



Joint FAO/IAEA Programme  
Nuclear Techniques in Food and Agriculture

# Insect Pest Control Newsletter



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## To Our Readers



*The tsetse and trypanosomosis suppression in the Niayes Senegal resulted in drastic reduction in disease transmission, significant increase in milk yields, and a ten-fold increase in the rate of imported more productive exotic cattle with a very positive overall return on investment.*

In Senegal, as in many other parts of West Africa, African animal trypanosomosis (AAT), a deadly disease carried by the tsetse fly, has long been a major obstacle to the development of more efficient and sustainable livestock production systems. For more than 50 years, the Government of Senegal invested in importing more productive improved breeds from Europe and elsewhere, but many did not survive because they had no natural tolerance to AAT. The frequency of the deadly disease transmitted by the tsetse has dropped dramatically in Senegal's Niayes region, as the long-term government-executed tsetse eradication campaign that involved the release of sterile tsetse flies has suppressed the pest population.



Now, in Niayes region, the incidence of the disease is close to zero and flies have been eliminated in all but one hotspot area. Nevertheless, the release of sterile tsetse flies will continue for several months in order to tackle any potential residual fly pocket and ensure that no wild flies remain.

Before the project's success, most farmers reared only local cattle breeds, 80–90 000 heads of which live in the area. These cattle had low milk and meat production and low reproductive rates, but the farmers kept them for their natural tolerance to trypanosomosis. Nowadays, the suppression of the tsetse population is allowing farmers to replace the low producing local cattle with more productive exotic breeds.

Working together, farmers in the region are progressively replacing their indigenous cattle that produce only 1–2 liters of milk a day with exotic cattle that produce between 20 and 40 liters a day and, hence, substantially increasing productivity. Furthermore, the improved milk and meat production of the exotic breeds will enable farmers to increase their income, including with smaller herds, which in turn will reduce the grazing pressure on this already fragile ecosystem.

Much credit for the success is given to the adaptive management approach of the project, which called for monthly project coordination meetings with the many different stakeholders, for transparency at all levels and for decision-making by consensus, with decisions for moving the project ahead based on scientific evidence.

Looking to the future, several countries are considering plans to set up similar programmes on their land. A genetic study, covering all West Africa, has identified ten tsetse-infested potential ecological islands, that would benefit from achievements and lessons learned in Niayes.

Another major achievement, during the past semester, has been the publication of a Special Issue in BMC Microbiology (<https://bmcmicrobiol.biomedcentral.com/articles/supplements/volume-18-supplement-1>) as part of the work conducted during the last five years of the Coordinated Research Project (CRP) titled 'Enhancing Vector Refractoriness to Trypanosome Infection'.



Volume 18 Supplement 1

## Enhancing Vector Refractoriness to Trypanosome Infection

**Research**

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Edited by Adly M.M. Abd-Alla, George Tsiamis, and Drion Boucias.

## Joint FAO/IAEA Programme

### Nuclear Techniques in Food and Agriculture

*Special Issue in BMC Microbiology.*

This CRP involved 23 scientists from 19 countries, bringing a wide range of expertise and experience, with two major goals: (i) to gain a deeper knowledge of the tripartite interactions between the tsetse fly vectors, their symbionts, and trypanosome parasites and (ii) to acquire a better understanding of mechanisms that limit the development of trypanosome infections in tsetse and how these may be enhanced. The research studies involved detailed investigations into the biology of the insect in relationship to the



causative trypanosomes, parasites, and symbionts, as well as epidemiological investigations of the disease in various parts of Africa.

This special issue comprises the final research results of the CRP, compiled in 19 research papers, together with an introductory review paper highlighting the objectives and the main achievements of the CRP and three reviews providing background to the project. We believe that this issue in BMC Microbiology is an important contribution to our knowledge on tsetse flies and their symbionts, parasites, and pathogens. It will provide the insect pathology and symbionts scientific community with extensive knowledge about the recent advances made in this field, with great potential to solve this crucial problem that continues to affect both humans and cattle in sub-Saharan Africa.

In meanwhile, other special issue was published at the Parasites & Vectors and can be downloaded at: (<https://parasitesandvectors.biomedcentral.com/articles/supplements/volume-11-supplement-2>). This special issue summarises the achievements made during the five years CRP on 'Exploring Genetic, Molecular, Mechanical and Behavioural Methods of Sex Separation in Mosquitoes'. For more detailed information please see page 20 of this newsletter.

Regarding fruit flies, an outbreak of *Bactrocera scutellata* was reported by the National Plant Protection Organization (NPPO) of Mexico on 2 February 2018 in Manzanillo, Colima, Mexico. The outbreak has now been eradicated, since from the last specimen captured, nine biological cycles have passed without any additional detection (25 April to 30 September 2018). The nine cycles were assessed using the degree-day model. The male annihilation technique (MAT) and the delimiting trapping were intensified in the last six cycles, following the recommendations of the FAO/IAEA expert. Eradication actions were enforced by the NPPO following the framework of the 'Action Plan Against *B. scutellata* on National Territory (March 2018)' and based on the International Standards of Phytosanitary Measures (ISPM 26) relative to the determination of pest status in an area and ISPM 9 relative to the directives for pest eradication programmes.

Following the Action Plan procedures, the MAT devices will be gradually retrieved from the field and disposed. Based on the assessed risk of introduction and spread of this transient actionable pest entry, the trapping network will be restructured in the urban area and at the Manzanillo Port, increasing the numbers from 48 to 567 permanent traps. In addition, the trapping network against non-native invasive species of quarantine significance will be reinforced along the Pacific Coast including: Topolobampo and Mazatlán, Sinaloa; Lázaro Cárdenas, Michoacán; Salina Cruz, Oaxaca and; Puerto Chiapas, Chiapas.

On the other hand, the Oriental fruit fly *Bactrocera dorsalis*, pictured next, was detected for the first time in Europe in Italy in late September 2018. The fly was detected in the

vicinity of Naples. The detection requires a delimitation of the infested area and an eradication plan if further specimens are detected. For more data on the detection, a recent paper was published in the Journal *Insects* 2018, 9, 182; doi:10.3390/insects9040182 (<https://www.mdpi.com/2075-4450/9/4/182>) 'First Record of an Invasive Fruit Fly Belonging to *Bactrocera dorsalis* Complex (Diptera: Tephritidae) in Europe'. This event shows the vulnerability of the Mediterranean countries to the introduction of *B. dorsalis* and the peach fruit fly, *B. zonata* and the consequent implications for the fruit industry of those countries.



On the subject of human disease vectors, research and development is making advances on the Sterile Insect Technique (SIT) package to control disease transmitting mosquitoes, including *Aedes aegypti* and *Ae. albopictus* the vectors for dengue, chikungunya, Zika and yellow fever virus with the ultimate goal of the SIT to be included in an Area-wide Integrated Pest Management (AW-IPM) approach.

This work has been implemented through in-house research and development at the Insect Pest Control Laboratory (IPCL) of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, in Seibersdorf, through Coordinated Research Projects and extra-budgetary Technical Cooperation Projects, as well as through direct collaboration with research institutes. In parallel, every possible effort is being made to foster capacity building and technology transfer to Member States through IAEA Technical Cooperation Projects.

Regarding IPC staff news, it is with great regret to inform you that our former colleague Ludwig Lang passed away on 1 November 2018. He retired from the Insect Pest Control Laboratory, where he worked in the plant pest group and was responsible of the quality control of fruit flies. We would like to express our sincere condolences to his family.

Finally, to each of you I extend my very best wishes for 2019.

**Rui Cardoso Pereira**

**Head, Insect Pest Control Section**

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# Forthcoming Events (2019)

## I. Research Coordination Meetings (RCMs) of FAO/IAEA Coordinated Research Projects (CRPs)

Second RCM on Integration of the SIT with Biocontrol for Greenhouse Insect Pest Management. 4–8 March 2019, Mendoza, Argentina.

Fourth RCM on Dormancy Management to Enable Mass-rearing and Increase Efficacy of Sterile Insects and Natural Enemies. 3–7 June 2019, Thessaloniki, Greece.

First RCM on Assessment of Simultaneous Application of SIT and MAT to Enhance *Bactrocera* Fruit Fly Management. 15–19 July 2019, Vienna, Austria.

First RCM on Generic Approach for the Development of Genetic Sexing Strains for SIT Applications. 7–11 October 2019, Vienna, Austria.

Third RCM on Improved Field Performance of Sterile Male Lepidoptera to Ensure Success in SIT Programmes. 21–25 October 2019, Mendoza, Argentina.

Fourth RCM on Comparing Rearing Efficiency and Competitiveness of Sterile Male Strains Produced by Genetic, Transgenic or Symbiont-based Technologies. 2–6 December 2019, Adelaide, Australia.

## II. Consultants and Expert Meetings

FAO/IAEA Consultants Meeting on Mosquito Radiation, Sterilization and Quality Control (under Regional TC Project RER5022). 25–29 March 2019, Vienna, Austria.

## III. Other Meetings/Events

FAO/IAEA Training on Basic Use of R Software to infer Demographic Parameters of Wild and Sterile Mosquitoes from Entomological Monitoring Data (under Regional TC Project RER5022). 18–22 February 2019, Seibersdorf, Austria.

FAO/IAEA/WHO Workshop to Initiate the Development of Joint Guidance for the Application of SIT for Mosquito Borne Diseases (under Interregional TC Project INT5155). 27 February–1 March 2019, Tapachula, Mexico.

FAO/IAEA Regional Training Course on Components of the Sterile Insect Technique: Principles and Practice (under Regional TC Project RLA5074). 27 February–1 March 2019, Buenos Aires, Argentina.

FAO/IAEA Workshop on Techniques for the Rearing, Quality Control and Radiation Sterilization of *Drosophila suzukii*. 10–12 March 2019, Mendoza, Argentina.

Fourteen Session of the Commission on Phytosanitary Measures, International Plant Protection Convention, FAO. 1–5 April 2019, Rome, Italy.

FAO/IAEA Regional Training Course on Marking, Handling, Transport and Release of Sterile Mosquitoes (under Regional TC Project RLA5074). 29 April–3 May 2019, Lima, Peru.

FAO/IAEA/WHO Regional Joint Meeting between Entomologists, Virologists and Epidemiologists to Boost the Collaboration for Vector Control in the European Region (under Regional TC Project RER5022). 6–10 May 2019, Tirana, Albania.

FAO/IAEA Interregional Training Course on The Use of the Sterile Insect and Related Techniques for the Integrated Area-wide Management of Insect Pests (under Interregional TC Project INT5155). 10 June–5 July 2019, Metapa de Dominguez, Chiapas, Mexico and Antigua / El Pino, Guatemala.

Technical Panel on Phytosanitary Treatments (TPPT), International Plant Protection Convention, FAO. 8–12 July 2019, Vienna, Austria.

FAO/IAEA/WHO Workshop to Finalize Joint Guidance for the Application of SIT for Mosquito Borne Diseases (under Interregional TC Project INT5155). 24–28 June 2019, Vienna, Austria.

FAO/IAEA Decision-makers Workshop on the Use of the SIT (under Interregional TC Project INT5155). 26–28 August 2019, Fort Myers, Florida, USA.

FAO/IAEA Regional Training Course on Modern Taxonomy and Identification Tools of Fruit Fly Species in Africa (under Regional TC Project RAF5074). 23–27 September 2019, Cotonou, Benin.

FAO/IAEA Regional Training Course on Communication within SIT Mosquito Projects (under Regional TC Project RER5022). 23–27 September 2019, Procida, Italy.

FAO/IAEA Regional Training Course on Area-wide Integrated Fruit Fly Management including Sterile Insect Technique (SIT) and Male Annihilation Technology (MAT) in Africa (under Regional TC Project RAF5074). 7–11 October 2019, Reduit, Mauritius.

FAO/IAEA Meeting on the review of Thematic Plan for the Development and Application of the Sterile Insect Technique (SIT) and Related Genetic and Biological Control Methods for Disease Transmitting Mosquitoes (under Interregional TC Project INT5155). 14–18 October 2019, Vienna, Austria.

FAO/IAEA Workshop on Preparation of SIT Pilot Trials against Mosquitoes (under Interregional TC Project INT5155). 11–15 November 2019, Valencia, Spain.

FAO/IAEA Workshop on a Best Practice Manual on Field Performance of Sterile Male Moths. 17–19 October 2019, Mendoza, Argentina.



## Past Events (2018)

### I. Research Coordination Meetings (RCMs) of FAO/IAEA Coordinated Research Projects (CRPs)

Fourth RCM on Exploring Genetic, Molecular, Mechanical and Behavioural Methods of Sex Separation in Mosquitoes. 19–23 February 2018, Bangkok, Thailand.

Second RCM on Improved Field Performance of Sterile Male Lepidoptera to Ensure Success in SIT Programmes. 12–16 March 2018, Palmerston North, New Zealand.

Third RCM on Comparing Rearing Efficiency and Competitiveness of Sterile Male Strains Produced by Genetic, Transgenic or Symbiont-based Technologies. 18–22 June 2018, Bangkok, Thailand.

First RCM on Colony Management of Insects for Sterile Insect Technique Application. 23–27 July 2018, Vienna, Austria.

Third RCM on Mosquito Handling, Transport, Release and Male Trapping Methods. 12–16 November 2018, Juazeiro, Bahia, Brazil.

### II. Consultants and Expert Meetings

FAO/IAEA Expert Meeting on Harmonization of Irradiation and Dosimetry Protocols for *Aedes* Invasive Mosquitoes (under Regional TC Project RER5022). 12–14 February 2018, Vienna, Austria.

FAO/IAEA Consultants Meeting on Dashboard for Process Control in Mass-rearing Facilities, and Automatic Insect Density Estimation for Sterile Insect Release. 23–27 April 2018, Tapachula, Mexico.

FAO/IAEA Consultants Meeting on Isolation of Genetic Markers towards the Development of Generic Methods for the Construction of Genetic Sexing Strains for Sterile Insect Technique (SIT) Applications. 3–5 October 2018, Vienna, Austria.

FAO/IAEA Expert Meeting on Required Parameters for Mosquito SIT Application (under Interregional TC Project INT5155). 8–10 October 2018, Vienna, Austria.

FAO/IAEA Expert Meeting on Technical Guidelines for Mosquito SIT Application (under Interregional TC Project INT5155). 3–7 December 2018, Vienna, Austria.

FAO/IAEA Consultants Meeting on *Aedes* Mosquito Sterile Insect Technique Package: Review and Future Perspectives. 10–12 December 2018, Vienna, Austria.

### III. Other Meetings/Events

FAO/IAEA First Coordination and Consultative Meeting of Managing and Controlling *Aedes* Vector Populations Using the Sterile Insect Technique (under Regional TC Project Asia Pacific RAS5082). 12–16 February 2018, Bangkok, Thailand.

SPS Committee Thematic Session on Pest Free Areas. World Trade Organization (WTO). 27 February 2018, Geneva, Switzerland.

FAO/IAEA Workshop to Develop a Best Practice Manual on Field Performance of Sterile Male Moths. 17 March 2018, Palmerston North, New Zealand.

FAO/IAEA First Coordination Meeting of Latin America Regional Project Strengthening the Regional Capacities in the Prevention and Progressive Control of Screwworm (under Regional TC Project RLA5075). 19–23 March 2018, Montevideo, Uruguay.

FAO/IAEA Regional Training Course on Methods for the Handling, Marking, Transportation and Release of Sterile Male *Aedes aegypti*, as Components of the SIT Package for the Control of Mosquito Disease Vectors of Zika, Dengue and Chikungunya (under Regional TC Project RLA5074). 19–23 March 2018, Juazeiro, Brazil.

Thirteenth Session of the Commission on Phytosanitary Measures, International Plant Protection Convention, FAO. 16–20 April 2017, Rome, Italy.

10th International Symposium on Fruit Flies of Economic Importance. 23–27 April 2018, Tapachula, Mexico.

FAO/IAEA Second Coordination Meeting of Latin America Regional Project Strengthening Fruit Fly Surveillance and Control Measures Using the Sterile Insect Technique in an Area-wide and Integrated Pest Management Approach for the Protection and Expansion of Horticultural Production (under Regional TC Project RLA5070). 23–27 April 2018, Tapachula, Chiapas, Mexico.

FAO/IAEA Stakeholder Engagement Meeting for Sterile Male Mosquito Release Pilot Trials in Europe (under Regional TC Project RER5022). 23–27 April 2018, Vienna, Austria.

FAO/IAEA Meeting of the New World Screwworm Pest Risk Analysis Panel (under TC Project RLA5075). 1–5 May 2018, Sarasota, Florida, United States.

FAO/IAEA Second Coordination Meeting of the Latin America and Caribbean Regional Project Strengthening Regional Capacity in Latin America and the Caribbean for Integrated Vector Management Approaches with a Sterile Insect Technique Component, to Control *Aedes* Mosquitoes as Vectors of Human Pathogens, particularly Zika Virus (under Regional TC Project RLA5074). 7–11 May 2018, Vienna, Austria.

FAO/IAEA Project Mid-Term Review Meeting (under Interregional TC Project INT5155). 14–17 May 2018, Bologna, Italy.

FAO/IAEA Workshop on Recent Developments and Impact Assessment of Sterile and Incompatibility Insect Techniques for Mosquito Control (under Regional TC Project RAS5082). 4–8 June 2018, Singapore.

FAO/IAEA Stakeholders Meeting for Discussions on Alternative Pest Management Approaches Including SIT against the Parasitic Fly *Philornis downsi* on the Galapagos Islands (under Regional TC Project RLA5070). 11–13 June 2018, Vienna, Austria.

Scoping Meeting on the Issues and Implications of Guidance on International Trade and Transport of Insects. 19–20 June 2018, London, United Kingdom.

FAO/IAEA Training Course on Data Management, GIS and Modelling to Optimize SIT Control Efforts of Invading Mosquito Species in Europe (under Regional TC Project RER5022). 9–13 July 2018, Montpellier, France.

FAO/IAEA Mid-term Coordination Meeting of Africa Regional Project Enhancing Capacity for Detection, Surveillance and Suppression of Exotic and Established Fruit Fly Species through Integration of Sterile Insect Technique with Other Suppression Methods (under Regional TC Project RAF5074). 23–27 July 2018, Accra, Ghana.

Meeting of an Expert Panel on Drafting a Procedures Manual on Establishment and Maintenance of Pest Free Areas (PFA) and Areas of Low Pest Prevalence (ALPP) in support of International Standards of Phytosanitary Measures of the Plant Protection Convention (IPPC). 23–27 July 2018, Raleigh, North Carolina, USA.

FAO/IAEA Regional Training Course on Identification and Diagnostics of New World Screwworm (under Regional Latin America RLA5075). 13–17 August 2018, Panama City, Panama.

FAO/IAEA Regional Training Course on the Use of GIS and International Standards for Phytosanitary Measures (ISPMs) for Fruit Fly Activities in Africa (under Regional TC Project RAF5074). 20–24 August 2018, Gaborone, Botswana.

FAO/IAEA Regional Training Course on Early Detection of Invasive Non-native Fruit Fly Species and Emergency Response (under Regional TC Project RAS5076). 14–18 October 2018, Amman, Jordan.

FAO/IAEA Workshop on the Development of a Regional Project to Assess the Feasibility of the SIT for the Cocoa Pod Borer (under regional project RAS0081). 15–19 October 2018, Kota Kinabalu, Malaysia.

China International Conference on Nuclear Technology Applications. 17–19 October 2018, Dongguan, China.

FAO/IAEA Regional Training Course on the Use of Georeferenced Fly Rounds for Cost-Effective Entomological Surveillance of Savannah Tsetse Species (under Regional TC Project RAF5080). 29 October–2 November 2018, Arusha, United Republic of Tanzania.

FAO/IAEA Regional Joint Meeting between Entomologists, Virologists and Epidemiologists to Boost the Collaboration for Vector Control in the European Region (under Regional TC Project RER5022). 5–9 November 2018, Tirana, Albania.

FAO/IAEA Regional Training Course on Molecular Tools Applied to Tsetse Control Programmes (under Regional TC Project RAF5080). 19–30 November 2018, Bobo Dioulasso, Burkina Faso.

FAO/IAEA Regional Training Course on Early Detection of Invasive Non-native Fruit Fly Species and Emergency Response (under Regional TC Project RAS5076). 2–6 December 2018, Tel-Aviv, Israel.

FAO/IAEA Regional Training Course on Epidemiological Surveillance and Emergency Plan for the Eradication of NWS Outbreaks (under TC Project RLA5075). 3–7 December 2018, Panama City, Panama.

FAO/IAEA Final Coordination Meeting and Workshop on Phytosanitary Measures for International Trade of Fruits and Vegetables (under TC Project RLA5070). 3–7 December 2018, Guatemala City, Guatemala.

FAO/IAEA Meeting for Launching of the MEDNIP Database (under TC Project RAS5076). 10–14 December 2018, Vienna, Austria.

FAO/IAEA Regional Coordination Meeting on Progresses and Perspectives on the Control Programmes for *Aedes* Invasive Mosquitoes (under Regional TC Project RER5022). 17–19 December, Vienna, Austria.

## Technical Cooperation Projects

The Insect Pest Control subprogramme currently has technical responsibilities for the following technical cooperation projects that are managed by the IAEA's Department of Technical Cooperation. They can be classed under four major topics, namely:

- Biocontrol using radiation
- Human disease vectors
- Livestock pests
- Plant pests

Country	Project Number	National Projects	Technical Officer
Bolivia	BOL5022	Reducing Fruit Fly Populations in Different Regions Introducing an Integrated Pest Management Approach Including the Use of the Sterile Insect Technique	Walther Enkerlin
Botswana	BOT5013	Using the Sterile Insect Technique Integrated with Other Suppression Methods for Managing <i>Bactrocera dorsalis</i>	Daguang Lu
Brazil	BRA5060	Using the Sterile Insect Technique to Evaluate a Local Strain in the Control of <i>Aedes aegypti</i>	Rafael Argiles
Burkina Faso	BKF5020	Strengthening the Insectarium to Create Agropastoral Areas Permanently Liberated from Tsetse Flies and Trypanosomiasis	Adly Abdalla
Chad	CHD5007	Contributing to the Eradication of <i>Glossina fuscipes fuscipes</i> to Improve Food and Nutritional Security	Rafael Argiles
Chile	CHI5051	Implementing Pilot Level of Sterile Insect Technique for Control of <i>Lobesia botrana</i> in Urban Areas	Walther Enkerlin
China	CPR5020	Integrating the Sterile Insect Technique (SIT) for Area-wide Integrated Pest Management of Tephritid Fruit Flies	Rui Cardoso Pereira
Cuba	CUB5021	Demonstrating the Feasibility of the Sterile Insect Technique in the Control of Vectors and Pests	Rafael Argiles
Ecuador	ECU5029	Improving Integrated Fruit Fly Management in Fruit and Vegetable Production Areas	Walther Enkerlin
Ethiopia	ETH5021	Enhancing Livestock and Crop Production Through Continued Consolidated and Sustainable Control of Tsetse and Trypanosomosis	Rafael Argiles
Fiji	FIJ5001	Examining Options for the Management of Fruit Flies	Daguang Lu
Guatemala	GUA5019	Strengthening National Capabilities for the Control of Agricultural Pests Using Nuclear Technologies	Walther Enkerlin
Israel	ISR5021	Assisting in the Development of a Strategy to Counteract <i>Bactrocera zonata</i>	Walther Enkerlin
Libya	LIB5011	Enhancing Area-wide Integrated Management of Fruit Flies	Walther Enkerlin



Mexico	MEX5031	Using the Sterile Insect Technique to Control Dengue Vectors	Danilo Carvalho
Morocco	MOR5035	Implementing the Sterile Insect Technique in the Souss Valley	Walther Enkerlin Carlos Cáceres
Oman	OMA5007	Strengthening Sterile Insect Technique Based Area-wide Integrated Management of Date Palm Pests	Marc Vreysen
Palau	PLW5002	Improving the Quantity and Quality of Fruits for Exportation and Domestic Consumption Through Area-wide Integrated Pest Management of <i>Bactrocera</i> Fruit Flies in Tropical Fruit and Vegetable Production Areas (Phase II)	Daguang Lu
Papua New Guinea	PAP5001	Supporting a Feasibility Study on Using the Sterile Insect Technique against the Cocoa Pod Borer	Marc Vreysen
Philippines	PHI5033	Building Capacity in Using the Sterile Insect Technique against Dengue and Chikungunya Vectors	Wadaka Mamai
Senegal	SEN5037	Supporting the National Programme to Control Tsetse and Trypanosomosis	Marc Vreysen Rafael Argiles
South Africa	SAF5014	Assessing the Sterile Insect Technique for Malaria Mosquitos in a South African Setting, Phase II	Hanano Yamada
South Africa	SAF5015	Supporting the Control of Nagana in South Africa Using an Area-Wide Integrated Pest Management Approach with a Sterile Insect Technique Component - Phase I	Marc Vreysen
Seychelles	SEY5009	Suppressing Melon Fruit Fly Species through Environment-Friendly Techniques to Enhance Food Security	Rui Cardoso Pereira
Sri Lanka	SRL5047	Establishing a National Centre for Research, Training and Services in Medical and Molecular Entomology for Vector-borne Disease Control	Kostas Bourtzis
Sudan	SUD5038	Implementing the Sterile Insect Technique for Integrated Control of <i>Anopheles arabiensis</i> , Phase II	Adly Abdalla
Thailand	THA5052	Developing Sustainable Management of Fruit Flies Integrating Sterile Insect Technique with other Suppression Methods	Daguang Lu
Uganda	UGA5036	Demonstrating the Feasibility of a Sterile Insect Technique Component as Part of an Area-wide Integrated Pest Management Approach to Increase Livestock Productivity	Rafael Argiles
Viet Nam	VIE5021	Integration of the Sterile Insect Technique with Other Suppression Methods for Control of <i>Bactrocera</i> fruit flies in Dragon Fruit Production	Rui Cardoso Pereira
Zimbabwe	ZIM5023	Improving Crop and Livestock Production through the Eradication of Bovine and Human Trypanosomiasis in Matusadona National Park	Rafael Argiles

		Regional Projects	
Regional Africa	RAF5074	Enhancing Capacity for Detection, Surveillance and Suppression of Exotic and Established Fruit Fly Species through Integration of Sterile Insect Technique with Other Suppression Methods	Daguang Lu
Regional Africa	RAF5080	Supporting Area-Wide Tsetse and Trypanosomosis Management to Improve Livestock Productivity - Phase IV	Rafael Argiles
Regional Asia (ARASIA)	RAS5076	Harmonizing and Strengthening Surveillance Systems to Prevent and Control Exotic and Native Fruit Flies Including the Use of the Sterile Insect Technique	Walther Enkerlin Adly Abdalla
Regional Asia	RAS5082	Managing and Controlling <i>Aedes</i> Vector Populations Using the Sterile Insect Technique	Marc Vreysen Hamidou Maiga
Regional Europe	RER5022	Establishing Genetic Control Programmes for <i>Aedes</i> Invasive Mosquitoes	Jeremy Bouyer
Regional Latin America (ARCAL)	RLA5070	Strengthening Fruit Fly Surveillance and Control Measures Using the Sterile Insect Technique in an Area-wide and Integrated Pest Management Approach for the Protection and Expansion of Horticultural Production (ARCAL CXLI)	Walther Enkerlin
Regional Latin America	RLA5074	Strengthening Regional Capacity in Latin America and the Caribbean for Integrated Vector Management Approaches with a Sterile Insect Technique Component, to Control <i>Aedes</i> Mosquitoes as Vectors of Human Pathogens, particularly Zika Virus	Hanano Yamada Rui Cardoso Pereira
Regional Latin America	RLA5075	Strengthening the Regional Capacities in the Prevention and Progressive Control of Screwworm	Walther Enkerlin
		Interregional Project	
Interregional	INT5155	Sharing Knowledge on the Sterile Insect and Related Techniques for the Integrated Area-wide Management of Insect Pests and Human Disease Vectors	Jeremy Bouyer Rui Cardoso Pereira

## Highlights of Technical Cooperation Projects

### Improving Integrated Fruit Fly Management in Fruit and Vegetable Production Areas (ECU5029)

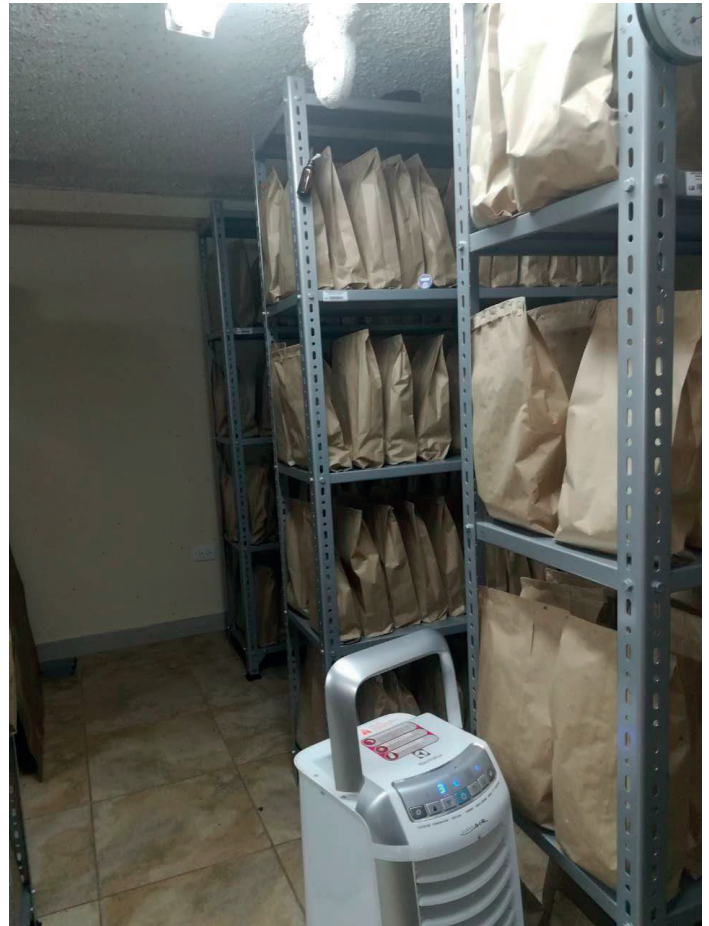
The Agencia Ecuatoriana de Aseguramiento de la Calidad del Agro (AGROCALIDAD) of the Ministry of Agriculture of Ecuador, with assistance from the IAEA and FAO, has built and equipped a sterile emergence and release facility. The facility is strategically located near a carefully selected area where SIT will be applied on a pilot scale for the suppression of Mediterranean fruit fly populations and establishment of areas of low pest prevalence. On the selected pilot areas, fruit crops are cultivated for sales on the national and international markets, through a pest risk mitigation scheme known as systems approach.



*Sterile Mediterranean fruit fly pupae shipped from El Pino facility in Guatemala.*

The first shipment of sterile Mediterranean fruit fly pupae reached Quito, Ecuador on 8 November 2018. Five million sterile pupae were shipped from the El Pino Mediterranean fruit fly mass rearing and sterilization facility in Guatemala to Quito, Ecuador. The pupae were then transported to the fly emergence and release facility for holding until adult emergence and field release, scheduled for 14 November 2018. Adult sterile flies will be released on the pilot areas in the provinces of Pichincha, Imbabura and Tungurahua. Due to the relatively small scale of the operation, the sterile flies will be packed in paper bags and released by ground on a grid of evenly distributed release spots. Weekly shipments of sterile Mediterranean fruit fly pupae from El Pino should continue for at least the next 24 to 36 months. The impact of SIT on the ground, will be measured through a trapping network placed across the targeted pilot area for adult pest population monitoring and through fruit sampling to assess infestation levels. Both the trapping and the fruit sampling are technologies that have been transferred to Ecuador as part of this project.

The SIT will be integrated with other pest control techniques to achieve population suppression. A continued, participative and coordinated effort between AGROCALIDAD, the growers and local communities will be fundamental for the project success.



*Sterile Mediterranean fruit fly pupae packed in paper bags for ground releases.*

### Enhancing Capacity for Detection, Surveillance and Suppression of Exotic and Established Fruit Fly Species through Integration of Sterile Insect Technique with Other Suppression Methods (RAF5074)

#### Regional Training Course on the Use of GIS and International Standards for Phytosanitary Measures (ISPMs) for Fruit Fly Activities in Africa, Gaborone, Botswana, 20–24 August 2018

This regional training course was organized under the Africa regional TC project RAF5074 and was attended by 24 participants from 18 countries (Benin, Botswana, Burundi, Cote d'Ivoire, Eswatini, Ghana, Kenya, Madagascar, Mozambique, Namibia, Nigeria, Senegal, Seychelles, Sudan, Tunisia, Uganda, Zambia, Zimbabwe). The course was hosted by Botswana National Plant Protection Division of



the Ministry of Agricultural Development and Food Security, and was held in Gaborone, Botswana from 20–24 August 2018.



*Participants of Regional Training Course on the Use of GIS and International Standards for Phytosanitary Measures (ISPMs) for Fruit Fly Activities in Africa (Gaborone, Botswana).*

The programme of the training course consisted of theoretical lectures, demonstrations and practical exercises, and covered the following main topics:

- International Plant Protection Convention (IPPC) introduction and Standards relevant to Fruit fly ISPMs including ISPM4 Requirements for the establishment of pest free areas; ISPM5 Glossary of phytosanitary terms ISPM6 Surveillance; ISPM9 Guidelines for pest eradication programmes; ISPM10 Requirements for the establishment of pest free places of production and pest free production sites; ISPM14 the use of integrated measures in a systems approach for pest risk management; ISPM22 Requirements for the establishment of areas of low pest prevalence; ISPM26 Establishment of pest free areas for fruit flies (Tephritidae); ISPM35 Systems approach for pest risk management of fruit flies (Tephritidae); ISPM37 Determination of host status of fruit to fruit flies (Tephritidae); and the revision of the fruit fly standards.
- Cartography and GIS introduction; GPS and collection of data in the field; GIS data collection, storage, analysis and display; GIS and decision making.



*Participants in lecture.*

With the knowledge on the fruit fly ISPMs, countries will be better equipped to find solutions for reducing their fruit

fly infestations and facilitate trade. For example, host plant research can show that certain fruits or varieties are not or hardly attacked by fruit flies, thereby creating an opportunity for the market.

The GIS training was conducted using the example of Medfly Program in Guatemala, which allowed the participants to apply the GIS to evaluate the Sterile Insect Technique (SIT) in an actual fruit fly management programme. Participants learned techniques, procedures and standards on how the trapping data can be collected, stored, analysed and displayed with a geographic perspective and with the support of GPS and GIS. They also learned how to overlay and integrate data with other information to evaluate SIT performance. At the end, the participants were able to generate and present their results. The training was provided using Open Source Software, which will allow the participants to follow-up by themselves using a software freely available.

### **Mid-term Coordination Meeting, Accra, Ghana, 22–27 July 2018**

The meeting was held to review and evaluate the progresses and achievements under the Africa Regional Technical Cooperation Project RAF5074, to share and harmonize methodologies on fruit fly activities and plan its future project activities. The meeting was attended by 24 participants including the project counterparts from 20 African Member States (Benin, Botswana, Burkina Faso, Burundi, Cote d'Ivoire, Eswatini, Ghana, Kenya, Libya, Madagascar, Mozambique, Namibia, Nigeria, Senegal, Seychelles, Sudan, Tunisia, Uganda, United Republic of Tanzania, Zimbabwe) and the representatives from Belgium Royal Museum for Central Africa (RMCA) and the French Agricultural Research Centre for International Development (CIRAD).



*Participants of Mid-term Coordination Meeting of RAF5074 (Accra, Ghana).*

Under the technical cooperation project RAF5074, the project participating Member States conduct various activities for the surveillance and control of fruit flies in the region. The major activities being conducted were: studying the fruit flies population dynamic, identification of fruit flies associated with mango and other hosts, assessing losses caused by fruit flies, sensitizing actors in the fruits and

vegetable sectors to alert them on the dangers of fruit flies and how to manage or control them.

The participants identified challenges and future needs and agreed on future plans of the project such as: the Member States emphasis on the expansion of the surveillance system to include the areas of likely introduction of non-native invasive species, and the intensification of surveillance for established species. Participants were also aware that the varying capacity on fruit fly detection, monitoring and suppression among participating Member States. The harmonization and standardization of the surveillance activates especially on fruit fly data collection need to be improved.

### Implementing Pilot Level of Sterile Insect Technique for Control of *Lobesia botrana* in Urban Areas (CHI5051)

In October 2018, the *Lobesia botrana* adult recovery system was set-up at the facility in Arica, Chile. A trial run of the moth collection system was executed, and the set-up worked well with recommendations provided for improvement.



Unpacking of the adult moth collection system developed by the USDA and transferred to Chile.

In relation to rearing operations, in the past two years there has been substantial progress, including: (1) Addition of more larval rearing space (increase of 9 m<sup>2</sup> of area), (2) Development of a diet disposal and excess inset material disposal area, (3) A hot box and autoclave sterilization for spent diet, and (4) Separation of most clean rearing areas including diet production, egg treatment, rearing tray infes-

tation from 'dirty' activities where moth scales are produced and diet disposal areas.

There is a walk in cold room and a storage area which is being converted for use of the adult collection system. There is a total of about 100 m<sup>2</sup> dedicated to the rearing operation which includes a small laboratory and office space.



European grapevine moth (*Lobesia botrana*).

Overall there is a good biosecurity system in place with an outside area for changing into clothing dedicated for working inside the rearing space. Current *Lobesia* production is about 8 000 pupae per week. Of these ~5 000 are used to set egg cages and the remainder are available for different testing needs, including for use in irradiation studies and field cage test or open field testing. Because the production methods are still under development, the number of eggs available per day and the number seeding per tray have been variable, ranging from 250 to 1 500 eggs per tray. Rates of eclosion in individual trays ranged from 50% to 90%, from December 2017 until June 2018.

Consistent eclosion rates of 90% or higher should be achievable. Under the current conditions of the rearing system, there are two collections of pupae, one at 23 days after the egg seeding date and a second one on day 25 by separating pupae from the diet and silk. The number of pupae harvested per tray is variable ranging from less than 100 to close to 1 000. This variation may be due to lower eclosion rates that are observed on occasion or because of variable numbers of egg being seeded.

Pupae eclosion rates are generally high ranging from 85% to 95%. While pupal harvest numbers are highly variable the performance of this rearing system suggests that high rates of production are possible with this diet and tray combination. The number one recommendation will be to develop methods to achieve more consistent production on a per tray basis.

A laboratory-based flight ability test is now in use and an outdoor field cage system is being constructed for use as a test arena to assess sterile moth competitiveness such as



attraction to pheromone, longevity and mating performance.

There is no technical reason that a Chilean programme could not be developed to produce several millions of *Lobesia* per week in a manner similar to other successful SIT moth mass-rearing programs such as for the pink bollworm, codling moth, the false codling moth and others. The scope, size and investment needed for a larger facility will be dependent on future *Lobesia* programme goals and the areas where SIT releases would be made. For example, development of an SIT capacity intended solely for an urban Santiago suppression programme would require less moth production capacity than a larger programme that would also include grape crops and perhaps other crops at risk. Strategic decisions about how to best make use of an SIT capacity will depend on these factors and others such as the extent of mating disruption used within the programme.

### Supporting Area-Wide Tsetse and Trypanosomiasis Management to Improve Livestock Productivity - Phase IV (RAF5080)

#### Regional Training Course on Georeferenced Fly-Rounds for Cost Effective Entomological Surveillance of Savannah Tsetse Species, Arusha, United Republic of Tanzania, 29 October–2 November 2018



Preparation of the new system to conduct a tsetse survey (left), installed on the vehicle for survey (right).

A regional training course on Georeferenced Fly-Rounds for Cost Effective Entomological Surveillance of Savannah Tsetse Species was hosted by the Vector and Vector Borne Disease Research Institute of the United Republic of Tanzania. Fourteen participants from 9 tsetse affected countries attended the theoretical and practical lessons and demon-

stration of the system conducted to survey *Glossina swynnertoni* in the Tarangire National Park.

After the training was completed, the participants expressed strong interest to use this new tool for entomological surveys in large areas in their respective countries in the 2019 and discussed different possibilities for the optimisation of the system.

#### Regional Training Course on Molecular Tools in Tsetse Intervention Programmes, Bobo-Dioulasso, Burkina Faso, 19–30 November 2018

The Regional Training Course, was hosted by the 'Centre International de Recherche-Développement sur l'Élevage en Zone Sub-humides' (CIRDES), Bobo Dioulasso, Burkina Faso and was attended by 18 participants from nine countries (Burkina Faso, Chad, Ethiopia, Kenya, Senegal, Uganda, United Republic of Tanzania, and Zambia) and two lecturers from Belgium and France. The workshop aimed to use the molecular tools in tsetse species identification, Trypanosome detection and identification and determination of tsetse population genetics.



Participants of the Regional Training Course on Molecular Tools in Tsetse Intervention Programmes (Bobo-Dioulasso, Burkina Faso).

The first week focused on the extraction of DNA from whole tsetse flies followed by measuring the DNA quality and quantity using spectrophotometer (nanodrop) and run PCR with several primer pairs for tsetse species identification. In addition, DNA was extracted from trypanosome infected blood on tissue paper and the abdomen of trypanosome-infected tsetse flies was conducted followed by PCR for trypanosome identification. The second week focused on the partial extraction of DNA for tsetse population genetics using Chelex method, followed by PCR for four different microsatellites each marked with different dye. The generated data was examined and used for the population genetics analysis.

During the second week the workshop was evaluated by the participants, who rated the course as very good, however many of them expressed that the time was short, in particular for the genetic population topics. The closing session was attended by Ms Valentine Yapi-Gnaore (DG-



CIRDES), who appreciated the collaboration with the IAEA for selecting CIRDES to organize this workshop. During the closing session, participants expressed their appreciation of both the IAEA and CIRDES for the organization of this workshop and its effectiveness.

## Managing and Controlling *Aedes* Vector Populations Using the Sterile Insect Technique (RAS5082)

### Workshop on Recent Developments and Impact Assessment of Sterile and Incompatibility Insect Techniques for Mosquito Control, Singapore, 4–8 June 2018

In the context of the IAEA TC RAS5082 project, a workshop was organized in Singapore on ‘Recent developments and impact assessment of sterile and incompatibility insect techniques for mosquito control’. Thirty-three participants from 15 countries (Bangladesh, Cambodia, China, Fiji, Indonesia, Lao PDR, Malaysia, Myanmar, Nepal, Pakistan, Philippines, Singapore, Sri Lanka, Thailand and Viet Nam) as well as three experts (Australia, United Kingdom and WHO) attended this meeting.



*Participants of the IAEA TC RAS5082 Workshop on ‘Recent Developments and Impact Assessment of Sterile and Incompatibility Insect Techniques for Mosquito Control’ (Singapore).*

The workshop focused on the recent developments on sterile insect technique (SIT) and incompatibility insect technique (IIT) for the population suppression of mosquito vector species with an emphasis on *Aedes* mosquitoes transmitting dengue, chikungunya, Zika and yellow fever. In addition, the workshop focused on determining the impact associated with the implementation of SIT-based approaches (SIT, SIT/IIT) toward a risk assessment framework which will allow scientists, managers, regulators and national authorities to develop a systematic decision process from research through to deployment releases of sterile male mosquitoes for population suppression programmes.

## Assessing the Sterile Insect Technique for Malaria Mosquitos in a South African Setting, Phase II (SAF5014)

The South African national project has advanced significantly as the team continues to accomplish the necessary steps to prepare for first sterile male releases in Kwazulu-Natal in 2019. A national stakeholder meeting on the mosquito SIT project was held from 29 October to 2 November in Johannesburg, where the recent developments, and next steps in the project were presented and discussed. Operational funding has been secured for the coming year with strong support from South African stakeholders.



*Completed Anopheles mass rearing facility with part of the SIT team and stakeholders, Johannesburg, South Africa.*

The world’s first *Anopheles* mass-rearing facility with the capacity to produce 1 million sterile males/week of the genetic sexing strain GMK (based on the IPCL developed ANO IPCL1) has been completed and is due to be running at 250 000 sterile male/week for pilot studies by mid-2019. Alternative strains are under evaluation for improved male production capacity. Treatment protocols for female elimination and appropriate irradiation doses for sterile male releases have been established.

Field monitoring activities including 3 mark-release-recapture studies were completed in the designated pilot sites. Further tests for the improvement of sterile male transportation to the field sites, in 2019, are planned. A community awareness programme has been established and has been highly successful in getting support from the communities in and around the pilot sites. All these activities provide basis for the launch of the first SIT trial for the suppression of the malaria vector *Anopheles arabiensis* in the pilot project area.

## Using the Sterile Insect Technique to Control Dengue Vectors (MEX5031)

The MEX5031 project has made good progress on handling, transport and release of sterile male *Aedes* mosquitoes. The project has been implemented by the Regional Centre for Public Health Research (CRISP).



*SIT male release in Ejido Hidalgo (top) and drone demonstration to the local community (bottom).*

Information gathered, so far, allowed the project counterparts to develop their own holistic protocol, including entomological and epidemiological parameters to fully evalu-

ate the impact of SIT in the selected areas regarding the mosquito population and disease transmission.



*Daily activities performed by the counterpart in Tapachula, Chiapas Mexico, regarding rearing (top) and trap monitoring (bottom).*

For the last two months, the Mexican counterparts already released more than 1 million sterile male mosquitoes using a local strain of *Aedes aegypti* in Ejido Hidalgo. Drone release are also being evaluated, as alternative for the laborious and time-consuming ground releases. The Mexican counterparts continue to monitor the mosquito population using ovitraps and adult traps. Colony for egg production is being scaled up to reach the sterile male needs.



## Coordinated Research Projects (CRPs)

Project Number	Ongoing CRPs	Project Officer
D4.10.25	Dormancy Management to Enable Mass-rearing and Increase Efficacy of Sterile Insects and Natural Enemies (2014-2019)	Rui Cardoso Pereira
D4.20.16	Comparing Rearing Efficiency and Competitiveness of Sterile Male Strains Produced by Genetic, Transgenic or Symbiont-based Technologies (2015-2020)	Kostas Bourtzis
D4.40.02	Mosquito Handling, Transport, Release and Male Trapping Methods (2015-2020)	Rafael Argiles
D4.10.26	Improved Field Performance of Sterile Male Lepidoptera to Ensure Success in SIT Programmes (2016-2021)	Marc Vreysen
D4.30.03	Integration of the SIT with Biocontrol for Greenhouse Insect Pest Management (2017-2022)	Andrew Parker Carlos Cáceres
D4.20.17	Improvement of Colony Management in Insect Mass-rearing for SIT Applications (2018-2023)	Adly Abd Alla Carlos Cáceres
	<b>New CRP</b>	
D4.10.27	Assessment of Simultaneous Application of SIT and MAT to Enhance <i>Bactrocera</i> Fruit Fly Management (2019-2024)	Carlos Cáceres Rui Cardoso Pereira
D4.40.03	Generic Approach for the Development of Genetic Sexing Strains for SIT Applications (2019-2024)	Kostas Bourtzis

### Third RCM of the CRP on *Comparing Rearing Efficiency and Competitiveness of Sterile Male Strains Produced by Genetic, Transgenic or Symbiont-based Technologies*. 18–22 June 2018, Bangkok, Thailand

The Third Research Coordination Meeting was held at the Regional R&D Training Center for Insect Biotechnology (RCIB), Department of Biotechnology, Faculty of Science, and The Salaya Pavilion Hotel, Mahidol University at Salaya Campus, Thailand. The meeting was attended by 27 scientists from 13 countries (Argentina, Australia, Brazil, China, Germany, Greece, Guatemala, Italy, Mexico, Panama, Switzerland, Thailand and United States of America).

Twenty-one scientific presentations on the development and/or refinement of male-only strains as well as on the quality of sterile males produced by classical genetic, transgenic or symbiont-based technologies were presented and reviewed. High quality male-only strains are urgently needed to address the increasing demand for environment-friendly and sustainable integrated pest management approaches with a sterile insect technique (SIT) component to

control populations of insect pests of agricultural, veterinary or human health importance. Emphasis is particularly given to the rearing efficiency and male mating competitiveness, which are critical parameters as concerns the quality of male-only strains.



Participants of the Third Research Coordination Meeting on *Comparing Rearing Efficiency and Competitiveness of Sterile Male Strains Produced by Genetic, Transgenic or Symbiont-based Technologies* (Bangkok, Thailand).



In addition, the CRP has been assessing potential genetic instability and/or horizontal transfer phenomena towards the use of strains developed by classical genetic, transgenic or symbiont-based approaches for SIT applications.

### **First RCM of the CRP on *Improvement of Colony Management in Insect Mass-rearing for SIT Applications*. 23–27 July 2018, Vienna, Austria**

The first RCM was held at the Vienna International Centre, Vienna, Austria. Twenty participants from seventeen countries attended the meeting together with eight observers. The first two days of the meeting were devoted to presentations whereas during the remainder of the meeting the participants revised the CRP document prepared by the consultants, revised the logical framework based on the available expertise of CRP selected participants and prepared the individual work plan for the next 18 months. The discussion was conducted in three groups: the first focused on fruit fly strain management, the second focused on tsetse fly colony management and the third focused on pathogens diagnosis and management. In addition to the discussion in the working groups, a plenary discussion was held with insect pathologists and insect colony managers.



*Participants of the first RCM on “Improvement of Colony Management in Insect Mass-rearing for SIT Applications” (Vienna, Austria), during a visit to the new IPCL in Seibersdorf.*

The participants discussion recommended the use of meta-genomic and meta-transcriptomic for insect symbiont and pathogen identification. The CRP participants discussed, as well, the need to train participants on methods to assess the genetic diversity in the insect colonies, therefore the first workshop titled ‘Genetic Diversity Analysis and Colony Management’ is planned to be held in conjunction with the second RCM.

The participants of the RCM discussed the date and the venue for the next RCM and it was decided that the second RCM will be held on 6–10 May 2020, in Guatemala.

### **Third RCM of the CRP on *Mosquito Handling, Transport, Release and Male Trapping Methods*. 12–16 November 2018, Juazeiro, Bahia, Brazil.**

The Collaborating Centre Moscamed Brasil hosted the Third RCM of the CRP on Mosquito Handling, Transport, Release and Male Trapping Methods, which was attended by 22 participants and observers.



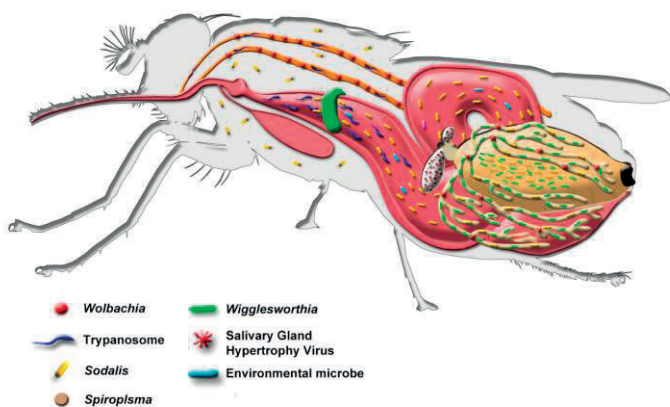
*A demonstration of mosquito aerial releases by drone that took place during the CRP on Mosquito Handling, Transport, Release and Male Trapping Methods (Juazeiro, Bahia, Brazil).*

Good progress has been made in the last 18 months, including a novel method for marking large numbers of sterile males with fluorescent dye with no impact on the quality of insects, assessment of optimal conditions for short and long-distance shipment of pupae and adult mosquitoes, development of mosquito release machines for remotely piloted aircraft systems (drones), smart traps for *Aedes* mosquitoes and resting traps for *Anopheles*. Live demonstrations of releases of mosquitoes with drones and quality control devices were highly appreciated by the participants.

## Special Issue on Enhancing Vector Refractoriness to Trypanosome Infection

Sleeping sickness or Human African Trypanosomiasis (HAT) affects people in approximately 37 countries in sub-Saharan Africa and is caused by *Trypanosoma brucei gambiense* and *Trypanosoma brucei rhodesiense*. *T. b. gambiense* is responsible for about 95% of the chronic cases of HAT in Central and Western Africa, whereas *T. b. rhodesiense* causes the acute form of HAT in Eastern Africa. The parasites are transmitted by blood-feeding tsetse fly species belonging to the genus *Glossina*. Other *Glossina*-transmitted trypanosomes also infect cattle and cause a disease called Nagana, a Zulu word meaning ‘to be depressed’. Nagana or African Animal Trypanosomiasis (AAT) results in millions of Euros of economic losses to countries that can ill afford such losses.

The management of Nagana, based on the recurrent treatment of livestock with trypanocidal drugs is costly and not sustainable due to increased resistance of the parasites. In attempts to develop more sustainable approaches to the management of AAT in Africa, several governments adopted the sterile insect technique (SIT). This technique, when integrated with other suppression tactics, has been successful in eradicating a population of *Glossina austeni* on the Island of Unguja (Zanzibar), Republic of Tanzania and in almost eradicating a population of *Glossina palpalis gambiense* in the Niayes region of Senegal. It relies on limiting the reproductive capacity of the tsetse flies by releasing large numbers of mass-reared sterile males.



The tsetse fly and its associated microorganisms (Kariithi et al., 2018).

So far, only projects that have an SIT component for tsetse and trypanosomiasis control focused on AAT-infected areas have been implemented. Future projects could be designed to target human disease transmission by *T. b. gambiense*. In this context, it is imperative to take strict measures to ensure that released sterile tsetse flies cannot transmit the trypanosome parasites that are causing the disease. Therefore, development of tsetse fly strains refractory to trypanosome infections is highly desirable as a simple and effective method of ensuring vector incompetence of the released flies.

Coordinated Research Projects (CRP) are a very effective tool to bring together scientists from all over the world to work on solving important problems in FAO and IAEA Member States. The SIT is an environment friendly control tactic to manage insect pests within area-wide integrated pest management approaches. Five years ago, a CRP titled ‘Enhancing Vector Refractoriness to Trypanosome Infection’ was initiated under the auspices of the FAO/IAEA Joint Division of Nuclear Techniques in Food and Agriculture. This FAO/IAEA CRP included 23 scientists from 19 countries, representing a broad range of expertise, to gain a deeper knowledge of the tripartite interactions between the tsetse fly vectors, their symbionts, and trypanosome parasites and to acquire a better understanding of mechanisms that limit the development of trypanosome infections in tsetse and how these may be enhanced. The studies involved detailed investigations into the biology of the insect in relationship to the causative trypanosomes, parasites, and symbionts, as well as epidemiological investigations of the disease in various parts of Africa. The scientists convened four times at about 18-month intervals to report their findings and to coordinate their research.

The CPR results have been recently published in a special issue of ‘BMC Microbiology’ <https://bmcmicrobiol.biomedcentral.com/articles/supplements/volume-18-supplement-1>. This special issue comprises the final research results of the CRP, compiled in 19 research papers, together with an introductory review paper highlighting the objectives and the main achievements of the CRP and three reviews to provide background to the project. We believe that this issue in BMC Microbiology provides an important contribution to our knowledge on tsetse flies, their symbionts, parasites, and pathogens. It will provide extensive information to the insect pathology and symbionts community about the recent advances made in this field, which in turn will help solve this important problem that continues to affect both humans and cattle in sub-Saharan Africa.

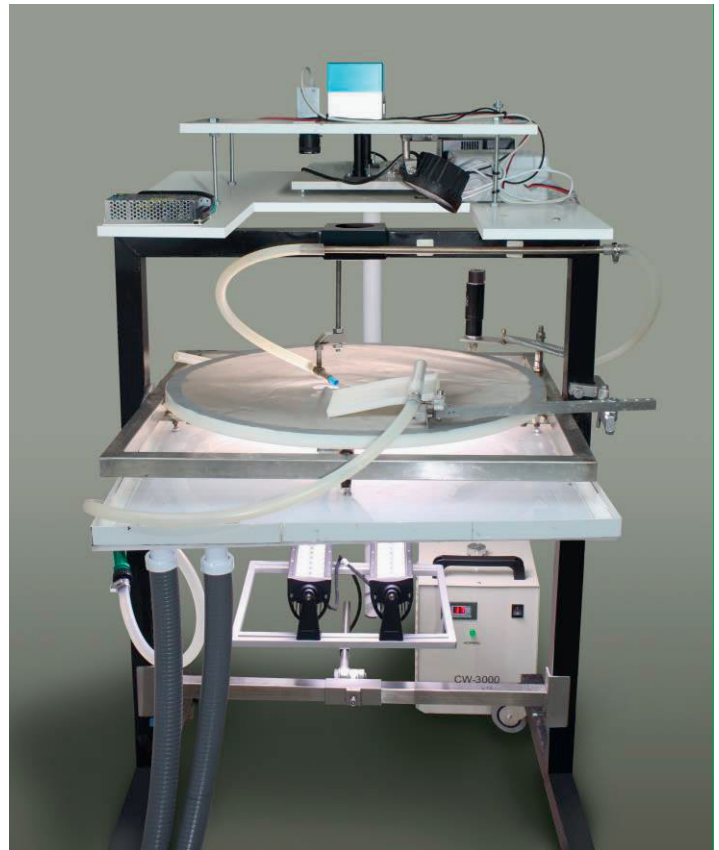


## Special Issue on Exploring Genetic, Molecular, Mechanical and Behavioural Methods of Sex Separation in Mosquitoes

Mosquitoes, and particularly some species belonging to the *Aedes* and *Anopheles* genera, transmit major human pathogens causing human diseases such as malaria, dengue, chikungunya, Zika and yellow fever. In the absence of efficient drugs and vaccines, current control methods are largely based on larval source reduction and the use of insecticides. However, these methods are also inefficient, and also not sustainable. Therefore, there is an urgent need of innovative strategies to suppress mosquito vector populations.

Several novel *Aedes* and *Anopheles* mosquito population suppression strategies, including the Sterile Insect Technique (SIT), depend on the mass production and release of sterile male mosquitoes. Male-only releases, is a key factor since female mosquitoes can bite, blood-feed and potentially transmit major human pathogens such as *Plasmodium*, dengue, Chikungunya, Zika and yellow fever.

However, currently available sex separation methods are largely inefficient to eliminate females and to ensure safe and bio-secure releases of sterile males only. To bridge this gap, the Insect Pest Control Subprogramme of the Joint Division of Nuclear Techniques in Food and Agriculture of the Food and Agriculture Organization (FAO) and the International Atomic Energy Agency (IAEA) initiated and completed a five year long coordinated research project (CRP) titled “Exploring Genetic Molecular, Mechanical and Behavioural Methods of Sex Separation in Mosquitoes”. Nineteen scientists from 14 countries participated in this CRP project, creating a critical mass of expertise required to address the challenge for the development of alternative and potentially robust and sustainable methods for mosquito sex separation. In the frame of the CRP, significant progress was achieved, accompanied by the development of genetic sexing strains, as well as mechanical and other sex separation methods aiming to eliminate females and achieve male-only releases.



*A prototype used as sorting system for different species of mosquitoes (Zacarés et al., 2018).*

Strains and methods developed, now need to be validated under mass rearing conditions and in small scale pilot trials, prior to their integration and use in large scale operational programmes. The scientific result of this CRP is presented in a series of review and original research articles in a special issue of the peer-reviewed scientific journal *Parasites and Vectors*

(<https://parasitesandvectors.biomedcentral.com/articles/supplements/volume-11-supplement-2>).



# Developments at the Insect Pest Control Laboratory (IPCL)

## INSECT GENETICS AND MOLECULAR BIOLOGY

### Isolation of recombination suppressors in *Aedes aegypti*

In the previous newsletter we referred to the importance of recombination suppressors in the construction of genetic sexing strains (GSS) using classical genetic approaches. The restriction (and even better the elimination) of recombination in male mosquitoes is crucial for the development of a genetically stable and sustainable sexing strategy, especially since recombination in male *Aedes aegypti* occurs in similar frequencies as in females.

Following irradiation with a dose of 30 Gy of 24-30 h old male pupae, a series of genetic crosses was carried out to isolate inversions that suppress recombination in chromosome pair I, especially the chromosome that harbours the M factor. Among the isomale lines retrieved, at least one line exhibited the desirable properties. In this line, the recombination was suppressed in the genomic area harbouring the M factor and neighbouring selectable markers. This line has been stable for more than 5 generations and has reduced initial recombination frequency about 10 times. Further characterization of this line is ongoing, including genetic stability and quality control tests.

This line is very promising; as it can increase the stability of genetic sexing strains that are based on the genetic linkage of selectable markers with the M chromosome. Such markers can either be naturally present in this genomic area or be transferred there following the induction of chromosomal translocations and/or inversions. This line, and inversions in general, can be regarded as a possible 'component' of a package that can stabilize (or make more accurate) a genetic sexing strain. This line can be combined with genetic sexing strains developed by other laboratories, to enhance their genetic stability.

### A promising selectable marker for the development of a genetic sexing strain for *Aedes albopictus*

The IPCL has recently invested in the isolation and characterization of morphological markers for *Aedes albopictus*. Such markers could be directly incorporated in a genetic sexing strategy and/or be used in combination with mutations such as the *temperature sensitive lethal (tsl)* ones. Even if a *tsl* mutation would be available, a morphological marker that would be tightly linked with such a mutation would still be valuable; as it would enable the preservation of a *tsl* based GSS and the quick screening of the final product prior to a male only release, as in the case of the VIENNA GSS strains of the Mediterranean fruit fly.

Two approaches have been followed to isolate morphological mutations: a) ethyl methanesulfonate (EMS) induced mutagenesis and, b) screening of natural populations. The latter approach has recently delivered at least one morphological colour marker, that is evident from larval stages to adults, but more prominent in the pupal stage. The genetic characterization of this mutation shows that it is recessive with respect to the wild type allele. This line has been stable for consecutive generations, producing consistently progeny with full penetrance and expressivity of the relevant phenotype. Additional genetic crosses have shown that it is sex-linked although not tightly linked with the male determining factor.

This selectable marker is further being characterized with respect to both quality control and the possibility to incorporate it in a genetic sexing strategy. At the same time irradiation experiments will follow, aiming to create chromosomal rearrangements (inversions) that will enhance the genetic linkage of this marker with the male determining region.

### Towards the development of a SIT-based package against *Drosophila suzukii*

*Drosophila suzukii* has been recently established in several niches globally and this has escalated into a serious threat for small stone fruit production in the US and Europe. In the absence of an appropriate control method, a scientific consortium was established to serve the goals of the SUZUKILL project. The SUZUKILL project was set up following a call for a joint Austrian-French project and it has recruited researchers from ECOBIO/Rennes, LBBE/Lyon, IFFF/BOKU Vienna, and the Joint FAO/IAEA Insect Pest Control subprogramme.

The IPCL is actively participating in the SUZUKILL project through the development of an SIT package that will be initially applied in greenhouses. The IPCL is also proactively responding to potential future challenges by developing a combined SIT/IIT approach, as a back-up method. The LBBE/Lyon colleagues, working on the development of the IIT branch, have provided us with two *Wolbachia*-infected *D. suzukii* lines which were introgressed with the *D. suzukii* line originating from Italy, and that was used in the mass-rearing group to align their genetic backgrounds. In addition, a *Wolbachia*-free *D. suzukii* line was created following a treatment with tetracycline for four generations.

Several biological traits were assessed in all these lines including fertility, fecundity, pupal weight, sex ratio, cytoplasmic incompatibility (CI) expression, and potential effect of males' age on CI levels. The results provided better insight in several fitness traits and the impact of *Wolbachia*

on the biological quality of the lines. A noteworthy outcome of the quality control analysis is the high fecundity level of the *Wolbachia*-free line we created in the IPCL. The results of the fitness assays will allow us to evaluate if these lines are adequate to be employed in a combined strategy. The combined approach will be tested in IPCL with both *Wolbachia*-infected lines and at several radiation doses. The radiation dose that will confer complete female sterility will thereafter be assessed to define if it affects the biological quality of the flies and/or the male mating competitiveness. The completion of these experiments will determine if the combined approach to control *D. suzukii* is feasible and efficient.

### Identification and manipulation of gut microbiota of the olive fruit fly for the reinforcement of area-wide integrated pest management (AW-IPM) approaches with a sterile insect technique (SIT) component

As previously reported, the IPCL has been an active participant of the EU Project “Breeding Invertebrates for Next Generation Bio-Control Training Network” (BINGO). Our main objective is the characterization of the gut bacterial profile of the olive fruit fly *Bactrocera oleae*, and the manipulation of these gut microbiota and its parasitoid wasps to improve the mass-rearing of both the fly and the parasitoid toward the implementation of AW-IPM programmes with an SIT component.

To that end, several bacteria from wild populations of both the olive fruit fly and its parasitoid have already been identified using Next Generation Sequencing (NGS) of 16S rRNA gene approaches and several bacterial species have been isolated from the olive fruit fly gut using cultivable approaches in bacterial growth cultures. Our data confirmed the predominance of *Erwinia dacicola* in the olive fruit fly gut and revealed the presence of a broad gut microbial diversity that is affected by the origin of the flies among other parameters, such as the developmental stage of the fruit fly. A diverse number of genera belonging to the Enterobacteriaceae family such as *Erwinia*, *Providencia*, *Enterobacter* and *Klebsiella* were detected as well as members of the Bacilli class and the Deinococcus-Thermus and Actinobacteria phyla.

The subsequent step is currently in progress and includes the use of the gut microbiota as supplements in the insect larval diet. Several isolated bacterial cultures have been added to the fruit fly larval diet and are tested for their ability to improve insect and parasitoid productivity under small-scale rearing conditions. A variety of life history traits related to the efficiency of mass production for both the fruit fly and its parasitoid wasps, such as pupal and adult recovery, pupal weight and adult survival under stress conditions of the fruit fly and the parasitism rate of the parasitoid wasps are currently assessed under the exposure of seven different bacterial isolates.



Female olive fruit fly egg oviposition on artificial oviposition site consisted of wax covered fabric (top), olive fruit fly larvae and pupae reared in artificial larval diet containing probiotic supplements (middle), female and male *Diachasmimorpha longicaudata* parasitoid wasps (lower).

## PLANT PESTS

### Progress on the development of sterile insect technique for *Drosophila suzukii*: long distance shipments of sterile pupae

The rapid dispersal of *D. suzukii* and its subsequent economic losses, encouraged the development of different approaches of pest management. The sterile insect technique (SIT) potentially can be integrated using area-wide integrated pest management (AW-IPM) approaches to control this pest under confined environment systems such as greenhouses. Staff of the IPCL has been working on the development of the SIT package for *D. suzukii* including determination of the optimum sterilization dose, development of effective mass-rearing procedures and quality control protocols as well as the assessment of their mating behaviour. This work has been done at the IPCL or in collab-



oration with other research institutes in FAO and IAEA Member States.



*Irradiated and untreated pupae of *D. suzukii* packed in styrofoam boxes for shipments to Italy.*

The IPCL has been collaborating with the ‘Fondazione E. Mach - Dep. Sustainable Agroecosystems and Bioresources, Agricultural Entomology Unit, San Michele, All. Adige, Italy’, on a project to assess some aspect of the mating behaviour of *D. suzukii*. The focus of this project is on sperm competition and sperm displacement between sperm from sterile or fertile males. As part of this experiment irradiated and fertile pupae have been shipped weekly from the IPCL to San Michele, Italy. The pupae were shipped in styrofoam boxes that were holding ice packs to maintain a constant temperature. To assess the effect of shipping duration and packing conditions on the pupae, quality control tests were carried out at the IPCL (before shipment) and at San Michele, Italy, upon receipt of the pupae.

### **Promoting quality of irradiated *Drosophila suzukii* for SIT using cold exposures**

Mr Thomas Enriquez, a PhD student from the University of Rennes in France, has been researching the quality of irradiated *D. suzukii* after exposure to low temperatures. There are several steps in the process of mass-producing insects towards the actual field release that may compromise the quality of the insect and therefore the success of the SIT operation. Irradiation is an event that may induce somatic damage to the pupae that is partially due to oxidative stress. The antioxidant capacity of insects might be increased by conditioning before or during irradiation using e.g. a low oxygen atmosphere (hypoxia). When insects are exposed to cold temperatures they produce antioxidants and heat shock proteins to better tolerate the stress. Consequently, it was hypothesised that cold exposure of *D. suzukii* pupae before irradiation may induce antioxidant production, offering better protection from the deleterious effects of irradiation. We tested a large array of cold conditioning, but none of them significantly increased insect quality after irradiation.

Shipment of sterile insects after irradiation is another key step of an SIT programme. Generally, insects are shipped as pupae, and therefore shipping conditions need to be

adapted to prevent adult emergence during transport and before being released in the field. In addition, pupae are often shipped under cold or hypoxia conditions. However, the effects of long-term cold exposures of irradiated *D. suzukii* pupae are presently unknown. Furthermore, hypoxia is stressful in itself, and may induce injuries during long-term exposure. Therefore, the second aim of Mr Enriquez’s experiment was to test the effect of several shipping conditions (hypoxia alone, cold alone (approximately 0°C) or a combination of both treatments) on the quality of irradiated pupae of *D. suzukii*. The shipping conditions were simulated at the IPCL and treatments lasted for 24 or 48 h. None of the treatments had a negative effect on emergence rate of pupae, but after 48 h of hypoxia alone, emerged flies were not capable of flying. However, when hypoxia was combined with cold treatment, the flying rate of the treated flies was similar to that of untreated flies. In conclusion, we showed that the use of cold in combination of hypoxia seemed to be a promising method to avoid emergence of flies while preserving their quality. Supplementary experiments are planned to investigate the physiological basis of these phenotypical data.

### **The FAO/IAEA/USDA project on phytosanitary treatment**

The phytosanitary research under the USDA/IAEA collaborative agreement, “Harmonization of phytosanitary treatments for exotic fruit flies”, is currently evaluating whether low-oxygen conditioning can increase the radiotolerance of third instar larvae of *Anastrepha fraterculus* and *A. ludens* reared in mangoes. Preliminary results have shown that six hours of severe-hypoxia (~0.5% O<sub>2</sub>, ~22% CO<sub>2</sub>) or hypoxia (~5.5% O<sub>2</sub>, ~15% CO<sub>2</sub>) does not increase the survival of insects irradiated with 70 Gy of gamma rays. This research can contribute to the revision of restrictions applied by regulatory agencies to phytosanitary irradiation of fresh fruits packed under modified atmosphere.



*Mango being naturally infested by *Anastrepha ludens* for phytosanitary irradiation research.*

Research comparing cold tolerance of four morphotypes of the *A. fraterculus* complex at 1.1°C for up to 15 days using

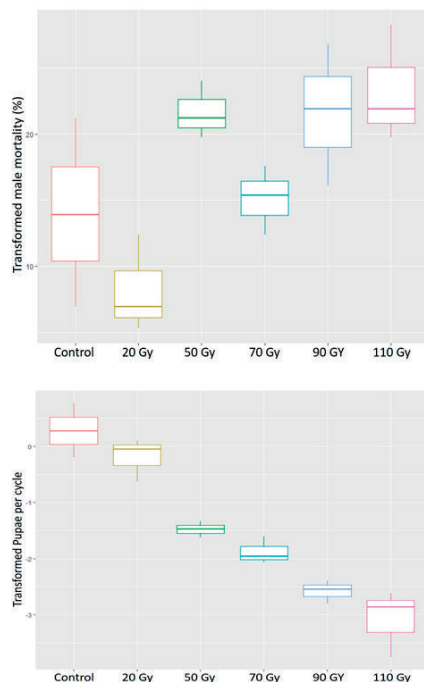


naturally infested nectarines has shown differences in cold tolerance only at sub-lethal doses (3-10 days). Cold treatment at 1.1°C for 15 days seems to be effective against most morphotypes of the *A. fraterculus* complex. This result can help guide regulatory decisions to proactively avoid any resulting trade barriers from the description of new species out of the *A. fraterculus* complex.

## LIVESTOCK PESTS

### Impact of ionizing radiation of tsetse fly fertility

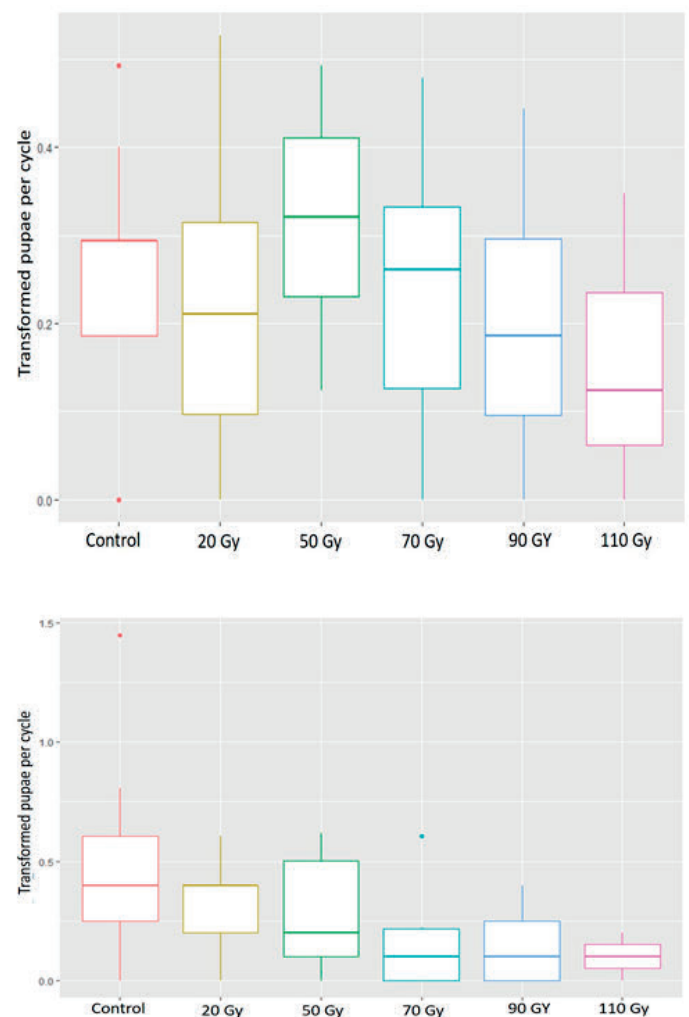
The sterile insect technique was successfully used to eradicate a population of the tsetse fly *Glossina austeni* from Unguja Island, Zanzibar, Republic of Tanzania in 1997. Thereafter, there were several attempts to implement the SIT in several African countries in mainland Africa i.e. Ethiopia and Senegal. However, due to the special biology of the tsetse, i.e. their low reproductive capacity and being hematophagous, the rearing of tsetse flies in large numbers represents a significant challenge. The establishment of large tsetse mass-rearing facilities to produce sufficient sterile males for the implementation of the SIT requires a capital investment. Therefore, smaller projects like the eradication of a tsetse fly population (*Glossina palpalis gambiensis*) from the Niayes around Dakar, Senegal, adopted the strategy of procuring the sterile male pupae from abroad (the CIRDES in Burkina Faso, the Slovak Academy of Sciences, Bratislava, Slovakia and the Insect Pest Control Laboratory, Seibersdorf, Austria). As the sterile males are not 100% sterile, some concerns were raised with respect to the potential of introducing new genetic material in the target area, in those cases where the strain released does not originate from the release area.



Mortality of male *Glossina morsitans morsitans* exposed to increasing radiation doses (top), and pupae production of virgin females mated with the irradiated males (bottom).

To evaluate the fate of offspring from irradiated male matings with untreated females, *G. morsitans morsitans* males were exposed to various doses of ionizing radiation (20, 50, 70, 90, and 110 Gy) as 29-day old pupae. Non-irradiated male pupae served as the control. After emerged males were mated with virgin females, the mortality of the males and the productivity of the mated females were recorded. The results indicated that the survival of males was significantly reduced with irradiation doses of 50, 90 and 110 Gy in comparison with the non-irradiated control. In addition, there was a significant decrease in the productivity of mated females with increasing irradiation dose of the males.

To assess the productivity of the F<sub>1</sub> offspring, emerged males were mated with 3-day old virgin females and emerged females were mated individually with 7-day old males, the productivity and mortality of both combinations were recorded. The results indicated that productivity of females mated with F<sub>1</sub> males was not affected by the irradiation doses used for the parent, however, the productivity of F<sub>1</sub> females mated with 7-day old males was significantly reduced with increasing dose used for the parents.

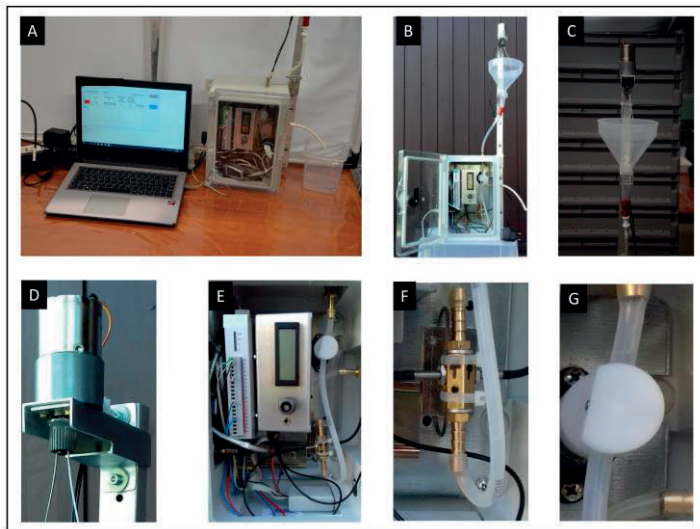


Productivity of untreated colony females mated with F<sub>1</sub> males (top), and productivity of F<sub>1</sub> females mated with 7-day-old untreated colony males (bottom).

## HUMAN DISEASE VECTORS

### The efficiency of a new automated mosquito larval counter

To achieve consistent and standardized rearing of mosquito immature stages, it is crucial to control the initial number of larvae present in each larval tray. In addition, maintaining an optimal and synchronized development rate of larvae is essential to maximize pupal production and optimize male sorting in a mass-rearing setting. Manual counting is labour intensive, time consuming and error prone. We evaluated the use of a customized automated counter for the quantification of mosquito larvae. The present prototype of the mosquito larval counter uses a single counting channel consisting of three parts: a larvae dispenser, an electronic counting unit and computer control software.



The automated mosquito larval counter. (A) General view of the larval counter, (B) general display of the electronic counting unit and the input larvae container, (C) funnel as input larvae container, (D) stirrer unit, (E) electronic counting unit, (F) optical sensor head, and (G) pinch valve.

The prototype was highly efficient to count mosquito larvae and provided repeatable and reproducible results (accuracy of ~94%) without impacting mosquito quality.



New mosquito larval diet component. From the left to the right, adult black soldier fly, mature larvae and pupae and ground powder, used to reduce the cost of mosquito larval diet.

Refinement (with a bigger larvae input container and a configuration with multiple channels) is ongoing, aiming to reduce the number of operations and ultimately speed up the process for potential use in mass-rearing settings, which requires 18 000 first instar larvae per rearing tray. This

technology could also be used in the future to evaluate larval density in the tray before pupal collection as a quality control tool to estimate mortality.

### Developing cost-effective mosquito larval diets to reduce rearing costs

Larval diet is one of the most critical and costly components of the rearing process of mosquitoes. Within the development of the mosquito SIT, a standard artificial larval diet consisting of powdered tuna meal, bovine liver powder and brewer's yeast has been developed and is currently used in the IPCL and in other laboratories. Although this reference diet adequately supplies the necessary components for larval growth including sugars, fatty acids, proteins and vitamins, the bovine liver powder is expensive and is not widely available in various Member States. Availability and affordability of diet components are of utmost importance for developing an artificial diet. Hence, alternative diet ingredients that are cost-effective and readily available in large quantity are urgently needed for mass-rearing *Aedes* spp.

Insects such as *Tenebrio molitor* (yellow mealworm), *Musca domestica* (house fly), and *Hermetia illucens* (black soldier fly) were tested for their potential to be used to feed mosquitoes. The results were promising regarding to mosquito development and quality parameters. Importantly, adding black soldier fly powder to the larval diet allowed a cost reduction up to 80% in comparison to the current larval diet.

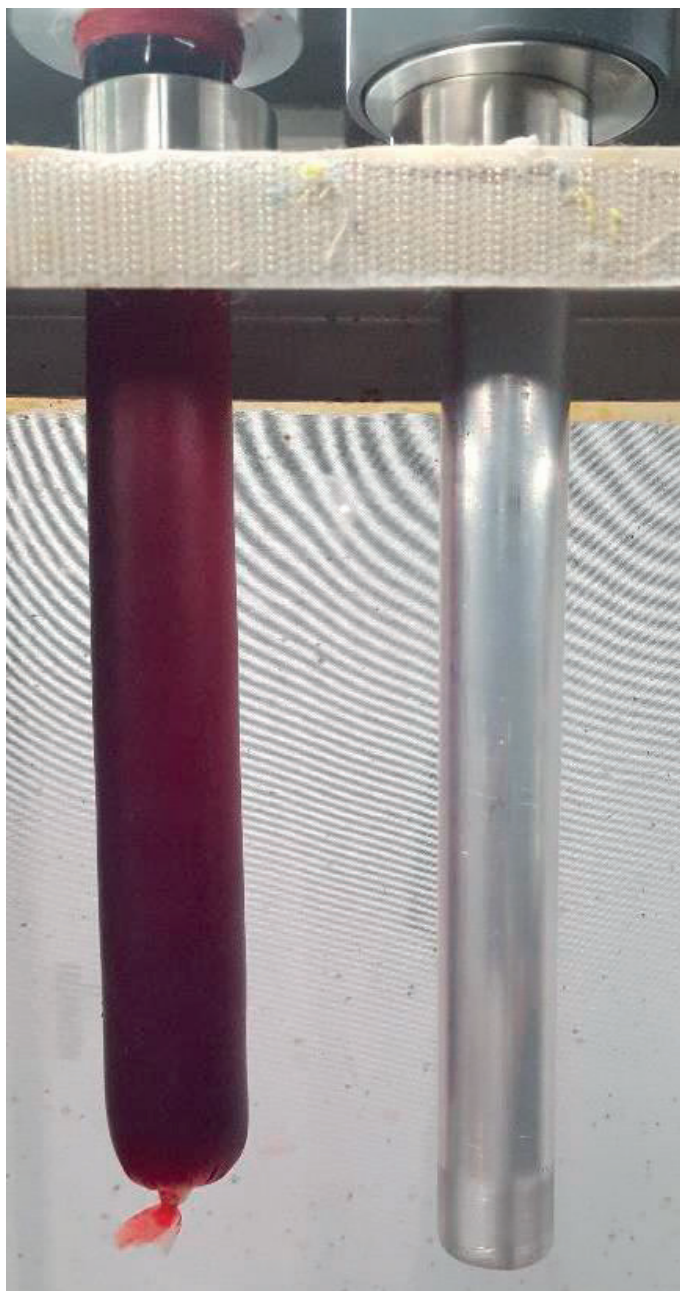
### Optimizing blood feeding of *Aedes aegypti* for mass-rearing

The sterile insect technique for mosquitoes relies on the release of large numbers of sterile males in the target area and females require blood meals for egg production. In the development of a system for mass-rearing insects, cost-efficiency is of utmost importance. The availability of high-quality blood from the slaughterhouse, and the management of the blood stock are also critical in a mosquito mass-rearing facility. Although artificial blood feeding using sausage casings has been more efficient than live animals, reducing blood quantity per sausage and exploring different ways to stock the blood such as irradiation and freezing are needed.

Different types of blood (pig, bovine) and different ways of processing the blood (fresh, frozen, irradiated) were tested in terms of fecundity, fertility and adult female survival. In addition, a sausage heating device was developed and tested in terms of reducing the quantity of blood per sausage. The device is made of aluminium material and can be plugged to a Haemotek heating device to maintain a constant temperature. Female *Aedes aegypti* had a better fecundity when fed with fresh pig blood as compared with the other blood types and processes. We achieved up to a 2/3 reduction of the blood that was previously necessary to



feed a mass-rearing cage stocked with 12 000 females using sausage casings.



*Sausage heating device developed at the IPCL to reduce the amount of blood necessary to feed a mass-rearing cage.*

The use of the sausage heating device would reduce the quantity of blood needed and thus the cost in a rearing facility. However, more work is needed to mass-rear mosquitoes using long-term stored blood without compromising egg production, and female survival.

### Identifying factors affecting the radio-sensitivity of mosquito pupae

Standardizing methods for inducing sexual sterility in mosquitoes following radiation exposure is essential to obtain reliable and reproducible results in the frame of the SIT. Numerous factors affect the physical and biological effects during irradiation procedures. Some have been identified

and characterized while others still need careful assessment.

It has been shown that older pupae (or life stages) are more resistant to radiation than younger ones. To see the extent of the relationship between age and effect, five groups of *Aedes aegypti* pupae with increasing ages were irradiated simultaneously at a fixed dose and induced sterility, and their longevity was assessed. There was a strong negative correlation between pupal age and radio-sensitivity even when age differences were small, with a 6% difference in residual fertility when comparing the youngest (10-24 hours) and oldest age groups (45-50 hrs) exposed to 40 Gy. Longevity of adult sterile males was also affected by age at which they were irradiated. Irradiating males with 40 Gy at ages less than 24 hrs reduced their mean longevity by 40% compared to untreated controls, while those irradiated at ages over 42 hrs survived slightly longer (25%) than controls.

The size of the *Aedes aegypti* pupae did not affect induced sterility. Small pupae (<0.900mm) and large pupae (>1.100mm) of the same age and cohort were irradiated simultaneously at a diagnostic dose of 30 Gy. The mean induced sterility in small and large pupae was 76.5% and 75.9% respectively.

Handling and sample preparations for pupae irradiation can significantly alter the biological responses, especially when the atmospheric conditions are changed. Submerging pupae in water during irradiation induced changes in the level of dissolved oxygen as pupae continue to absorb oxygen under water. Lowering oxygen levels creating a hypoxic environment changes cellular response and provides radioprotective effects. Pupae irradiated in such an environment suffer less damage and therefore show a higher residual fertility than pupae irradiated in air. Experiments comparing induced sterility following irradiation of pupae in water and in air confirmed the reduced radio-sensitivity in 3 mosquito species - *Aedes aegypti*, *Ae. albopictus*, and *Anopheles arabiensis*.

Several additional factors which may impact dose-response are under investigation and will provide important information for the development of standardized protocols for irradiating mosquitoes in the frame of the SIT.

### Use of stable isotopes in mosquito mating studies

Stable isotopes can be useful tools to study the mating behaviour and the fate of sperm in mosquito mating studies. Two groups of males such as sterile and fertile males, or males of different strains can be marked with stable isotopes such as  $C^{13}$  and  $N^{15}$ , and following a period of mating, the presence (or absence) of these isotopes can be detected in female spermathecae. Previous studies have indicated that virgin female mosquitoes can accept sperm from 2 males if the two mating occurs within a 20 minutes time frame. Therefore, further experiments were designed to ascertain whether sperm from sterile and fertile males are



separated or mixed in spermathecae, and whether sperm selection (for the 'better' fertile sperm) occurs in double-mated females. Preliminary conclusions are that sperm distribution and use seems random, and there is no evidence of a mechanism for sperm selection. Essentially, the first male that finds and mates the female will father future offspring. If both sterile and fertile males mate with a female within a 20-minute window, both fertile and sterile sperm will be used and will result in intermediate sterility of eggs.

This information provides important insight in *Aedes* mosquito mating behaviour which will assist in the improvement of sterile male release strategies.

### **Novel quality control tool adapted to *Anopheles arabiensis***

In the last newsletter we reported on the development of a simple flight ability test that can accurately predict the quality of male *Aedes aegypti* and *Ae. albopictus*.

However, there is currently no standardized method of assessing the quality of male *Anopheles arabiensis* mosquitoes and we tested the device developed for *Aedes* species in a series of preliminary experiments. It became apparent that the diameter of the individual flight tubes was too narrow for the slightly larger body size of male *An. arabiensis*. We therefore enlarged the width of the tubes to 1 cm of internal diameter. Initial testing was successful and subsequently we have manufactured a set of 10 new flight ability devices for *An. arabiensis*.

Validation experiments are currently underway with male *An. arabiensis* exposed to stress treatments including irradiation, chilling and compaction. The protocols were similar to those used for the validation experiments with *Aedes* species earlier this year. Survival and insemination results will be used as reference methods and compared to the flight ability results. It is hoped that this tool will prove to

be a useful tool to measure quality in other important human disease vectors. An update will be given in the next newsletter.



*A prototype flight test device for Anopheles arabiensis.*

## Reports

### Annual Meeting of the EU Project “Breeding Invertebrates for Next Generation Bio-Control Training Network” (BINGO), 6–9 July 2018, Capri, Italy

The final annual scientific meeting of the EU project BINGO took place at the Expo Convention Centre, Naples, Italy and at ‘Villa Orlandi’ Congress Center, Anacapri, Capri, Italy on 7–9 July 2018. BINGO is an EU-funded Innovative Training Network (ITN) in the frame of Marie Skłodowska-Curie Actions, which aims to advance current knowledge in biocontrol practice. In the frame of BINGO, 13 early stage researchers (ESRs) have been recruited as PhD students and trained in biocontrol practices, through a variety of scientific and technological platforms in different universities, research institutes and international organizations from eight countries.



Participants of the final annual meeting of BINGO (Breeding Invertebrates for Next Generation BioControl Training Network) held at ‘Villa Orlandi’ Congress Center (Anacapri, Capri, Italy).

On the first day, a BINGO workshop on ‘Biological Control and Integrated Pest Management’ took place as a separate session of the XI European Conference on Entomology. The workshop consisted of the following scientific presentations: ‘Improving the genetics of natural enemies for biological control with lessons not yet learned from the last century’ (Richard Stouthamer), ‘Trends in biological control’ (Jacques Brodeur), ‘An increased need for research on biological control’ (Tom V.M. Groot), ‘Using genetics to improve biological control’ (Bart A. Pannebakker), ‘Selective breeding of the biological control agent *Orius laevigatus*: biotic potential and tolerance to environmental stress’ (Pablo Bielza), ‘Is vegetarianism in the genes? Investigating the genetic basis of phytophagy in *Nesidiocoris tenuis*’ (Milena Chinchilla-Ramirez), ‘Effect of mass-rearing on the performance and the genetic diversity of the predatory mite *Amblyseius swirskii* Athias-Henriot (Acari: Phytoseiidae)’ (Angeliki Paspatis), ‘Intrinsic competition

between two European egg parasitoids of the brown mar-morated stink bug’ (Judith Stahl) and ‘*Trichogramma* populations in organic German cabbage fields – a genetic, ecological, and behavioural overview’ (Sophie R. Chattington). The following three days of the BINGO meeting were hosted at ‘Villa Orlandi’ Congress Center, Anacapri, Capri, Italy. During these days, Giuseppe Saccone delivered a scientific speech on ‘Changing sex of agricultural pests: from basic studies to genetic control by gene editing and more’ and all thirteen ESRs presented the progress of their research projects followed by Q&A and discussions about each project. Finally, the overall progress of the deliverables and milestones of BINGO project, the ESR training progress as well as the overall dissemination of the BINGO scientific findings were thoroughly discussed, and a preliminary discussion was held related to the final deliverable report of the BINGO project, the finalization of which was planned for December 2018.

### The International Congress on Invertebrate Pathology and Microbial Control and the 51th Annual Meeting of the Society for Invertebrate Pathology, 12–16 August 2018, Gold Coast, Queensland, Australia

More than 300 researchers in insect pathology, covering microbial, fungal, microsporidia, nematode and viral diseases participated in the International Congress and the 51<sup>th</sup> Annual Meeting of the Society for Invertebrate Pathology. During this annual meeting, many research papers were presented focusing on the insect pest control in south-east Asia.

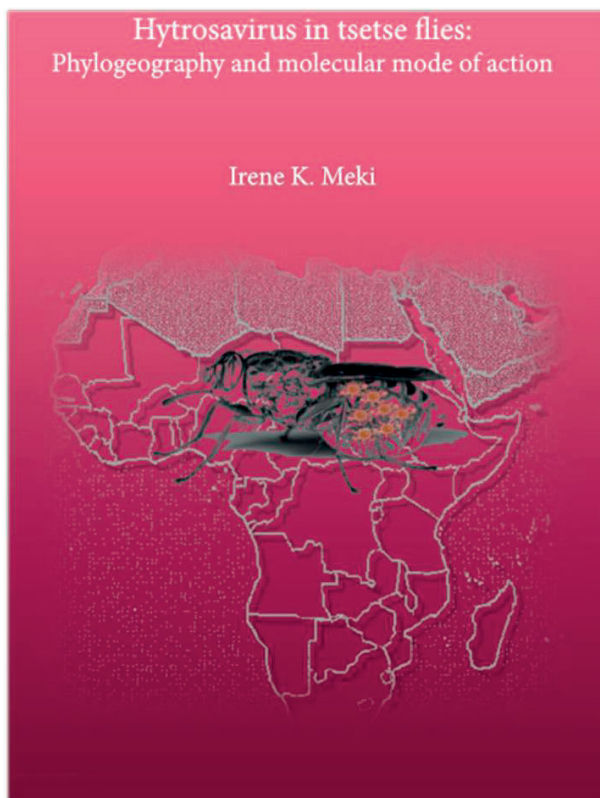
During the conference, IPC staff held a meeting with Prof Monique van Oers and Dr Vera Ros, from Wageningen University, to discuss the current and future collaboration with the IPCL and the work plan of the PhD candidate, Ms Caroline Mirieri who is currently conducting her research at the IPCL in collaboration with Wageningen University. Moreover, IPC staff met with Mr Stephen R Sharpe from the Hawkesbury Institute for the Environment, Western Sydney University who mentioned that several viruses were discovered in the Queensland fruit fly. In addition, IPC staff met with Drs Sean Moore and Craig Chambers from Citrus Research International (CRI), South Africa, who are involved in the SIT project for the false codling moth (FCM) and discussed the granulovirus infection problem in the FCM facility. Dr Moore mentioned that the virus is causing serious problems due to seasonal outbreaks. After presenting the topic of the new CRP D42017 on ‘Improvement of colony management in insect mass-rearing for SIT applications’ and its objectives and the current list of participants, Drs Moore and Chambers requested addi-



tional information on this CRP and requested the possibility to attend the next RCM as observers.

### Ms Irene K. Meki obtains her PhD from Wageningen University

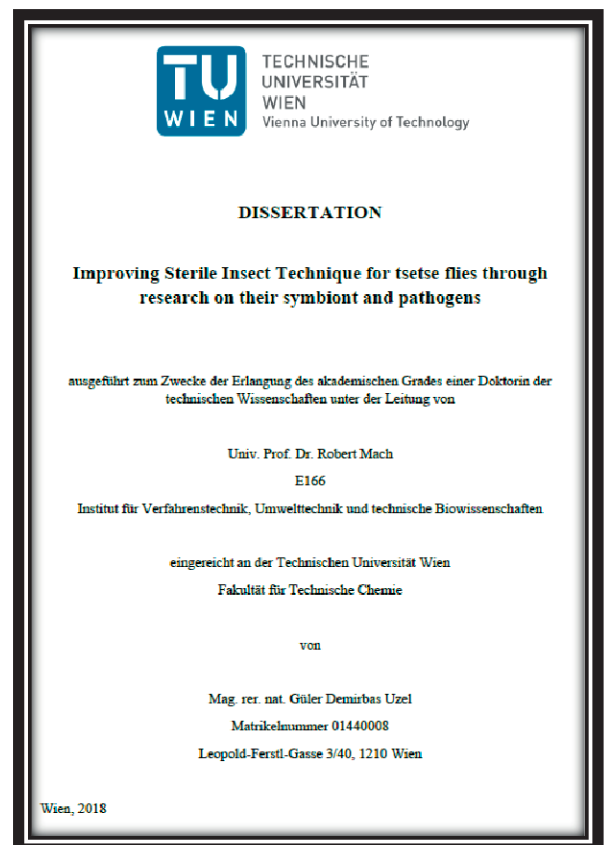
Ms Irene K. Meki from Kenya successfully defended her PhD thesis in early October 2018 in Wageningen University, The Netherlands. Irene completed her PhD research under a sandwich programme, in which she spent 80% of her time doing research at the Insect Pest Control Laboratory (IPCL) in Seibersdorf and the remaining time at the Laboratory of Virology of the Wageningen University.



Her PhD thesis is entitled 'Hytrosavirus in tsetse flies: Phylogeography and molecular mode of action', and focused on various aspects of Salivary Glands Hypertrophy Virus (SGHV)-tsetse interactions, including deciphering some of the factors that influence the outbreaks of the salivary gland hypertrophy (SGH) syndrome that is directly linked to collapse of tsetse colonies, identification of SGHV strains in wild and laboratory-bred tsetse species, elucidation of host and viral molecular mechanisms that maintain the virus in covert infection status (e.g. small RNAs). The volume and quality of the research conducted was impressive, and her research findings were published in eight scientific papers in renowned peer-reviewed journals. Irene's research has also greatly contributed in setting up a platform on which she hopes to continue. The data obtained from Irene's thesis have not only been instrumental in furthering our understanding of the pathobiology of this virus, but it also suggests new approaches that are key in the management of viral infections in insect mass-rearing for the benefit of vector-based control of insect pests.

### Ms Güler Demirbas Uzel obtains her PhD from the Vienna University of Technology

Ms Güler Demirbas Uzel from Turkey, successfully defended her PhD thesis in March 2018 at the Vienna University of Technology, Vienna, Austria. Güler did her PhD research under a sandwich programme, in which she spent 90% of her time doing her research at the Insect Pest Control Laboratory (IPCL) in Seibersdorf, under the supervision of IPCL staff Adly Abdalla and Andrew Parker, and the remaining time at the Vienna University of Technology under the supervision of Prof Robert March.



Her PhD thesis, titled 'Improving sterile insect technique for tsetse flies through research on their symbiont and pathogens', focused on various aspects of symbionts, parasites and pathogens-tsetse interactions, including assessing the impact of ionizing radiation on tsetse symbionts, the prevalence of SGHV, *Wolbachia* and trypanosome infection in wild tsetse populations. The volume and the quality of the research conducted was very impressive, and the research findings were published in eight scientific papers in renowned peer-reviewed journals. The data obtained from Güler's thesis have been instrumental in furthering our understanding of the host range of this virus, and it suggests that the virus management strategy developed to manage its infection in tsetse fly *Glossina pallidipes* should be implemented for other tsetse species even without observing the salivary gland hypertrophy. In addition, Güler's results open the door to implementing the paratransgenesis approach with the SIT of tsetse fly to release sterile males' refractory to trypanosome infection.

# Announcements

## Call for Submission of Research Proposals for a new FAO/IAEA Coordinated Research Project (CRP) on *Generic approach for the development of genetic sexing strains (GSS) for SIT applications*

The application of the Sterile Insect Technique (SIT) in area-wide integrated pest management (AW-IPM) programmes continues to increase in response to requests from Member States. These requests include the development and refinement of SIT packages for programmes to control populations of different insect pests of agricultural, veterinary and human health importance. The development and operational application of such programmes with an SIT component against insect pests and disease vectors continue to reveal research areas where new technologies could further improve efficiency and thus lead to more efficacious programmes. One such critical area, where important advances need to be made to increase the cost-effectiveness of the technique, or where it is a prerequisite before any SIT application is conceivable, concerns the development of genetic sexing strains (GSS). In SIT programmes against agriculture pests, the release of both sexes is primarily of economic concern; however, in SIT programmes against some insect disease vectors (e.g. mosquitoes), it is an essential prerequisite to release only males as females are blood feeders and may potentially transmit the diseases even if sterile. One example of how a GSS can significantly enhance SIT applicability and efficiency has been their use in the Mediterranean fruit fly, *Ceratitis capitata* AW-IPM programmes, a technology developed at the FAO/IAEA Agriculture and Biotechnology Laboratories in Seibersdorf in conjunction with the Agency's CRP programme. Using irradiation and classical genetic approaches, a series of genetic sexing strains were developed for the Mediterranean fruit fly. These are currently being used in all mass-rearing facilities producing this pest for large-scale SIT programmes, including the VIENNA 7 and VIENNA 8 strains. These GSS consist of at least two principal components: (a) a Y-autosome translocation, T(Y;A), which is required to link the inheritance of this marker to the sex and (b) a selectable marker which is necessary for sex separation or female killing. The latter include a *temperature-sensitive lethal (tsl)* gene, which is located on chromosome 5 and has useful biological properties rendering it an integral component of the new generation of Mediterranean fruit fly GSS currently used in mass rearing facilities and operational programmes worldwide. This is because, in the VIENNA 7 and VIENNA 8 GSS, *tsl*<sup>+</sup> (temperature insensitive) individuals are males while females are homozygous for the *tsl* allele and can be killed through the incubation of eggs at elevated temperatures. Despite the importance of the *tsl* marker or other morphological colour markers,

GSSs have not been developed in many SIT targeted species because the isolation of such naturally occurring mutants is a tedious, random and labour-intensive process. Furthermore, the current process to develop a GSS in one species, even if successful, is not tractable to the transfer or development of similar GSSs in other, even closely related, species. However, such mutants could now be specifically induced through new technologies. If successful, this would open the way to detect and isolate the orthologous genes in all other SIT targeted species (fruit flies, mosquitoes, tsetse flies, moths etc) and/or induce similar mutations which could then be used for the development of GSS. In other words, the successful isolation and characterization of such marker genes and their associated mutations may provide a versatile (generic) tool towards the development of GSS for SIT applications against diverse species.

**Objectives:** The main objective of this CRP is the development and evaluation of generic approaches for the construction of genetic sexing strains (GSS) to be used for SIT applications, as part of AW-IPM programs, to control populations of agricultural pests and disease vectors. In particular: (a) to develop generic strategies for the construction of GSS for SIT applications; (b) to assess the efficiency, applicability and the range of the species transferability of the generic approaches and (c) to evaluate, at small scale, GSS developed through the generic approaches.

**Main Activities:** The CRP will focus on the following: (a) to identify at least two markers which could be used for generic strategies for the development of GSS for SIT applications against targeted agricultural pests and disease vectors identified; (b) to establish and evaluate at least two strains carrying these selectable markers to be used for the development of genetic sexing strains for SIT applications against targeted agricultural pests and disease vectors; (c) to develop at least two GSS based on generic approaches for SIT applications against targeted agricultural pests and disease vectors developed and (d) to evaluate these GSS under small scale rearing conditions.

**Duration:** The expected duration of the CRP is 6 years (2019–2024) and the first Research Coordination Meeting is planned for 7–11 October 2019 in Vienna, Austria.

**Applications:** Scientists and researchers who are interested in collaborating in this new CRP should contact Kostas Bourtzis (K.Bourtzis@iaea.org) or Rui Cardoso Pereira (R.Cardoso-Pereira@iaea.org). Information on the IAEA Coordinated Research Programme and how to apply for research contracts and research agreements can be found at <http://cra.iaea.org/>. Applications should be submitted by **28 February 2019** to [research.contracts@iaea.org](mailto:research.contracts@iaea.org).



## Interregional Training Course on *The Use of the Sterile Insect and Related Techniques for the Integrated Area-wide Management of Insect Pests*, 10 June–5 July 2019, Metapa de Dominguez, Chiapas, Mexico and Antigua / El Pino, Guatemala

**Context:** Food insecurity is inherently linked to pests and diseases. The losses caused by diseases and pests at both the pre- and post-harvest levels average at 30-40% of agricultural outputs. These are hampering the efficient use of agricultural investments in land, seeds, water, fertilizer, animal feed, labour and other inputs available to feed the growing human population.

Current reliance on pesticides and drugs is not sustainable, impairing the natural balance and causing outbreaks of secondary pests, contaminating the environment and leaving residues on food commodities, and leading to the development of resistance to pesticides used.

In addition, because of increasing crop and animal movement and trade, as well as climate change, there is an unprecedented increase of invasive animal and plant pests with dire socio-economic consequences.

An area-wide integrated approach that targets the management of total populations of major pest insects, although management-intensive and logistically more complex, can contribute, in most cases to a more effective and sustainable control.

**Purpose of the Course:** The purpose of this four-week interregional course is to provide a broad overview on the application of nuclear-related techniques, within the context of area-wide integrated insect pest management programmes, to managers of insect control programmes, animal health and plant protection officials and applied research entomologists.

The course will include radiation-induced sterility, the sterile insect technique (SIT), F-1 sterility, other methods of insect control, integration of control methodologies for area-wide insect management, the biology, ecology and dynamics of pest insect populations subjected to control, economic analysis of area-wide programmes and reviews of successful and ongoing area-wide programmes with an SIT component.

The aim is to widen the knowledge and horizon of current and future decision makers to a broader list of major insect pest problems, including pests or vectors of diseases that are currently not yet established in the participants' countries.

**Participants:** The course is directed at top-level disease vector and pest control management personnel who are or will likely become high-level decision-makers and senior managers of pest control programmes or campaigns. A key aspect of this training is to train good pest control managers

in Member States with the broad background and skills required to conduct complex area-wide programmes. There is a need to transfer technology while training the required managers of projects to effectively integrate the SIT. Future decision makers need to be made aware of upcoming risks, develop a sense of preparedness and be trained on preventive and management strategies against potential new major pests and disease vectors.

**Application Procedure:** Nominations should be submitted on the standard IAEA application form for training courses (<https://www.iaea.org/technicalcooperation/How-to-take-part/train-course/index.html>). Completed forms should be endorsed by and submitted through the official channels established (either the Ministry of Foreign Affairs, the National Atomic Energy Authority, the Office of the United Nations Development Programme, the Office of the FAO Resident Representative or the Ministry of Agriculture). **(Deadline for nominations: 28 February 2019).**

**Participants' Qualifications:** The course is open to about 24 participants from IAEA and FAO Member States in all geographical regions. Preference will be given to qualified candidates from developing countries. Applicants must have at least a Bachelor of Science degree or equivalent in entomology or a related biological field. As the course will be conducted in **English**, participants must have an adequate working knowledge of that language.

The key criterion is the candidate's actual participation in operational area-wide pest control programmes or the potential when he/she has returned home to provide leadership in area-wide pest management and the use of the SIT in future programmes. Preference will be given to those in pest control policy-formulating positions or involved in preparing applied pest control programmes, or who have had at least several years of practical experience in applied research or teaching on pest control.

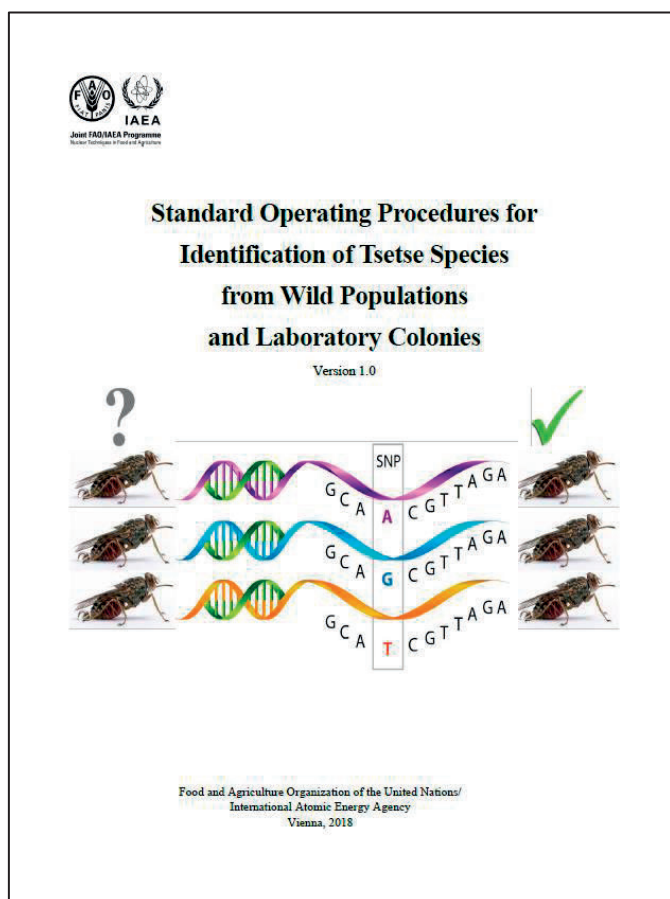
## Standard Operating Procedures for Identification of Tsetse Species from Wild Populations and Laboratory Colonies

Tsetse flies are solely responsible for the cyclical transmission of the trypanosomes that are the causative agents of sleeping sickness or human African trypanosomiasis (HAT) and nagana or African animal trypanosomiasis (AAT) in livestock. There are about 31 tsetse fly species and subspecies placed in the genus *Glossina* of the family Glossinidae. All are restricted to Sub-Saharan Africa, but only 8-10 of these species are of economic or human health importance. The genus *Glossina* is divided into three distinct taxonomic groups based on morphological characters such as external genitalia of the male flies, their habitat requirements and preferred hosts.

There are several strategies that have been applied to suppress the vectors to manage trypanosomiasis, which include the use of the sequential aerosol technique (SAT), station-

ary and mobile attractive devices, the live bait technique and the sterile insect technique (SIT). The SIT involves the production of the target insect species in large numbers in rearing factories, and subsequently males are sterilized by irradiation. This is followed by the sustained and systematic release of the sterile males over the target area in numbers large enough to out-compete the wild male population for wild females. Mating of sterile males with virgin wild females result in no offspring, which will lead to a decrease of the population of the target species with each generation.

The application of the SIT requires the mass-rearing of the correct target species. Although in many instances the colony will be established from samples from the targeted area, in other cases they might be imported from a regional facility and this would require confirmation of the correct species status. The morphological identification of especially closely related species of tsetse species has been challenging. These standard operating procedures (SOP) have been developed to enable the quick and easy identification of tsetse species using molecular techniques, thereby facilitating the implementation of the SIT against selected tsetse species in Africa.

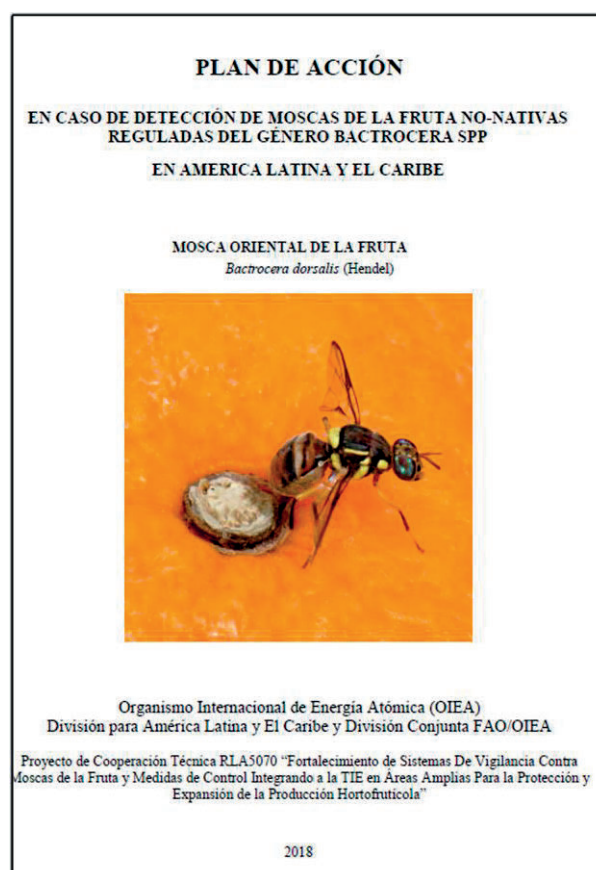


This document provides useful information to correctly identify specimens of nine tsetse flies species/subspecies (*Glossina brevipalpis*, *G. palpalis gambiensis*, *G. morsitans morsitans*, *G. m. centralis*, *G. m. submorsitans*, *G. pallidipes*, *G. fuscipes fuscipes*, *G. tachinoides* and *G. swynnertoni*) derived from field collections or laboratory colonies using molecular techniques. These SOP are in-

tended for staff involved in tsetse rearing with sufficient training or experience in molecular biology techniques such as DNA extraction, the Polymerase Chain Reaction (PCR) method, and sequencing. This procedures manual was developed based on the publication Augustinos et al., (in press). The SOP can be downloaded at: [http://www-naweb.iaea.org/Nafa/ipc/public/SOP-for-tsetse-species-identification-Final\\_8.pdf](http://www-naweb.iaea.org/Nafa/ipc/public/SOP-for-tsetse-species-identification-Final_8.pdf)

## Action Plan in Case of Detection of Non-native Regulated *Bactrocera* spp in Latin America and the Caribbean

The information contained in this Action Plan is aimed at cases when a detection or outbreak of a non-native regulated *Bactrocera* and *Zeugodacus* species is officially declared. The Action Plan should be used as a guide in the implementation of procedures for delimitation, containment and eradication of fruit fly species responding to the male attractants methyl eugenol and cuelure, to prevent its spread and introduction to other areas free of the pest.



The methods and technologies described have been validated and used in action programmes against these pests by National Plant Protection Organizations in various FAO and IAEA Member States. This is part of the efforts being conducted through technical cooperation projects, to strengthen early detection and emergency response capacity against invasive insect pests in Latin America and the Caribbean (the document is available only in Spanish at: [http://www-naweb.iaea.org/nafa/ipc/public/Plan-de-Accion-Bactrocera-spp\\_agosto2018-Final.pdf](http://www-naweb.iaea.org/nafa/ipc/public/Plan-de-Accion-Bactrocera-spp_agosto2018-Final.pdf)).



## In Memoriam

### Roger Vargas (1947–2018)

On 10 July 2018, Roger Vargas suddenly passed away, and the entomology world lost one of its greatest and most beloved scientists. Roger spent most of his career working on Tephritid fruit flies, many of which are the most economically damaging pests in the world. His areas of expertise were integrated pest management, insect ecology, biological control, crop protection, insect mass rearing, demography, trapping, and toxicology. Roger was widely regarded as the world's greatest fruit fly ecologist. His over 40-year record of research on fruit flies was truly outstanding: He published over 240 scientific papers, won many awards and was awarded patents for his work on fruit fly rearing. His research productivity was considerable, as demonstrated in a 2018 IAEA/APHIS survey ranking him # 4 worldwide for number of fruit fly publications.



He received Master's degree in Zoology from San Diego State University in 1974 and moved to Honolulu where he enrolled a PhD program at the University of Hawaii's Department of Entomology. Under the guidance of Dr Toshiyuki Nishida he received his doctorate in 1979 and was hired by the USDA's Agricultural Research Service in Manoa, Hawaii. Roger initially worked on fruit fly mass-rearing, developing new fruit fly mass rearing procedures.

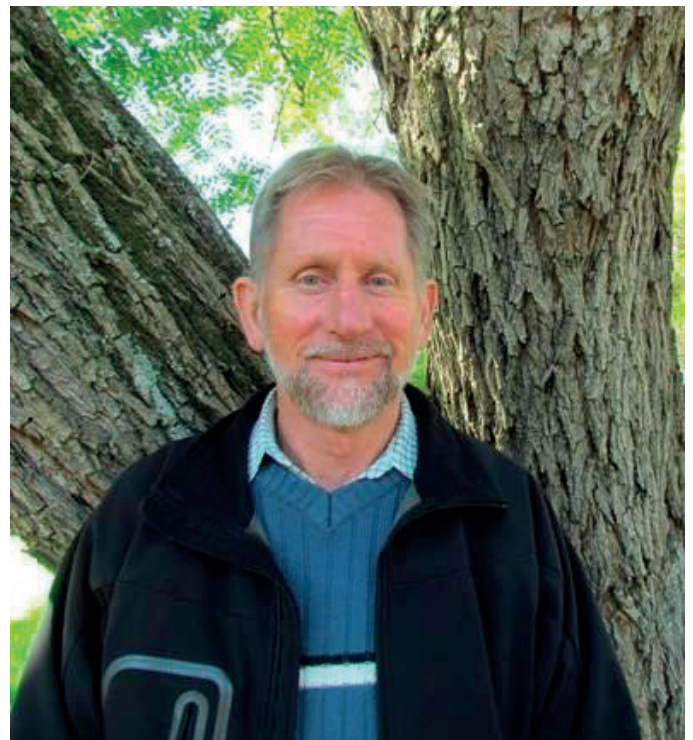
He received patents for his work on rearing, producing flies used in sterile release programmes. Roger became the Research Leader of the Rearing, Radiation, and Genetics Unit from 1986-90, after which he moved to the Big Island and worked at the USDA-ARS Hilo laboratory for the rest of his career. Roger was in great demand for his expertise and was constantly traveling around the world to work with governments on developing and improving fruit fly control, particularly in the tropical and semitropical regions where fruit flies are economically important.

*Source: Proceedings of the Hawaiian Entomological Society.*

### Tom Blomefield (1944–2018)

Tom Blomefield, Research Entomologist retired from the ARC Infruitec-Nietvoorbij Research Institute in Stellenbosch, died suddenly on 18 August 2018. He was 74.

Tom and I worked together in the Pest Management Division at Infruitec in Stellenbosch for nearly 40 years, although our paths first crossed in 1965 at the University of Natal, Pietermaritzburg.



When he arrived in Stellenbosch in 1973, Tom was writing up an MSc on ants. But ants weren't top priority pests in fruit orchards. By quirk of fate, Tom seemed to attract projects involving lepidopteran pests. Over the years most of his research was on leafroller, false codling moth, oriental fruit moth and codling moth. But Tom's signature work was on the bionomics of codling moth in apple orchards in South Africa. His years of dedication, many, many kilometres walked through orchards, thousands of leaves, twigs and fruit inspected and cut open, and the collection of vast amounts of data (Tom hated throwing anything away), culminated in his PhD. His thesis became a go-to reference work on codling moth in South Africa.

Towards the end of his career Tom became involved with an IAEA CRP on codling moth, working closely with IAEA Experts Stephanie Bloem and Jim Carpenter on the compatibility of wild and sterile moths, and the strain compatibility between South African and Canadian codling moth populations. They published on this together.

After retirement Tom developed a passion for cycling. Twice a week he and another retired entomologist reli-

giously hopped on their bikes, often covering up to 100 km at a time. Age appeared to be no barrier.

Tom was a once-off – those who knew him would know why his university professor called him ‘inimitable’. Rest in Peace Tommo. Our thoughts are with Pam, Jessica and Stephen.

Source: Brian Barnes.

## Kishor Mehta (1937–2018)

It is with great sadness that we report the death of our colleague and friend, Kishor Mehta, on the 19th October 2018. He was an internationally recognized and respected authority in radiation dosimetry with many scientific publications.



After leaving Atomic Energy of Canada Ltd, Kishor worked in the Dosimetry and Medical Radiation Physics Section (DMRPS) from July 1992 until his retirement in December 1999. Soon after his arrival in the Agency, in addition to his official duties in DMRPS he also became involved with assisting first the Food & Environmental Protection subprogramme with dosimetry for phytosanitary irradiation and then the IPCS with dosimetry for radiation sterilization of insects for the sterile insect technique. His main focus was on establishing and improving the application of practical, economical and reliable dosimetry to im-

prove the reproducibility and consistency of results in these areas.

Very aware of the potential complications of dosimetry and the lack of dosimetry skills in most insect rearing programmes, he focused on developing clear guidelines and protocols to enable all rearing programmes to use dosimetry to improve their confidence in the doses being used and to improve the consistency and comparability of their results. For this he produced a *Standard Operating Procedure for Gafchromic® film dosimetry system for gamma radiation*, which has been widely used and cited.

Recognizing the security problems with utilizing radioactive sources for insect sterilization, Kishor assisted the IPCS with identifying suitable X-ray systems as an alternative. This raised issues with calibrating these sources and Kishor spent the last few years working on characterization of X-ray sources and calibration of dosimeters for low energy X-ray. This resulted in him producing a guideline for selecting X-ray irradiators (*Technical specification for an X-ray system for the irradiation of insects for the sterile insect technique and other related technologies*).

Kishor's cheerful personality, deep knowledge and willingness to help in solving our problems will be sorely missed.

Publications with the IPCS:

BAKRI, A., MEHTA, K., LANCE, D.R., “Sterilizing insects with ionizing radiation”, 3.3 (DYCK, V.A., HENDRICH, J., ROBINSON, A.S., Eds), Springer, Dordrecht (2005) 233–268.

PARKER, A., MEHTA, K., Sterile insect technique: a model for dose optimization for improved sterile insect quality, *Florida Entomologist* **90** (2007) 88.

MEHTA, K., Radiation sources supporting the use of natural enemies for biological control of agricultural pests, *Bio-control Science & Technology* **19**, S1 (2009) 335.

MEHTA, K., PARKER, A., Characterization and dosimetry of a practical x-ray alternative to self-shielded gamma irradiators, *Radiation Physics & Chemistry* **80** (2011) 107.

MEHTA, K., PARKER, A., TESSIER, F., Gafchromic® film dosimetry for low energy X radiation, *Radiation Measurements* **67** (2014) 48.

KHOURY, H.J. et al., Alanine-EPR as a transfer standard dosimetry system for low energy X radiation, *Radiation Physics & Chemistry* **116** (2015) 147.

KHOURY, H.J., MEHTA, K., DE BARROS, V.S., GUZZO, P.L., PARKER, A.G., Dose assurance service for low energy X ray irradiators using an alanine-EPR transfer dosimetry system, *Florida Entomologist* **99** (2016) 14.

Source: Andrew Parker, FAO/IAEA.



## Other News

### Citizen Science and Asian Tiger Mosquito Monitoring and Control on Procida Island, Italy

Since September 2015 the Department of Biology of the University of Naples Federico II, in collaboration with the Insect Pest Control Section of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture and with the University of Rome La Sapienza, is conducting a research project on the Mediterranean island of Procida (Campania Region, Southern Italy) to collect baseline data on the spatial and temporal dynamic of the *Aedes albopictus* population with the aim to apply SIT to control this vector. The project, led by Marco Salvemini, has been designed with the idea to develop, as a first step, a successful protocol of community engagement to prepare the ground for the subsequent control steps, which could greatly benefit from strong stakeholders' participation. Since April 2016, local political administrators and 'citizen scientists' are collaborating with researchers of the Department of Biology, collecting data from ovitraps.



*Sterile males release day on Procida island with volunteers, students of the University of Naples Federico II and local administrators (third from the left the Procida Major, Dr. Raimondo Ambrosino and fifth from the left the councillor Dr. Rossella Lauro).*

During a one year-long monitoring program twelve volunteers were involved to manage ovitraps weekly and 40 families participated to the project, giving the access to their private properties for the placing of 101 ovitraps. In September 2018, a mark-release-recapture experiment (MRR) was performed on an area of 12.5 ha and over twenty days with the release of about 20 000 sterile males, produced by the IAEA Italian Collaborating Center in Crevalcore (Bologna) starting from eggs collected on the island in June 2018. During this test, researchers successfully improved the collaboration network on the island. About 39 families were involved to place on their properties BG-traps, ovitraps and human landing catch (HLC) stations. One family hosted a field laboratory in its garden and two families provided free accommodation to the research team, composed by eight persons, for the whole period of the study; the

Procida Major, a municipal councillor and three volunteers participated to the activities of the release days.



*Public event on Procida island with students of primary school "I.C. Capraro" during the egg counting challenge and the sterile-male catching challenge.*

During the study period, the research team distributed informative materials in the streets of the island and organized a public event involving schools and citizens (in the frame of the European Researchers Night 2018 - <http://www.meetmetonight.it/>) with the aim to increase their engagement by letting people themselves test the monitoring methods utilized in the field on the island. Two demonstrations "competitions" were organized: an egg counting challenge: with people asked to count by stereomicroscope the eggs, collected using ovitraps in the MRR area, and a sterile-male catching challenge with people asked to collect sterile males released in a big cage using electric aspirators.

The activities on the island were very successful in terms of participation of citizens and of children from primary and secondary schools and in terms of media coverage with a radio interview on Radio2 (national radio channel) and an article on the national weekly newspaper 'il Venerdì di Repubblica'. Further activities are being planned for the spring of 2019, in collaboration with science teachers of the 'I.C. Capraro' school of the island, with the idea to involve children in a door-to-door sanitization project to eliminate larval breeding sites in the private properties.

*Source: Marco Salvemini.*

## Kickoff of a French SIT Think Tank: Collectif TIS

France starts taking a serious look at the use of the Sterile Insect Technique to control health and agricultural pests.

Various parties that are involved or interested in the development of the SIT in France convened on October 2nd at the first meeting of the Collectif TIS, a think tank built to study the potential and coordinate the implementation of SIT projects on French territories.

This think tank is integrated within the Consortium Biocontrôle and the network Vectopole Sud. More than 90 participants from various expertise and field of activities (human and animal health, agriculture, environment) have followed the meeting from Montpellier and five videoconference sites (Paris, Bergerac, Sophia-Antipolis, Corse, Ile de la Réunion).



The setup and implementation of SIT requires various steps and a reflection articulated on different interconnected components: technical, regulatory, societal and ecosystemic. Through the activities of five working groups, the Collectif TIS wants to consider all these components in a concerted manner with participants from diverse backgrounds (entomologists, sociologists, agricultural federations, public authorities, biocontrol industries, environmental associations).

The enthusiastic participation of the various stakeholders and the experience-sharing between Health and Agricultural fields, have made it possible that the kick-off of the Collectif TIS was a success. The working groups have now started drawing their road maps with the goal of achieving transversal and concrete deliverables that will help implementing efficient, profitable and long-term SIT projects in French territories.

### Objectives of the Working Groups (WG):

*WG 1 - Methodology and evaluation of pilot studies:* establish transversal guidelines enabling defining the milestones and prerequisites for the implementation of small-scale pilot-studies.

*WG 2 -Evaluation of socio-ecosystemic impacts:* brainstorm and design suitable studies to assess the impact of

area-wide SIT implementation at various scales in terms of social and agricultural practices and biodiversity.

*WG 3 – Regulation and import:* work together with the relevant ministries to avoid that regulatory framework gaps or ambiguity hinder the implementation of small-scale trials or large-scale deployments.

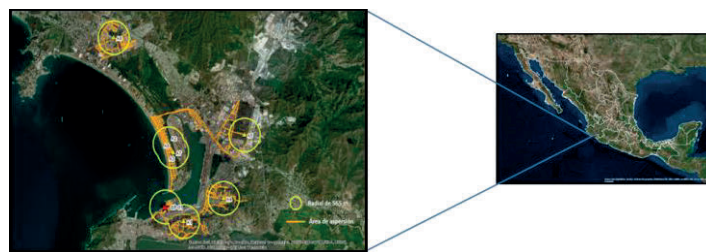
*WG 4 -Economical studies and governance:* assess the best suited modalities for the deployment of area-wide SIT on a regional scale, according to the species and the local context. Design and plan the governance for a multi-species mass-rearing facility in France.

*WG 5 – Concertation and information:* consider the outreach and prior consultation efforts that are prerequisite to a proper implementation of area-wide SIT projects.

Source: Clelia Oliva, Collectif-TIS.

## Eradication of a Transient Actionable Entry of *Bactrocera scutellata* (Hendel) in Manzanillo, Colima, Mexico. Plant Protection General Directorate SENASICA-SAGARPA, Mexico City, 7 November 2018

An outbreak of *Bactrocera scutellata* (Hendel) was recorded on 2 February 2018 in Manzanillo, Colima, Mexico. The outbreak has now been eradicated, since from the last specimen captured, nine biological cycles have passed without any additional detection (25 April to 30 September 2018). The nine cycles were assessed using the degree-day model. The male annihilation technique (MAT) and delimiting trapping were intensified in the last six cycles, following the recommendations of IAEA expert. Eradication actions were enforced following the framework of the 'Action Plan Against *Bactrocera scutellata* (Hendel) on National Territory (March 2018)' and based on the International Standards of Phytosanitary Measures N° 8 relative to the Determination of Pest Status in an Area and N° 9 relative to the Directives for Pest Eradication Programmes.



Location of *Bactrocera scutellata* transient entry, Manzanillo, Colima Mexico.

Corrective actions were implemented in a delimited area of 398 km<sup>2</sup>, where 1 104 traps were installed, with 29 790 trap inspections, 1 457 fruit host samples dissected, 10 145 MAT devices deployed (Jackson Traps baited with Cuelure + Malathion) and the localized spraying of 5 657 litres of the Spinosad-based insecticide-bait. 99 technicians participated in the Action Plan during different time periods.



In addition to the service provided by SENASICA structure *per se*, the financial investment of the Federal Government on the execution of the Plan was US \$1.3 million.



*Implementation of the Male Annihilation Technique (MAT).*

Following the Action Plan procedures, the MAT devices will be gradually retrieved from the field. Based on the assessed risk of introduction and spread of this transient actionable pest entry, the trapping network will be restructured in the urban area and at the Manzanillo Port, increasing the numbers from 48 to 567 permanent traps. In addition, the trapping network against not-native invasive species of quarantine significance will be reinforced along the Pacific Coast including: Topolobampo and Mazatlán, Sinaloa; Lázaro Cárdenas, Michoacán; Salina Cruz, Oaxaca and; Puerto Chiapas, Chiapas.

With the eradication of this pest outbreak and the strengthening of the trapping network, a total area of 473 000 hectares of cultivated hosts is being protected with an estimated production of 11.3 million tonnes valued at US \$3.3 billion (Servicio de Información Agroalimentaria y Pesquera, 2018).

*Source: Francisco Ramirez.*

## **USDA: Pink Bollworm is no Longer an Economic Threat**

In an October 19 announcement in Washington, DC, Agriculture Secretary Sonny Perdue proclaimed that “U.S. cotton is free – after more than 100 years – of the devastating pink bollworm.” Perdue said the pink bollworm has cost U.S. producers tens of millions of dollars in yearly control costs and yield losses. He stated that “thanks to rigorous control and regulatory activities carried out by USDA, state departments of agriculture, the U.S. cotton industry, and growers, the pink bollworm has been eliminated from all

cotton-producing areas in the continental United States. As a result, USDA is lifting the domestic quarantine for pink bollworm, relieving restrictions on the domestic and international movement of U.S. cotton.” Perdue noted that cotton growers were critical to this success, banding together to carry out a coordinated, multi-state programme and shouldering 80% of the program’s cost.

The National Cotton Council (NCC) welcomed the USDA announcement. During the ceremony, Clyde Sharp, an Arizona grower who co-chairs the NCC’s Pink Bollworm Action Committee with Ted Sheely, a California grower, said, “On behalf of the cotton producers who have battled the pink bollworm most of their career, and those that battled the pink bollworm before us, we thank everyone that has made this day possible. Amazingly, the pink bollworm eradication program relied mostly on insect control measures that were least disruptive to the agro-ecosystem.”

According to USDA, the pink bollworm was first detected in the United States in Hearne, TX, in 1917. By the mid-1950s, the pest had spread to surrounding states and eventually reached California in 1963. In 1955, USDA’s Animal Plant Health Inspection Service (APHIS) established domestic pink bollworm regulations. At the height of the program, 10 states – Arizona, Arkansas, California, Louisiana, New Mexico, Oklahoma, Texas, Nevada, Mississippi and Missouri – were quarantined for this pest. Many of these infestations were suppressed through cooperative federal, state and industry programmes.

By 2003, only Arizona, California, New Mexico and Texas remained under regulation. Eradication of pink bollworm took years of committed research by USDA’s Agricultural Research Service (ARS) and included planting transgenic cotton, using insect pheromones to disrupt mating, releasing sterile insects to prevent reproduction, and extensive survey. Many of the research findings by ARS became management strategies used by APHIS and cotton producers in their battle against pink bollworm.

NCC President/CEO Gary Adams, who also participated in the ceremony, said the NCC appreciated the united effort of its producer members, the dedication and coordination of APHIS staff, the collective contributions of many scientists, and the federal funding support from Congress that enabled USDA-APHIS to complete this monumental task.

“The pink bollworm, a destructive insect pest of cotton that once required multiple insecticide applications while continuing to reduce yields, is no longer present in U.S. cotton production,” Adams said. “The benefits of this program are shared by society, the environment, and the united producer membership who led this battle to victory.”

*Source: Cotton Grower Staff.*

## Relevant Published Articles

### Synonymy of *Toxotrypana* Gerstaecker with *Anastrepha* Schiner (Diptera: Tephritidae)

Allen L. Norrbom,<sup>1</sup> Norman B. Barr,<sup>2</sup> Peter Kerr,<sup>3</sup> Ximo Mengual,<sup>4</sup> Norma Nolasco,<sup>5</sup> Erick J. Rodriguez,<sup>6</sup> Gary J. Steck,<sup>7</sup> Bruce D. Sutton,<sup>8</sup> Keiko Uramoto<sup>9</sup> and Roberto A. Zucchi<sup>10</sup>

<sup>1</sup>Systematic Entomology Laboratory, USDA, ARS, Washington, DC, USA

<sup>2</sup>Center for Plant Health Science and Technology Mission Laboratory, USDA-APHIS, Edinburg, TX, USA

<sup>3</sup>California State Collection of Arthropods, Plant Pest Diagnostics Branch, California Department of Food and Agriculture, Sacramento, CA, USA

<sup>4</sup>Zoologisches Forschungsmuseum Alexander Koenig, Bonn, Germany

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#### Abstract

Based on a recent phylogenetic analysis, *Toxotrypana* Gerstaecker (1860) is here placed in synonymy with *Anastrepha* Schiner (1868). Although *Toxotrypana* is the senior name, *Anastrepha* is recognized as valid because of the much greater economic significance of its species. Changing the names of the many pest species of *Anastrepha* would cause major nomenclatural instability and confusion in the applied literature, therefore a proposal to the International Commission on Zoological Nomenclature has been submitted to validate this usage. The following nomenclatural changes are proposed: *Anastrepha australis* (Blanchard 1960), new combination, *Anastrepha curvicauda* (Gerstaecker 1860), new combination, *Anastrepha littoralis* (Blanchard 1960), new combination, *Anastrepha nigra* (Blanchard 1960), new combination, *Anastrepha picciola* (Blanchard 1960), new combination, *Anastrepha proseni* (Blanchard 1960), new combination, *Anastrepha recurcauda* (Tigrero 1992), new combination (all transferred from *Toxotrypana*), and *Anastrepha nigrina* Norrbom, new name for *Anastrepha nigra* Norrbom and Korytkowski (2009). Some changes to the species group classification within *Anastrepha* are also discussed.

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### Rates and Patterns of Laboratory Adaptation in (Mostly) Insects

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#### Abstract

Insects and other invertebrates can readily adapt to a range of environmental conditions and these include conditions used in artificial rearing. This can lead to problems when mass-rearing insects and mites for release as biocontrol agents or in sterile insect control programs, and when using laboratory strains to understand field population dynamics. Laboratory adaptation experiments also help to understand potential rates of trait evolution and repeatability of evolutionary changes. Here, we review evidence for laboratory adaptation across invertebrates, contrasting different taxonomic groups and providing estimates of the rate of evolutionary change across trait classes. These estimates highlight rapid changes in the order of 0.033 (median) haldanes and up to 2.4 haldanes, along with proportional changes in traits of more than 10% per generation in some cases. Traits tended to change in the direction of increased fitness for Coleoptera, Diptera and Hymenoptera, but changes in Lepidoptera were often in the opposite direction. Laboratory-adapted lines tend to be more sensitive to stress, likely reflecting relaxed selection for stress-related traits. Morphological traits show smaller changes under laboratory conditions than other types of traits. Estimates of evolutionary rates slowed as more generations were included in comparisons, perhaps reflecting nonlinear dynamics although such patterns may also reflect variance differences among trait classes. The rapid rate of laboratory adaptation in some cultures reinforces the need to develop guidelines for maintaining quality during mass-rearing and highlights the need for caution when using laboratory lines to represent the performance of species in vulnerability assessments.

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