzibar eradication project, and whilst there has been involved with both

John Kabayo joins Regional Africa Project

John Kabayo has just joined the Regional Africa Project, RAF/5/040, SIT for Tsetse and Trypanosomosis Management in Africa. He will work on a strategy for raising awareness at

the Ethiopia project and a new project proposal from Mali, and has continued to visit Zanzibar to provide advice and assistance.

We wish Marc and his family all the best for the future, and look forward to working with him on future projects.

the highest levels within African Member states of the tsetse and trypanosomosis problem and the potential of area wide control to provide significant benefits.

Abdeljelil Bakri leaves section

Abdeljelil leaves the section after a short contract, during which time he has been working on the medfly and *Anastrepha fraterculus* sites on the GPPIS database (see page 22) and compiled information on mass rearing facilities (page 23). We wish him all the best on his return to Morocco, and look forward to working with him in the future.

J. PUBLICATIONS

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