## 5. BENEFITS AND OPPORTUNITIES

The OKSIR programme is an AW-IPM programme that is able to combat insect infestations across multiple local community jurisdictions. It is a programme that requires close collaboration between four different regional governments, growers, urban host-tree owners, fruit packers, industry advisors, tree-fruit retailers and researchers. Therefore, there are benefits and responsibilities at all levels, and the continued commitment of all partners is vital.

Prior to the AW-IPM programme and the release of sterile insects, spraying organochlorine and later organophosphate insecticides was for many years the main method to control the codling moth. Growers had to apply multiple cover sprays each season, and concerns that the codling moth might become resistant to insecticides mounted. In addition, there were growing concerns regarding chemical residues on the crop and the insecticide load in the environment. Residents of the Okanagan Valley have a strong preference for reduced insecticide use on their properties as well as on neighbouring farms, and for reduced insecticide residues on food (Cartier 2014). Insecticide poisoning of farmworkers and groundwater contamination are also serious concerns in many fruit-growing areas (Witzgall et al. 2008), and run-off from farmers' fields may have direct toxic effects on the water supply and aquatic organisms.

Following the introduction of the OKSIR programme, the reduced reliance on insecticides used for codling moth control in the region has significantly minimized the potential risks to the environment, including the local biodiversity and water for both people and crops, and has significantly minimized the risks to workers' safety while maintaining the economic viability of the crop (Tracy 2014). Residents who live near the orchards now have lower exposure to insecticides and access to sustainably produced fruit, and they enjoy the benefits of a thriving local agriculture and the green spaces it provides. Also, the local economy is improved because the agritourism and agricultural industries are supported by the programme.

Currently, a trend towards "social economy" is becoming apparent in Canada, in which individuals and groups take into account the social consequences of economic activity, including the increased attention to social issues. Local non-insecticide control of the codling moth can be seen as a "shared value," which is defined as "policies and operating practices that enhance the competitiveness of a company while simultaneously advancing the economic and social conditions in the communities in which it operates" (European Commission 2013).

This trend is evidenced by the willingness of the vast majority (90.4%) of nonagricultural residents in the area to pay for the programme. Under the current funding structure of the area-wide approach, growers pay 65% less than they would for the same service without the programme (Cartier 2014).

Both the provincial and federal governments contributed toward the cost of constructing the codling moth mass-rearing facility (CAD 7.4 million), which is a major seasonal employer in the region. Total employment associated with the programme contributes CAD 2.2 million, or CAD 620 per ha, to the regional gross domestic product (Cartier 2014). Cartier's (2014) benefit-cost analysis revealed that, for every CAD 1 in cost there was CAD 2.5 in benefit, for both producers and society as a whole.

As the mass-rearing facility has the capacity to produce 780 million sterile codling moths annually, of which only a portion is used seasonally to treat the areas in the Okanagan Valley, this has opened opportunities to serve other regions or countries through sales of sterile moths or egg sheets, and offers options for codling moth virus production. As an example, the OKSIR programme currently sells excess production of sterile moths to researchers in the USA working on pheromone research. The New Zealand Institute for Plant and Food Research is conducting a pilot project for codling moth control using sterile codling moth shipped from Canada (Horner et al. 2016). The OKSIR programme also sells egg sheets for commercial virus production.

There are examples of for-profit operations selling sterile insects and related services commercially, such as the onion fly *Delia antiqua* (Meigen) in the Netherlands (Vreysen et al. 2006), Mediterranean fruit fly in Israel and South Africa (Bassi et al. 2007; Barnes et al. 2015), and the false codling moth in South Africa (Boersma, this volume).

As costs of controlling the codling moth decrease because of the success of areawide application of the SIT, other resources can be redirected to address threats of other/new invasive insect pests. Due to climate change and increasing global trade, invasive insect pests are migrating to new habitats throughout the world.

Subsequent to the successful suppression of the codling moth to below economic levels, the local fruit industry has requested the programme to use its surveillance infrastructure to monitor other existing pests, e.g. leafrollers, and new invasive pests, such as apple clearwing moth *Synanthedon myopaeformis* Borkhausen, apple maggot *Rhagoletis pomonella* Walsh, and brown marmorated stink bug *Halyomorpha halys* Stål. Since programme staff visit the pome orchard properties weekly for codling moth management purposes, there is an opportunity to incrementally add monitoring services for other pests and take advantage of economies of scale.

Insecticide resistance is also increasing. The SIT reduces insecticide use and, therefore, reduces the likelihood of insecticide resistance. There is an opportunity to use codling moth SIT to supplement the use of other biocontrol techniques, such as pheromone-mediated mating disruption or biological insecticides, e.g. *Cydia pomonella* granulosis virus (CpGV) (Judd and Gardiner 2005; Eberle and Jehle 2006; Cardé 2007; Witzgall et al. 2008). The SIT is very efficacious and sustainable, and hence is the ideal tool to supplement other methods as needed (Dyck et al. 2021).

A further benefit to a region of an AW-IPM programme with an SIT component is the provision of employment opportunities for area residents. A major challenge can be the availability and recruitment of sufficient staff for the planned facility and field operations when choosing the physical location of the programme. Central administration of the programme is important because it ensures continuity between areas and allows for the consistent provision of full IPM support services, including monitoring, education and enforcement, at a lower cost. The commitment of staff to the outcome is also critically important; experienced staff is the programme's most valuable resource.

Looking globally, apples and cherries from the area can now be exported into highvalue restricted markets, such as Taiwan, China and Japan (the presence of the codling moth restricts the movement of cherries and apples in some high-value Asian markets). The SIT promotes areas of low pest prevalence, which can allow for the biosecurity of the product to be almost guaranteed, while the cost for a rejected shipment of fruit in such restricted markets and the closure of such markets can be very costly to the exporting nation.

The OKSIR programme is also working with leaders worldwide in addressing invasive pests in a changing climate by developing projects that transfer knowledge using AW-IPM approaches and the SIT. In addition, the programme has successfully exported quality sterile moths to New Zealand, South Africa and the USA for use in IPM pilot projects and research (Blomefield et al. 2011; Carpenter et al. 2012; Horner et al. 2016; Adams et al. 2017).

In recent years, the visibility of the OKSIR programme on the international scene has increased; in 2015 at the 8th International IPM Symposium the programme received the International IPM Award of Excellence for a regional integrated pest management programme (IPM 2015). Also, a panel of international experts recommended the OKSIR programme as a role model for AW-IPM using the SIT (Carpenter et al. 2014).

The Food and Agriculture Organization of the United Nations, the World Bank and other international organizations promote the deployment of biological control agents to suppress pests and replace chemical controls (IAASTD 2009). The Stockholm Convention on Persistent Organic Pollutants, a global environmental treaty effective from 2004, aims to eliminate or restrict persistent organic pollutants (Stockholm Convention 2009). These actions epitomize the global need for environment-friendly pest management programmes like the OKSIR programme. Following improved technology and research, the SIT could play an important role in this area, particularly as a supplement to other biocontrol techniques applied on an area-wide basis (Cardé and Minks 1995; Hendrichs et al. 2007).

## 6. CHALLENGES

According to Tracy (2014), common obstacles to adopting a programme like the OKSIR are:

1. Low levels of investment in research and development.

2. Lack of coordination among growers in adopting the different methods.

3. Weak or conflicting regulatory framework.

4. Absence of market incentives and consumer awareness.

The OKSIR programme's experience of more than 20 years has revealed vital lessons. The goal in sharing these lessons is to assist other regions and pome fruit growers who want to incorporate the SIT for codling moth control into their pest management system and start their programmes as efficiently and smoothly as possible (Carpenter et al. 2014).

Support from political partners (policy/regulatory) and the public is critically important for establishing and sustaining an AW-IPM programme like OKSIR. The impact of the pest and the techniques used to manage it can be influenced by both politics and emotions. It is vital that all stakeholders share common values, and that their expectations are identified from the beginning and managed along the way. This is crucial for building trust in the programme, especially in its initial phases before the programme results can provide convincing proof.

Champions for the programme outside the administration who represent the interests of each of the stakeholder groups are extremely useful to move the initiative forward and maintain ongoing support for an AW-IPM programme.

Committed partners in the programme are critical to its success. It may be easy to identify and join forces with potential partners, but what happens if this collaboration fails or is unproductive? It is crucial to define how the partnership is constituted. The responsibilities of each partner must be mutually agreeable, such that the integrity and efficacy of the programme can be maintained. Not doing so can jeopardize the success of the entire programme. Defining procedures to allow exit and entry of partners is invaluable.

In the initial phase of the OKSIR programme, many growers resented having the programme "imposed" on them. In addition, it was difficult to convince growers, whose livelihoods depend on their crops, to trust the SIT component of the AW-IPM approach and to apply supplemental chemical control only when needed. However, trust grew over time, in large part due to the real-time trapping and monitoring data made available to growers on the website, and especially with the obvious decline in codling moth populations and crop damage. Growers are now very supportive of the AW-IPM approach because it protects them from potential infestation coming from their neighbours' poor management practices (both commercial and residential). In addition, the high number of hits on the website's real-time data pages shows that growers are checking their monitoring data and scrutinizing moth thresholds before spraying, rather than simply applying "comfort sprays" (Lefebvre et al. 2015a).

This was the first area-wide use of the SIT for controlling the codling moth in commercial orchard plantings. Challenges in the early years of the programme included overcoming public resistance to the mandatory requirement to control the pest on all properties, and for the growers to receive a mandatory applied control measure. This was caused in part by ineffective communication with the public and philosophical disagreements between the programme and individual property owners. However, over time, due to improved communication, the roles and responsibilities of all stakeholder groups were better understood, resulting in better cooperative actions and more effective codling moth control. Furthermore, the legal authority for programme staff to enter properties to conduct activities was initially met with resistance from growers and the public. This authority is vitally important for the success of the programme.

Convincing consumers to embrace AW-IPM practices has also been a challenge. Compared with the sale price of fruit produced by conventional production systems, it has been shown that consumers are willing to pay significantly more for certified organic fruit, and moderately more for fruit produced through IPM practices (Lefebvre et al. 2015a). The OKSIR programme supports a relatively small organic pome fruit industry, i.e. less than 10% of British Columbia's pome fruit production (Macey 2013). These growers enjoy the economic benefits of marketing their fruit as certified organic. However, the remaining conventional growers have not yet taken steps to capitalize on market opportunities afforded by agroecological and socially responsible practices in the same way that growers in the USA have benefited from the "Responsible Choice" label (Tracy 2014). During the planning stages of an AW-IPM programme, it is important to understand what the benefits and costs of the programme are, and to understand the long-term expectations of the stakeholders once the programme achieves its goals. A major challenge to maintain a sustainable programme is to obtain continued funding when the pest is no longer deemed a problem. The success of an AW-IPM approach means that the reason for its existence appears to diminish. However, removing the AW-IPM measures will create a resurgence of the pest to levels existing prior to the programme. For example, once success has been achieved, keeping the population levels below treatment thresholds requires a risk management approach.

The local governments in the community-based OKSIR programme are also under pressure to allocate tax revenue to other uses, and they must justify the continued areawide service to their constituents (who are also benefactors of the programme). In addition, there has been an overall decrease in pome-fruit area in the region because of low market returns. Because of their northern location, growers in British Columbia are faced with higher production costs than growers in the nearby USA. Also, higher profit margins can be made growing alternative crops such as sweet cherries and wine grapes. Even though a decreasing pome-fruit area equates to reduced parcel-tax revenue, the OKSIR programme has remained financially strong, and in the past seven years there has been no tax increase.

Expansion of the programme also presents challenges. There are challenges for a public-sector regulatory service when considering diversifying the revenue model to include expansion of scale and scope, such as producing a commercial supply for sales of sterile moths or other by-products, and in assisting the setting up of programmes in other regions/countries. There can be complications due to the governance/business model, as well as different languages and cultures when providing technical support to those areas purchasing sterile insects. There are regulatory, shipping and logistics timing issues involved in selling a product internationally (Blomefield et al. 2011; Adams et al. 2017; Suckling et al. 2017). Biological control agents and beneficial insects are still a developing sector. Unfamiliarity with import regulations of these new products by the many customs officials and private companies along the transport chain can impede movement over international borders (FAO 2017).

A small, locally funded programme cannot afford large-scale investment in research to improve the technology, and generally it can afford to focus only on critical operational improvements as situations arise. The OKSIR programme is currently working with collaborators in New Zealand and the USA on advancing the use of unmanned aerial systems for more efficient release of the sterile codling moth in field implementation trials using the SIT to control the codling moth.

The OKSIR programme has relied on the senior government and international research community for technical assistance, and for advancing AW-IPM and the SIT for the codling moth. Due to the increasing public pressure to reduce the use of chemical insecticides around the world, a global investment in SIT research is needed to advance the technology. A few areas worthy of investigation include if or how released sterile female codling moths could create an effect similar to commercially applied pheromone-mediated mating disruption, how SIT affects parasitoid populations, what are effective sterile: wild ratios for extremely low wild populations, and methods to monitor extremely low wild densities (Judd and Gardiner 2005; Witzgall et al. 2008; Bau and Cardé 2016).

#### 7. CONCLUSIONS

The OKSIR programme is a highly effective and easily transferrable model to control orchard pest infestations. There is greater demand for biological control methods to meet mounting pressures of climate change, increasing insect pest populations, impacts of global trade, and concerns over affordability and availability of methods. Concerns are mounting that the codling moth is becoming resistant to the insecticides used commonly for control, with few replacement chemical options being developed. Biological control options also face pressure because they are not stand-alone technologies, often relying on compatible, supplemental control products and crop management practices.

The OKSIR programme clearly illustrates that area-wide integration of the SIT can successfully manage codling moth populations in an environmentally sound way. In addition, it can easily be integrated with other biological control methods such as pheromone-mediated mating disruption and CpGV. Most importantly, the SIT can replace control products that are no longer environmentally or economically viable, and hence provide an excellent biologically sustainable solution for controlling insect pests.

Despite its proven success, the SIT is often considered a curiosity rather than an effective, environment-friendly technology that dovetails into many modern IPM programmes. When considering the success of the OKSIR programme, other regions and pome fruit growers around the world will be compelled positively to consider AW-IPM with a SIT component as a viable management strategy for the codling moth.

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# AREA-WIDE MANAGEMENT OF MEDITERRANEAN FRUIT FLY WITH THE STERILE INSECT TECHNIQUE IN SOUTH AFRICA: NEW PRODUCTION AND MANAGEMENT TECHNIQUES PAY DIVIDENDS

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#### SUMMARY

A mass-rearing facility to produce sterile male Mediterranean fruit flies, Ceratitis capitata (Wiedemann), for a Sterile Insect Technique (SIT) programme in the Hex River Valley in the Western Cape Province started in the late 1990s. The programme was initially underfunded and could only produce about 5 million sterile male flies per week. The resultant aerial release rate of 500 sterile males/ha/week reduced wild Mediterranean fruit fly populations substantially, but not to sufficiently low levels. Due to financial considerations, in 2003 aerial releases were replaced with ground releases targeting all gardens, other hotspots and neglected host plants. It was clear that with more funding, fruit fly mass-rearing facility and field operations could be improved, better quality control could be implemented, and more and better quality male sterile flies could be produced and released. Increased government support in 2001 resulted in a larger mass-rearing facility, and further improvements included the implementation of a quality control management system and the introduction of a new genetic sexing strain (VIENNA 8). The resultant increase in the production of sterile Mediterranean fruit flies of better quality enabled the SIT programme to be systematically introduced to additional fruit production areas. The Mediterranean fruit fly SIT programme was privatised in 2003 and is now operated by FruitFly Africa (Ptv) Ltd. In 2009 a new approach to funding was adopted with a renewable Memorandum of Understanding (MoU) between the Department of Agriculture, Forestry and Fisheries (DAFF) and the deciduous fruit and table grape industry. Under the MoU, the DAFF provides 50% of the necessary funding, while 50% is collected from growers through statutory levies. In 2010 a new state of the art mass-rearing facility became operational and subsequent improvements in production processes and facility maintenance resulted in improved fruit fly production and quality. By 2016 sterile male production had increased to 56 million flies per week. After 12 years of ground releases of sterile Mediterranean fruit flies, aerial releases were resumed in three main production areas, and, at the time of writing, include approximately 15 000 ha of commercial deciduous fruit and table grapes. As a result of this well-funded area-wide integrated pest management (AW-IPM) programme, average wild Mediterranean fruit fly populations in the SIT areas have decreased by as much as 73%. The

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J. Hendrichs, R. Pereira and M. J. B. Vreysen (eds.), Area-Wide Integrated Pest Management: Development and Field Application, pp. 129–141. CRC Press, Boca Raton, Florida, USA. © 2021 IAEA South African Mediterranean fruit fly SIT programme now aims to manage some of the fruit production areas as areas of low pest prevalence. Increased funding and a stable income stream also enabled *FruitFly Africa* to apply early detection and rapid response programmes for invasive pests such as *Bactrocera dorsalis* in relevant areas.

Key Words: Ceratitis capitata, Ceratitis rosa, Ceratitis quilicii, Tephritidae, SIT, public/private partnership, sterile male releases, suppression, Western Cape, Northern Cape, Eastern Cape

### 1. INTRODUCTION

The Western Cape Province is the centre of the South African deciduous fruit industry, followed by the Northern and Eastern Cape Provinces and to a lesser extent by smaller production areas in other provinces. Two fruit fly species of economic importance were previously recorded as occurring in the Western Cape, i.e. the Mediterranean fruit fly (*Ceratitis capitata* (Wiedemann)) and the Natal fruit fly (*C. rosa* Karsch) (Blomefield et al. 2015). However recent studies have revealed distinct morphological and molecular differences within populations of *C. rosa*, differentiated further by environmental requirements such as by temperature and altitude (Virgilio et al. 2013; Karsten et al. 2016). This resulted in the description of a new species viz. *Ceratitis quilicii* (De Meyer et al. 2016; FAO/IAEA 2019). Comprehensive surveys have not yet been conducted to determine the prevalence of either species. It can be deducted, however, from the studies of Karsten et al. (2016) that *C. quilicii* is more prevalent in the Western Cape than *C. rosa*.

Mediterranean fruit fly is the predominant species in most areas in the Western Cape (De Villiers et al. 2013; Manrakhan and Addison 2014) and is categorised as a quarantine pest for most of South Africa's export markets for deciduous fruit. Globally, more than 260 different fruit species, including citrus, are hosts of Mediterranean fruit fly, and it can cause enormous crop losses to commercially-produced fruit and also some vegetables if not controlled (USDA 2019). Small-scale farmers, as well as communities with backyard fruit trees, are also seriously affected by this species (White and Elson-Harris 1994).

Deciduous fruit (pome and stone fruit) and table grapes are mostly grown in mountain valleys in the Western and Eastern Cape, and in a semi-desert area alongside the Lower Orange River in the Northern Cape. The valleys are fairly isolated by surrounding mountains, and the Lower Orange River production area by the surrounding semi-desert area, making possible area-wide integrated pest management (AW-IPM) programmes incorporating the Sterile Insect Technique (SIT) covering a number of separate and relatively isolated areas.

South Africa is a net exporter of fruit and for many decades has had an established, well organised and integrated deciduous fruit industry, which in 2016 exported approximately 880 000 metric tons of deciduous fruit with an estimated value of USD 1200 million (DAFF 2017). The deciduous fruit industry in South Africa is one of the largest employers in horticulture, representing a significant investment both in terms of human resources and foreign exchange earnings (DAFF 2017). The country cannot afford to jeopardise future exports by allowing fruit flies to hinder international trade.

It is therefore essential for the South African export fruit industry to reduce fruit fly interceptions to a minimum to ensure market access and to reduce production losses. The European Union is the destination of more than 40% of fruit exports, and other markets include Japan, Taiwan and the USA (DAFF 2017). The European Union will intercept and detain fruit consignments for any non-European fruit fly larvae detected in fruit, which often includes unidentified larvae of the Mediterranean fruit fly, but also other *Ceratitis* spp., if the consignment does not originate from the European Union.

The use of chemical insecticides has become increasingly complex due to pest resistance, environmental concerns, and restrictions on residue levels by importing countries. In the interests of reducing insecticide use, as well as pre- and post-harvest crop losses, while maintaining sustainable agricultural systems, AW-IPM programmes integrating the SIT have proved effective in supporting safe and environment-friendly international trade.

South Africa is one of the largest deciduous fruit exporting countries in the southern hemisphere, but with a relatively small SIT programme to suppress or eradicate fruit flies. South Africa's major competitors on the international fruit export market, such as Chile, are either fruit fly-free, well advanced in achieving this or have at least low pest prevalence status. Use of the SIT, which has successfully contributed to eradicating the Mediterranean fruit fly in Chile and North America, as well as parts of Argentina, Australia, Peru, and Central America, has resulted in substantial savings to these countries (Enkerlin 2021).

A key factor for the success of any SIT programme is availability of adequate funding and long-term commitment of stakeholders. Funds generated from growers by making use of, e.g. a statutory grower levy, need to be supplemented with government funding in view of the public benefits of such programmes. Political will to support AW-IPM programmes which include the SIT is therefore needed to ensure sustainable funding (Dyck et al. 2021a).

## 2. HISTORIC OVERVIEW OF THE SOUTH AFRICAN SIT PROGRAMME

The Mediterranean fruit fly SIT programme in South Africa originated in 1996 when the Agricultural Research Council's (ARC) Infruitec-Nietvoorbij Institute for Fruit, Vine and Wine in Stellenbosch approached the Joint Food and Agriculture Organization of the United Nations (FAO)/International Atomic Energy Agency (IAEA) Programme of Nuclear Techniques in Food and Agriculture for technical support for a project to investigate the feasibility of integrating the SIT to suppress or eradicate Mediterranean fruit fly in the Hex River Valley. The pilot area was chosen mainly because of its relative geographic isolation, its large production area of 5000 ha of table grapes, a major export crop, and the fact that Mediterranean fruit fly was the dominant fruit fly pest (Barnes 2016).

Aerial releases of sterile Mediterranean fruit flies using a fixed-wing aircraft started in 1999, and flies were dispersed at a density of 500 sterile male Mediterranean fruit flies per ha per week. Wild Mediterranean fruit fly populations were subsequently reduced by 80% (Barnes et al. 2015), but still inadequately.

Nevertheless, encouraged by these early results, the governing body of the deciduous fruit industry, the then Deciduous Fruit Producers' Trust (DFPT, later HORTGRO) assisted the ARC with limited funding for the implementation of the project. Overall management of project funding at local level was through a formal 'SIT Partnership' agreement between the ARC and the DFPT. Additional funding in 2001 from the Western Cape Department of Agriculture allowed for improved infrastructure.

In 2002, a quality management system was incorporated into the mass-rearing process, and in 2003 a new genetic sexing strain of Mediterranean fruit fly based on a temperature sensitive lethal (*tsl*) mutation, i.e. the VIENNA 8 strain, was introduced and reared as the main colony (Franz et al. 2021). All these factors significantly improved mass-production levels and quality of the sterile males (Barnes et al. 2015; Barnes 2016).

In 2003 the programme was privatised with the establishment of *SIT Africa (Pty) Ltd.* (Barnes 2007), and in 2004 the sterile male release programme was extended to two additional production areas (Barnes 2016). Financial considerations resulted in the replacement of aerial releases with ground releases in 2003. These were focussed on farm and town gardens and other hotspots where wild Mediterranean fruit fly populations remained high (Barnes 2016). The rationale behind this strategy was to achieve high sterile to wild fly ratios in these localities where wild flies overwinter in low numbers (Barnes 2008), thus minimizing the number of wild flies which are able to migrate back to commercial fruit plantings in summer. However, still insufficient sterile to wild fly ratios during summer often occurred (Manrakhan and Addison 2014).

Subsequently, substantial funds were made available by the fruit industry to introduce extensive fruit fly monitoring programmes in production areas in order to identify and further suppress fruit fly hotspots prior to sterile male releases to ensure that momentum in the SIT programme was maintained. Greater detail on the development, progression and results of the Mediterranean fruit fly SIT programme is given in Barnes (2007, 2016).

When it became clear that a new approach was required to ensure sustainable funding and industry-wide roll out of the Mediterranean fruit fly SIT programme, a 50:50 contractual funding partnership was formed in 2008 between the DFPT and the then National Department of Agriculture (NDA) (now Department of Agriculture, Forestry and Fisheries, DAFF) in the form of a Memorandum of Understanding (MoU).

The broad AW-IPM programme with a SIT component includes, at the time of writing, eight distinct fruit production areas at different levels of SIT implementation. The sterile flies are released in the Elgin, Grabouw and Vyeboom area (9600 ha), the Hex River area (Hex River Valley, De Wet and Brandwacht, 5700 ha), and the Warm Bokkeveld, Wolseley and Tulbagh area (7000 ha), all in the Western Cape Province. Pre-SIT baseline data collection is being carried out in the Hemel and Aarde Valley in the Western Cape (300 ha), the Langkloof Valley in the Eastern Cape (4700 ha), and in the Lower Orange River area in the Northern Cape (Kakamas and Keimoes, 4200 ha).

Fruit fly densities in commercial orchards are monitored with Chempac® bucket traps baited with a three-component lure (Biolure) that are deployed at a density of 1 trap per 20 ha (Barnes 2016). Baiting no longer includes organophosphate insecticides, but the organically certified spinosad-based product GF-120 NF NATURALYTE <sup>™</sup> bait.

The programme contributes towards various government priorities, such as export competitiveness, economic growth and development, job creation, food security, and reduced insecticide use. Monitoring for non-native fruit flies, including *Bactrocera dorsalis* Hendel, which is already established in northern parts of South Africa (Manrakhan et al. 2015), forms part of the national exotic fruit fly surveillance programme, which is augmented through the SIT monitoring programme. For these species, Chempac® bucket traps baited with methyl eugenol lures are used at a density of 1 trap per 100 ha.

### 3. FINANCIAL MODEL

By 2008, several factors had influenced the economic viability of the programme. These included the absence of sustained investment from government, an inability to raise venture capital from private institutions, a fruit industry which was under economic stress and the resultant difficulties in getting grower buy-in, and a too-small and aging Mediterranean fruit fly mass-rearing facility which could not produce the numbers of sterile flies required (Barnes 2007). It was in this context that the DAFF/DFPT MoU was formalised in 2008.

The DAFF/DFPT MoU is a 3-year renewable contract; in 2009 DAFF's contribution was approximately USD 460 000 (current value) and included an annual consumer price increase. Under the MoU, approximately the same amount was contributed by the fruit industry towards the monitoring and sterile Mediterranean fruit fly production components of the programme. The aerial baiting component of the programme is funded solely by producers.

The objectives of the MoU focussed on the concept that reduced fruit fly population levels can lead to areas of low pest prevalence, pest free areas and, possibly, eradication of invasive fruit flies. This would ensure maintenance of market access and increase South Africa's ability to export high quality, residue safe fruit. Furthermore, all producers within the relevant area would be able to participate.

Subsequently, the MoU has been renewed twice. The current MoU for the business years 2015/16 to 2017/18 has ensured a financial contribution to the SIT programme by DAFF of USD 770 000, USD 930 000 and USD 1.1 million, respectively.

In 2013 *SIT Africa* evolved into *FruitFly Africa (Pty) Ltd.* (FruitFly Africa 2019), which was recognised by DAFF as the implementation structure for the MoU. DAFF funding is allocated for major projects within the SIT programme, namely awareness and educational programmes, optimisation of fruit fly monitoring, preparation of new areas for sterile Mediterranean fruit fly releases, supplementary fruit fly bait applications, remedial action in hotspot areas, effective radiation sterilisation of pupae, increased production and quality of sterile flies, area-wide release techniques, and continuation of releases in existing areas.

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#### 4. ORGANIZATIONAL STRUCTURE

The current (2017) organizational structure of *FruitFly Africa* is given in Fig. 1. During the initial stages of sterile fly production at the Stellenbosch mass-rearing facility, 5 million sterile flies per week were produced by 15 rearing technicians. *FruitFly Africa* currently (2017) has eight rearing technicians; their production output per week has risen from 15 million in 2014 to the 56 million VIENNA 8 sterile males per week in 2017. Production efficiency has been improved not by additional automation, but by the efficient use of labour and the adoption of improved production and quality practices.

A quality control officer monitors the quality of the sterile male flies produced, ensures adherence to the protocol of aerial applications of GF-120 baits, and evaluates all other fruit fly management measures applied in the field.

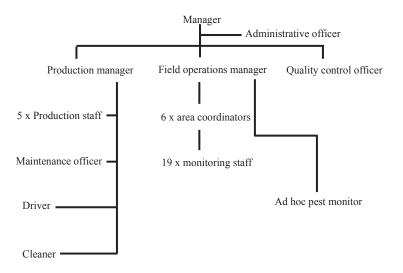


Figure 1: The current organizational structure of FruitFly Africa (Pty) Ltd.

Field surveillance staff carry out the day-to-day tasks in the areas that participate in the programme. Each SIT area has a coordinator who is responsible for planning and public relations in that area. Field monitoring staff report to the area coordinator in that area, who in turn reports to the field operations manager who supervises the implementation of monitoring and management strategies within that area.

#### 5. MANAGEMENT

In a SIT programme, sterile insects must be of the best quality, strategies and decisions need to be technically correct, and customer service has to be excellent. This, coupled with the fact that the broader community which is impacted also needs to contribute to fruit fly suppression, has a unique impact on programme management practices

(Dyck et al. 2021b). As the current programme evolved, it became evident that programme management needs to be able to rely on staff that are technically competent, and that are able to build good working relationships with all fruit industry stakeholders, e.g. community organizations and individual producers. People possessing both these skills are relatively rare, and in this programme much emphasis is put on recruiting suitably-skilled people for these tasks.

Since 2010, area-wide aerial baiting with GF-120 has also formed a crucial part of the fruit fly suppression strategy; in most areas four to six applications per season are applied shortly before harvest as an additional crop protection exercise. Groundreleased sterile flies are used as the main intervention in urban areas, farm gardens, on alternate hosts and in Mediterranean fruit fly hotspots.

In view of the encouraging results of aerial sterile fly releases during the 1999-2003 pilot phase in the Hex River Valley (Barnes 2016), and the improved funding base, aerial releases of sterile males were reconsidered and a pilot trial over 2200 ha was implemented during the 2014/15 season using a gyrocopter. Reasonable success was obtained (Barnes 2016), and in 2016/17 season-long, area-wide helicopter releases were carried out at the standard release rate of a 1000 sterile males/ha/week over  $\pm 39$  000 ha in three areas where the SIT forms part of the fruit fly management strategy. "Attract and kill" bait stations as well as mass-trapping were also used as part of the management strategy in programme areas, although their use has been limited to backyards and hotspots on farms.

Historically, fruit fly monitoring has been at a density of 1 trap per 20 hectares. These traps are not used in orchard-level decision making, but they are used to determine the area-wide distribution of both wild and sterile fruit flies, as well as to determine the ratios between the two. Because all farmers contribute financially in equal amounts to the programme, each of them feels entitled to the same AW-IPM service from FruitFly Africa in equal quantities. Once a strategy has been decided on for a season, it needs to be implemented equitably across all participating areas. Fruit fly trap catches have thus been not so much a tool for weekly management decisions, but more as an indication of whether a particular strategy for the season has been successful in that area. They are also a useful tool for timing other control interventions (e.g. host plant management), using historical trends. Trap catches have thus been used to compare fruit fly populations for a whole area across weeks and between seasons. Increasingly, farmers are opting for higher trap densities to enable them to make their own management decisions. For this, more detailed and timely information is necessary, and FruitFly Africa is thus developing an electronic database system that will be available to the farmers and provide detailed and realtime reporting on trap catches, and population levels and distribution.

## 6. PRODUCTION PRACTICES AND QUALITY CONTROL

During the period 2011 to 2015, systematic changes were made to the sterile Mediterranean fruit fly mass-rearing process based on experience gained by the *FruitFly Africa* quality control officer during a visit to the Moscamed El Pino Mediterranean fruit fly facility in Guatemala. These included a 20% reduction in adult fly density in the oviposition cages (from 4400 to 3600 adults – this equates to 0.0027

to 0.0034 cm<sup>3</sup> per fly), twice-daily brushing of eggs off the adult cage screen walls through which the females oviposit into water troughs, and the use of 1 kg 'starter packs' of diet for rearing first instar larvae.

The reduction in the number of adults per oviposition cage reduced the amount of stress on the flies that need to feed, mate and lay their eggs. Brushing the eggs more frequently from the screens through which they have been laid reduced the number of eggs that stuck to the screens and become desiccated, thereby increasing the percentage of viable eggs. The use of the 1 kg starter packs concentrated the recently-hatched larvae in a smaller volume of diet, thereby retaining essential metabolic heat.

Through a cascade effect, these measures resulted in better quality larvae, pupae and adults in the main colony, which in turn equated to better quality eggs, larvae, pupae and adults in the release stream. The end result is the release of better-quality sterile males (Barnes, 2016). The outcome of these changes is given below.

Additionally, a more stable flow of increased funding enabled the production team to make improvements to the facility infrastructure, as well as to the production equipment. This included improved illumination in the adult room, replacing eggbubbling aeration pumps with a single air supply line, better climate control equipment, and emergency standby services for equipment that is essential to production.

The stable flow of funding also enabled the mass-production facility to procure raw materials of a higher and more uniform quality from reliable sources. Examples of this are bran that is free of pesticide residues, vermiculite with low moisture levels, and yeast with a high and stable protein content.

The quality control parameters and production targets, calculated weekly, include daily egg production (volume), daily pupal production (volume), egg hatch (%), egg to pupa recovery (%), pupal weight (mg), adult flight ability (%), sterility in the release stream (%), and fertility of the main colony (%). All tests are carried out in accordance with the standards set out in the standard operating procedures and in the international product quality control manual (FAO/IAEA/USDA 2019).

## 7. RESULTS AND DISCUSSION

Following the inception of changes to the infrastructure and mass-rearing procedures described above, there was a marked improvement in the following production and quality control parameters in the release stream (Barnes 2016):

- Daily egg production per cage increased by 45.3%, with a decrease in standard deviation (SD) of 18.2%.
- Mean egg hatch improved from 39.6% to 42.6%, an increase of 7.6% (SD decreased by 50.0%). In 2011 the target of 40% hatch was often not met; this rarely happened in 2015.
- Egg to pupa recovery improved from 16.9% to 20.6%, an increase of 21.9%; there was no change in the SD.
- Mean flight ability increased from 82.2% to 87.5% (SD decreased by 64.8%). In 2011, the target of 80% was not achieved on a number of occasions; in 2015 it never dropped below 81%.

Egg to pupa recovery is a good indicator of the cost-effectiveness of production, since a large percentage of the variable costs of production is spent on rearing the fly from the egg stage to the pupal stage. With an increase in egg to pupa recovery it was not unexpected that the unit cost of production from 2011 to 2015 was reduced by 37% (nominal). In addition to the increased production efficiency, the increase in numbers of sterile males produced, coupled with a minimal increase in fixed costs, translated into lower unit costs, since the increase in total costs were not proportionate to that of total volumes. While the quality of sterile flies together with the cost-effectiveness of production are important to the success of an SIT programme, the effect of the programme on the degree of wild Mediterranean fruit fly population reduction has to be taken into account when considering the cost-effectiveness of such a programme.

The trend in wild Mediterranean fruit fly populations in the sterile male release areas during the period 2007 to 2017 is shown in Figure 2 (note the difference in the flies/trap/day (FTD) scale between the three areas).

The average wild fly population levels during the harvest season (first 20 weeks of the calendar year when ripe fruit is most abundant), in the large areas where the SIT forms part of the management strategy, decreased as follows:

- When comparing the average FTD for 2007-2008 (period before the MoU) with that of 2015-2017, the FTDs in the Hex River Valley decreased by 73% from an average of 4.32 to 1.14. This average includes hotspots that are focally supressed.
- The same comparison for the Elgin/Grabouw area indicates a population reduction of 19% (although the reduction from the 3-year period immediately following the MoU is 32%), from a FTD of 0.50 to 0.41.
- No reliable data prior to 2010 (implementation of the full programme) are available for the Warm Bokkeveld area. When comparing the average FTD for 2010-2011 with that of the period 2015-17, the FTDs in the Warm Bokkeveld decreased by 78% from an average of 1.46 to 0.32.

Over the period 2007-2017 the FTD values for the Hex River Valley were much higher than the FTD values of Elgin/Grabouw. This is mainly due to differences in wild and commercial host plants present and varieties cultivated, as well as harvesting processes and sanitation between the two areas (Barnes 2016), which made fruit fly management throughout the Hex River Valley more difficult. A further factor is climate; long-term data show that the Hex River Valley has higher average maximum temperatures than the Elgin/Grabouw area (Barnes et al. 2015), conditions which favour development of Mediterranean fruit fly (Nyamukondiwa et al. 2013).

Increased funding from DAFF from 2008 to 2017 allowed for essential improvements at the Mediterranean fruit fly mass-rearing facility. The timely integration of a combination of fruit fly management techniques made a positive difference to the outcomes of the programme. Production increased from 15 million sterile male flies per week in 2014 to 56 million per week by 2016, which enabled better sterile male to wild male overflooding ratios. Improved quality and quantity of sterile fruit flies produced and released, better release techniques, and overall, better on-farm management of fruit fly populations, have resulted in a generally steady decrease in average wild Mediterranean fruit fly populations over time, as illustrated in Fig. 2.

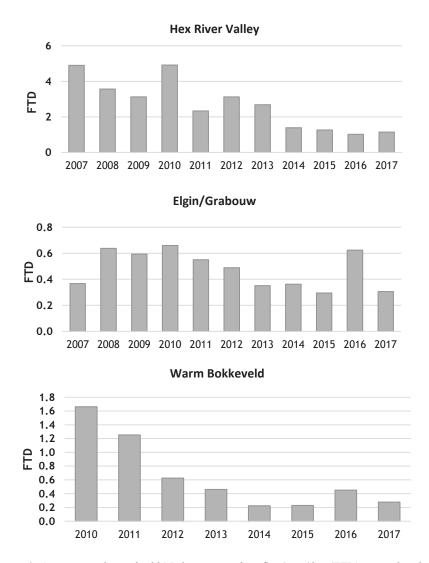


Figure 2. Average numbers of wild Mediterranean fruit flies/trap/day (FTD) trapped in three fruit production areas under SIT application during the first 20 weeks of the year (= harvest period) from 2007/2010 to 2017 (no data available for Warm Bokkeveld before 2010).

During the last 18 years, the South Africa Mediterranean fruit fly SIT programme has faced many challenges such as outdated infrastructure and inadequate equipment due to a poor local funding base, and initially a hesitant grower community, but, with excellent and sustained support from the FAO/IAEA, and later, better co-funding from government, determination by the SIT team, improved facilities and equipment, and a grower community steadily starting to believe in the programme, it has grown to a programme serving a total of 30 000 ha, and is destined for further growth.

Many lessons have been learnt along the way, the most important being:

- Broad-based, multi-organizational and sustainable funding must be available from the start of the AW-IPM programme.
- No single control measure is a stand-alone technology. All available tools for the management of the pest must be used in combination with each other. Efforts must be made to educate stakeholders accordingly.
- The area for the AW-IPM programme, especially at its initiation, must be carefully selected. Besides geographic or topographic isolation, the target pest should already be well managed by conventional methods and sanitation, with growers who are progressive in their pest management outlook.
- Effective management of alternative fruit fly host plants and active orchard/vineyard sanitation at farm level is crucial to the success of an AW-IPM programme.
- There must be buy-in and long-term commitment to the programme by all growers in the selected area(s). Ideally, there should be a 'push-pull' approach by the stakeholders: SIT technologists should 'push' (advocate) AW-IPM (including SIT) where it is appropriate, with a simultaneous 'pull' (a willingness/receptiveness) for the SIT on the part of the growers.
- Good relations and communication between AW-IPM service providers, growers, and the broader public is crucial, and should be based on transparent real-time reporting on trap catches, and population levels and distribution.
- High specification infrastructure, equipment and human capital must be available to produce good quality insects. A good quality management system must be in place in the rearing and release facilities and must include regular internal and external audits of procedures, processes and performance.
- Ground releases of sterile *C. capitata* are not a long-term solution to area-wide population suppression. Above a certain scale, releases should be by air if at all possible.
- Programme managers must keep abreast of the latest international developments in the field of AW-IPM and make good use of knowledge and input from international experts.
- AW-IPM programmes are not quick fixes to a problem. Population reduction exercises can take a couple of seasons to show results. Stakeholder expectations should be managed in this regard.
- Applied research and development should be on-going, and all cost-effective improvements in procedures and processes should be implemented.

South Africa is now aiming to identify some of the existing deciduous fruit and table grape areas in the AW-IPM programme and manage them as areas of low pest prevalence.

The invasion of *B. dorsalis* is officially controlled by DAFF in South Africa and preparedness plans are in place to immediately initiate eradication programmes in case of outbreaks. Official control actions include quarantine, delimiting surveys and eradication measures with the application of the male annihilation technique and bait application (Manrakhan et al. 2012).

Although SIT application for *B. dorsalis* is not envisaged at this stage, future expansion of the fruit production areas to be covered by an AW-IPM approach, which will include the SIT for Mediterranean fruit fly, is planned (Manrakhan 2020). A new MoU with DAFF is planned, which will provide the necessary support for such expansion. This expansion of SIT activities will mainly be within areas where other area-wide control measures (e.g. monitoring and aerial baiting) are already being implemented to effectively suppress populations. Such areas have already been identified in the Northern and Eastern Cape Provinces.

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# THE CHINESE CITRUS FLY, Bactrocera minax (DIPTERA: TEPHRITIDAE): A REVIEW OF ITS BIOLOGY, BEHAVIOUR AND AREA-WIDE MANAGEMENT

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#### SUMMARY

The Chinese citrus fly *Bactrocera minax* (Enderlein) is a major pest of citrus in some Asian countries. It is a univoltine, oligophagous pest, which strictly infests *Citrus* species and varieties, and has an exceptionally long pupal diapause. *B. minax* has great socio-economic importance in China and its neighbouring countries because citrus production is a key fruit industry in these countries. We review the biology and management of this pest with a focus on its distribution, life cycle, diapause, behavioural ecology, and host preferences. We further review potential area-wide integrated pest management (AW-IPM) strategies, including chemical control, but also various eco-friendly, locally developed and adopted techniques applied mainly in China and Bhutan. After years of continuous efforts in AW-IPM of *B. minax*, significant progress has been achieved in suppressing *B. minax* populations to a level of less than 5% infested fruit and a 60-80% reduction in the use of synthetic insecticides against this pest in China.

*Key Words:* temperate fruit fly, life cycle, behavioural ecology, diapause, *Tetradacus*, Dacinae, oranges, *Citrus*, AW-IPM, China, Bhutan, IPM

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#### M. A. RASHID ET AL.

## 1. INTRODUCTION

Citrus fruits rank first across the world in the international fruit trade in terms of value (Liu et al. 2012; Srivastava 2012). The Chinese citrus fly *Bactrocera minax* (Enderlein) (Diptera: Tephritidae) is a major pest of *Citrus* spp. in China, Bhutan, India, Viet Nam and other neighbouring countries (White and Wang 1992; Dong et al. 2014a). It is a univoltine insect with a long pupal diapause that feeds solely on different *Citrus* species and varieties (Chen et al. 2016). The Chinese citrus fly is thought to be endemic to China as its presence could have been recorded in a poem written about 1000 years ago during the Song dynasty:

"The yellow oranges drop to the ground of the garden due to the wind in autumn. When the oranges opened, there were maggots inside the oranges instead of dragon" (Yang et al. 2013).

In the 1940s *B. minax* was only recorded in Guizhou and Sichuan, China (Chen and Wong 1943). Currently, it is reported to occur in the major citrus growing provinces of China (Chongqing, Guangxi, Guizhou, Hubei, Hunan, Shanxi, and Sichuan, see Fig. 1), climatically ranging from temperate to subtropical (Wang and Zhang 2009; Gao et al. 2013).

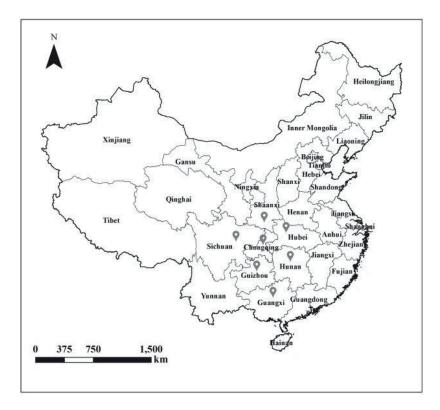


Figure 1. The occurrence and distribution of Bactrocera minax in China (indicated with dark marks showing location within different provinces).

Following a population outbreak in 2008 in south-western China, and the resulting heavy losses incurred by farmers, *B. minax* was removed from the national quarantine plant pest list in 2009 (Announcement No. 1216 of the Ministry of Agriculture of the People's Republic of China). Thus, the current management against *B. minax* in China has shifted from eradication to suppression.

## 1.1. Nature of Damage and Economic Importance

The adult female fly oviposits its eggs under the peel of green, immature citrus fruits with the aid of its elongated ovipositor (Fig. 2). The eggs hatch when fruit reach midlevel development and the larvae feed on the fruit flesh leading to premature ripening and fruit drop, ultimately resulting in economic damage and yield loss (Allwood et al. 1999; Liu et al. 2015). The larval stage is therefore considered as the most destructive life stage (Dorji et al. 2006). Heavy economic losses of USD 200 million were reported in 2008 due to the above-mentioned outbreak of *B. minax* in Guangyuan, Sichuan Province, China. Almost 1 million tons of oranges were destroyed during this flare-up. This outbreak and heavy infestation resulted in a ban on international trade of citrus commodities from China. The outbreak was reported by "The China Daily" in its headlines to highlight the serious damage to the citrus industry (Liu et al. 2015).

Similarly, *B. minax* is a major pest in the eastern Himalayan kingdom of Bhutan, where mandarin *Citrus reticulata* Blanco is one of the major fruit crops. Crop losses caused by *B. minax* infestation ranging from 35 to 75% are common in mid- and high-altitude orchards (>1100 m), and the fly is considered as one of the major barriers to citrus production (van Schoubroeck 1999; Dorji et al. 2006; Xia et al. 2018).



Figure 2. Female Bactrocera minax using its elongated ovipositor to lay its eggs into small green citrus fruit (20~30 mm diameter).

#### 1.2. Taxonomy, Distribution and Host Plants

The Chinese citrus fly was for the first time described in 1920 from specimens collected from Sikkim, India by Enderlein and named *Polistomimetes minax* (Thompson 1998). In 1940, the species was also collected in Sichuan Province, China. Drew (1979) provided a detailed description of the *B. minax* based on the specimens collected in 1920 and placing the species in the subgenus *Bactrocera* (Polistomimetes).

Subsequently, a lectotype of *B. minax* was ascribed to the subgenus *Bactrocera* (*Tetradacus*) by White and Wang (1992), who also recorded that *Bactrocera citri* Chen should be regarded as a junior synonym of *B. minax*. The fly is currently placed in the subgenus *Bactrocera* (*Tetradacus*), a small monophyletic clade evolutionary basal to all other *Bactrocera* species (Krosch et al. 2012).

Currently, *B. minax* is regarded as present in Bhutan, China, India (West Bengal and Sikkim), Nepal, and Viet Nam (Dorji et al. 2006; Drew et al. 2006). The host range of Chinese citrus fly is almost exclusively restricted to *Citrus* species and varieties. It has been recorded in citron *Citrus medica* L., lemon *Citrus limon* (L.) Burman f., meiwa kumquat *Fortunella crassifolia* Swingle, pummelo *Citrus maxima* (Burm.) Merr., sour orange *Citrus aurantium* L., sweet orange *Citrus sinensis* (L.) Osbeck, tangerine *Citrus reticulata* Blanco, navel orange *Citrus sinensis* Osb. var. *brasiliensis Tanaka*, pyunkyul *Citrus tangerina* Hort. ex Tanaka, grapefruit *Citrus paradisi* Macfad., and trifoliate orange *Poncirus trifoliata* L. (Nath 1972; Chao and Ming 1986; Liu et al. 2014). Among these, the preferred citrus host plant is sweet orange (Liu et al. 2014).

## 2. BIOLOGY AND LIFE CYCLE

#### 2.1. Seasonal Phenology

The phenology of *B. minax*'s life cycle may vary subtly depending on local climate conditions. However, the general pattern appears quite fixed. Based on the population of *B. minax* in Guizhou, China and in Bhutan, we summarize the life cycle of *B. minax* as follows:

1. The female oviposits eggs in small unripe fruit from mid-June to mid-July. Usually it takes two months for the eggs to hatch, much longer than other species in the *Bactrocera* genus; the eggs hatch in late August, after which the larvae go through three larval instars.

2. The larval stage lasts until the end of October, which is then followed by a pupal stage.

3. To survive the cold winter temperatures, the pupae enter a six-month overwintering diapause.

4. Adult emergence usually begins in May and mating starts about 25 to 30 days after adult emergence (Wang and Luo 1995; Dorji et al. 2006).

#### 2.2. Eggs of B. minax

The egg of *B. minax* is milky white, oblong and curved in shape, slightly pointed at one end and round at the other. It has a length of 1.1 to 1.5 mm and a maximum width of 0.2 to 0.4 mm (Sun 1961). Female *B. minax* lay eggs in clutches. Usually, each female oviposits about 14 to 17 eggs per oviposition event, with a maximum of 35 eggs per clutch. During its lifetime, a female can produce more than 100 eggs (Zhang 1989). After depositing eggs, the fruit surface is covered with juice around the oviposition wound. In the beginning, this juice is transparent to translucent for one to three days, then it gradually becomes yellow. The oviposition site bulges out, cracks and fruit skin around the oviposition site turns yellow to crimson-purple (Wang and Zhang 1993). Egg hatch starts in July and reaches its peak in late August (Xiong et al. 2016).

## 2.3. Larvae of B. minax

After eclosion, the larva feeds internally on the citrus flesh. The mature larva is milky white or pale yellow, 15 to 18 mm long, conical in shape and nearly transparent at one end. The mouth is equipped with sclerotized mouth hooks, and the body has 11 segments. The larval stage lasts 52 to 72 days depending on temperature, with an average of 63 days (Lu et al. 1997). Young larvae usually feed in a small group on a single fruit segment, later spreading to other segments: the average number of larvae in an infected fruit is 9.5 (Zhang 1989). Larval infestation leads to premature fruit-fall from October to November (Liu et al. 2015).

The larvae stay within the fruit for about 18-52 days after fruit drop. Such a long pre-pupal period is very unusual among the Dacinae and it suggests that long larval development occurs after fruit drop (Dorji et al. 2006). A mature, third instar larva usually leaves the fruit in the early morning and pupates within a day. Pupation starts in late October and reaches a peak in early to mid-November (Dong et al. 2013; Chen et al. 2016).

#### 2.4. Pupae of B. minax

The mature third instar larva pupates in the soil at 3 to 5 cm depth (Zhang 1989; Dorji et al. 2006). The long overwintering pupal diapause in *B. minax* is highly unusual within the genus *Bactrocera* and is considered as an adaptive strategy to survive the cold winter periods which occur in its native range (Fan et al. 1994). In Yichang city, where *B. minax* is present, the mean winter temperature usually ranges from 5 to 15 °C (Dong et al. 2013). The overwintering pupal phase lasts for 160-170 days, with the emergence of adults synchronised with the early fruiting season of citrus (Wang and Luo 1995). The pupa of *B. minax* is 9 to 10 mm long with a diameter of 4 mm, weight average of 77.5 mg. It is oval in shape and yellow-brown in colour. Prior to adult emergence, the pupal case becomes slightly dark brown (Zhang 1989; Wang and Luo 1995).

## 2.5. Diapause Termination in B. minax

Research on the development of efficient and sustainable *B. minax* management techniques is still very difficult because the fly has only one generation per year, accompanied by six months of pupal diapause (White and Wang 1992; van Schoubroeck 1999). This bottleneck made the mass-production of this fly very difficult, thereby limiting research capability and its potential use in area-wide integrated pest management (AW-IPM) programmes that have a Sterile Insect Technique (SIT) component (Lü et al. 2014). Therefore, options to break the diapause are considered essential for research and the development of the SIT package for this fly (see Section 4.2.3).

Pupal diapause is usually an evolved response in univoltine temperate tephritids in order to survive harsh environmental conditions and seasonal periods of host scarcity (Teixeira and Polavarapu 2001, 2005; Ragland et al. 2009; Papanastasiou et al. 2011; Moraiti et al. 2012). In *B. minax* pupal diapause is a vital strategy to tolerate cold stress and face the seasonal adversity. Research into the underlying mechanisms in terms of diapause termination, such as major cellular shifts, protein processing, differentially expressed genes and pathways, are still ongoing (Lü et al. 2014; Dong et al. 2014a; Wang et al. 2016, 2017).

Pupal diapause in *B. minax* is influenced by both chilling temperature and duration. A higher chilling temperature, coupled with longer chilling duration, results in a shorter pupal developmental time and improves the synchronisation of adult emergence (Dong et al. 2013). Apart from providing this chilling exposure, hormonal application of 20-hydroxyecdysone (20E) is considered a faster and more efficient method to break pupal diapause in *B. minax* (Dong et al. 2014a; Wang et al. 2014; Chen et al. 2016). Either injection or topical application of 20E can trigger a rapid termination of the pupal diapause in *B. minax* and the morphological changes are observed within 1 week at 22°C. On the tenth day after 20E treatment, the head, thorax and abdomen of the insect can clearly be distinguished, and the colour of body and eyes are milky (Chen et al. 2016). The 20E early-response genes, including *ecr*, *broad* and *foxo*, are up-regulated within 72 h of 20E exposure, indicating these genes are involved in diapause termination processes and pupal metamorphosis (Chen et al. 2016).

The gene sets involved in protein and energy metabolisms vary throughout earlylate- and post-diapause insects in response to cold stress. When diapause is terminated by 20E, many genes involved in ribosome and metabolic pathways are differentially expressed, which may mediate diapause transition (Dong et al. 2014a). The variation of transcriptomic and metabolomic profiles of pupae at five stages (pre-, early-, middle-, late-, and post-diapause) suggests major shifts in metabolism and signal transduction, as well as changes in the endocrine and digestive systems. Nine metabolites significantly contribute to the variation in the metabolomic profiles, especially proline and trehalose, which are well-known cryoprotective agents (Wang et al. 2017).

### 2.6. Adults of B. minax

The adult fly is 10 to 13.2 mm long, not including the female ovipositor, with a wingspan of ~10.8 mm (Drew 1979). The female possesses a long ovipositor of approximately 6.5 mm. The flies are of a brownish colour with yellow markings, the wings have a dark band along the outer margin, and the general appearance is wasp-like (Chen and Xie 1955). A morphological description of the adult is provided in Drew et al. (2007), who also note that the fly is probably the largest of all *Bactrocera* species (Fig. 2).

## 3. BEHAVIOUR AND ECOLOGY

Since the Chinese citrus fly is not attracted to methyl eugenol or cue-lure as are many species of the genus *Bactrocera*, thorough behavioural and ecological studies have been carried out over the past years in order to develop effective monitoring and control strategies in the long run.

## 3.1. Feeding Behaviour

It is well known that for most studied tephritid species, both males and females are anautogenous and forage for sugar and protein to fuel metabolic activities and to meet reproductive requirements (Aluja and Norrbom 1999; Drew and Yuval 2000; Taylor et al. 2013). Females need a protein diet for vitellogenesis and ovarian development (Harwood et al. 2015), while males feed on protein to reach sexual maturity leading to copulation which is crucial in achieving reproduction (Lushchak et al. 2013). Adult *B. minax* forage on non-host plants for honeydew, nectar, sooty mould and fruit juices to meet their dietary requirements during sexual maturation. The flies then shift to licking sooty moulds, bird faeces and, to a lesser extent, an unknown substance (probably leaf phylloplane bacteria and plant leachates) on citrus leaves and fruits during the mating and oviposition period (Hendrichs et al. 1993; Dong et al. 2014b).

## 3.2. Mating Behaviour

Male aggression and territoriality have been reported as a typical behaviour in some *Bactrocera* species (Shelly 1999; Weldon 2005; Benelli et al. 2014, 2015). For most tropical polyphagous *Bactrocera* species, males aggregate at a common place on foliage to attract and court females for mating, i.e. a non-resource-based lek mating system (Emlen and Oring 1977; Maan and Seehausen 2011). However, in *B. minax*, as in many temperate and oligophagous tephritids, male courtship behaviour is absent and the mating system is a resource-based defence polygyny, consisting of two phases: 1) males defend a resource (host fruit) (intrasexual selection), 2) where copulation takes place (intersexual selection) (Opp et al. 1996). In the wild, all mating events take place on citrus fruit (Dong et al. 2014b). Territory formation and copulation usually occurs on immature green fruits. It has been suggested that male flies that try to copulate in the vicinity of the ovipositional site have more chances to encounter and court receptive females (Prokopy 1976; Smith and Prokopy 1980).

Mating behaviour in *B. minax* is closely synchronised with the host fruiting season and has been described in the field as follows: (i) the male establishes its territory close to a potential oviposition substrate (citrus fruit); (ii) the female lands on the fruit and begins inspection and ovipositor probing on the fruit surface; and (iii), the male mounts and copulates with the female (Dong et al. 2014b). In this mating system females face trade-offs associated with the cost of additional, apparently unneeded matings on each fruit in return for access to resources. Like in the case of *Rhagoletis* species the resource is assumed to be the oviposition site (Opp et al. 1996; Opp and Prokopy 2000; Prokopy and Papaj 2000).

## 3.3. Oviposition and Host Preference

In tephritids, fruit flies use different cues for host finding behaviour and egg-laying behaviour. Usually, long-distance volatile chemicals are important before landing on host trees, visual stimuli act as short range once on the tree and contact chemicals on and inside host fruit influence female egg-laying decision.

*Bactrocera minax* oviposits solely into citrus fruits (Family *Rutaceae*) (Wang and Luo 1995; Dong et al. 2013). It is a large, powerful insect with a long ovipositor adapted for piercing through the thick skin of young, green citrus fruit (Liu and Zhou 2016). Visual cues including fruit shape, colour, and size are important for host finding (Prokopy and Owens 1983; Piñero et al. 2017). On the other hand, the egg-laying behaviour is greatly influenced by chemical stimuli, for example, semiochemicals, sugar content, levels of secondary plant compounds and physical properties of fruit (Bush 1969). The preference of *B. minax* oviposition on different citrus varieties is as follows, *Citrus sinensis* cv. Navel and *C. aurantium* > *C. sinensis* cv. Bintang, Amakusa and *C. reticulata* cv. Satsuma > *C. maxima* cv. Shatian > *C. reticulata* cv. Ponka. This ovipositional preference is positively correlated with larval survival and development; while in the field greater egg-laying occurs on those citrus fruits which are close to the surrounding vegetation and trees (Liu et al. 2014).

## 3.3.1. Visual Cues (Colour, Shape, and Size) for Oviposition

The hardness of a citrus fruit peel has an impact on the female insect's decision to oviposit (Lin et al. 2011). On the basis of egg oviposition marks on citrus fruit, it appears that *B. minax* significantly prefers to oviposit on the distal hemisphere rather than the basal hemisphere (Liu and Zhou 2016). Apart from peel hardness, the ovipositional behaviour of tephritid female flies is also influenced by fruit colour and shape (Alyokhin et al. 2000). It has been shown that tephritid flies respond to fruitmimics of the same colour or reflecting similar levels of light than host fruit of a particular fly species (Aluja and Norrbom 1999). For example, the Queensland fruit fly *Bactrocera tryoni* (Froggatt) showed attraction to spheres painted with cobalt blue pigments, which reflected the same UV spectrum as favoured blue-coloured host fruits occuring in its native rainforest environment (Drew et al. 2003). The oriental fruit fly *Bactrocera dorsalis* (Hendel) is attracted to white-yellow colour (Vargas et al. 1991) and the apple maggot *Rhagoletis pomonella* (Walsh), a pest of apples, is attracted to fruit-mimicking traps such as a red sphere (Duan and Prokopy 1995).

Both sexes of *B. minax* are attracted to orange or yellow/green spheres of 50 mm diameter (Drew et al. 2006). However, we found that *B. minax* adults prefer green over other colours, and this preference is significantly increased in sexually mature flies over immature flies (author submitted results).

## 3.3.2. Chemical Cues (Semiochemicals) for Oviposition

Chemical cues play an important role in foraging and oviposition of fruit flies (Sarles et al. 2015), and these chemicals are widely exploited in integrated pest management (Shrivastava et al. 2010). For example, the application of oviposition marking pheromone reduced *Rhagoletis cerasi* L. infestation up to 100% in cherry orchards (Katsoyannos and Boller 1976; Boller and Hurther 1998). Together with visual cues, semiochemical cues may also influence host finding and egg-laying of *B. minax*. The peel odours of different varieties of orange preferred by *B. minax* produce different volatile blends, including acids, aldehydes, alcohols, and oils. It is presumed that these volatile compounds directly influence the olfactory orientation of *B. minax* females. However, there is as yet no proof if these volatiles released by host plants have a direct impact on the oviposition preference of *B. minax* (Liu and Zhou 2016).

From the perspective of the biology, ecology and behaviour, *B. minax* is more reminiscent of flies in the temperate genus *Rhagoletis* than other pest species in the *Bactrocera* genus. Therefore, some components of pest management can be drawn from the extensive scientific literature on apple maggot fly *R. pomonella*, European cherry fruit fly *Rhagoletis cerasi*, and other fruit flies (Vargas et al. 2016). For example, the application of fruit volatiles in conjunction with visual traps may yield good results for the control of *B. minax*.

## 4. TOWARDS THE AREA-WIDE MANAGEMENT OF B. MINAX

Several different control tactics have been used to manage populations of the Chinese citrus fly. These include chemical control and "attract and kill" techniques using protein/food baits and fruit-mimicking traps. In addition, pilot trials of the Sterile Insect Technique (SIT) have been assessed for *B. minax* control (Wang et al. 1990; Wang and Luo 1995), and farmers in China and Bhutan have adopted locally developed suppression techniques.

## 4.1. Use of Chemicals

Pesticide applications (cover sprays) are the most commonly used conventional control practices against insects pests, especially in the case of outbreaks. Though they are effective in reducing the losses caused by fruit fly infestation, the negative impacts of pesticides on humans, the environment and non-target organisms have raised much concern. Different insecticides have been used to suppress *B. minax* populations, including phoxim, dichlorvos, chlorpyrifos, abamectin, botanically derived pesticides, and pyrethroids. Amongst these, abamectin and dichlorvos proved to have the highest and lowest toxicity, respectively. However, chlorpyrifos had the strongest effect on pupae, and phoxim had the strongest influence on emergence.

These chemicals are not recommended due to toxicity to non-target organisms and long residual effects, but they are effective against *B. minax* (Liu et al. 2015).

## 4.2. Eco-friendly Management

## 4.2.1. Field Sanitation

Field sanitation is an effective and important strategy to reduce *B. minax* populations for the next fruiting season. The collection of infested fruits from the ground every week from mid-September to late-November is essential to remove the breeding population from orchards. The protocol demands that these infested fruits are transferred into thick plastic bags (20-25 kg per bag), that can be supplemented with aluminium phosphide to facilitate the killing of larvae. However, this is not critical if the bags are kept in the field under the sun for 7~10 days. Finally, the rotting fruits serve as fertilizer. The plastic bags can be recycled and used again (Liu et al. 2011; Li et al. 2013).

In Bhutan, cultural practices such as the application of soil tillage, along with natural predation (pupae picking by birds), seem to have a role in reducing the number of pupae. However, this reduction is not significant. Thus, it is not recommended as the only control measure for reducing the overall *B. minax* population in the wild (Dorji et al. 2010).

## 4.2.2. Protein and Food Baits

Spraying a mixture of protein bait, with a small quantity of insecticide added, has proven to be an effective strategy for large-scale control of fruit fly populations (Conway and Forrester 2011). Bait sprays are effective for fruit fly population control as newly emerged females require protein to become sexually mature (Perez-Staples et al. 2007; McQuate 2009). In China, it is a widely accepted approach for the farmers to use vinegar, sugar and wine mixtures, plus detergent, as baits station/spots spray for the control of *B. minax*, which is simple and cheap (Zhou et al. 2012). Attractants such as GF-120, and other locally available commercial products, are also used for the suppression of the Chinese citrus fly. Fresh enzymatically-hydrolysed beer yeast (H-protein) liquid protein bait effectively attracted and killed more *B. minax* flies than GF-120 sprayed in the field (Zhou et al. 2012).

## 4.2.3. Use of Fruit-Mimicking Traps

Usually semiochemicals and plant derived volatiles are used to trap fruit flies (Díaz-Fleischer et al. 2014), but the males of *B. minax* are not attracted to either of the standard *Bactrocera* male lures (i.e. methyl eugenol and cue-lure) (Drew et al. 2006). Visual traps have been used as an alternative, and in Bhutan, both sexes of the fly were most attracted to green-yellow or orange fruit-mimicking spheres in the field (Drew et al. 2006). In recent years, a specific fruit-mimicking trap (spherical green sticky trap) has been developed and widely applied to monitor and control *B. minax* in China. After field deployment of spherical traps in sweet orange orchards in Zigui, China, the infested fruit rate dropped to 2.7% compared with 28.6% in untreated control orchards (Yi et al. 2015). Efficiency of the control effort was closely

associated with appropriate trap deployment density and time (Chen et al. 2017). Considering the cost of commercial traps, as well as efficacy, spherical green sticky traps with a diameter of 7 cm were recommended at a deployment rate of 20~30 traps per 1000 m<sup>2</sup> in citrus orchards (Chen et al. 2017; Gong et al. 2017).

#### 4.2.4. Sterile Insect Technique (SIT)

The SIT has been successfully used to manage fruit flies including *Ceratitis capitata* (Wiederman), *B. dorsalis*, the melon fly *Zeugodacus cucurbitae* (Coquillett) and the Mexican fruit fly *Anastrepha ludens* (Loew) (Calkins et al. 1994; Koyama et al. 2004; Dhillo et al. 2005; Enkerlin et al. 2017). In AW-IPM programmes that have an SIT component, the production of sterile insects in large numbers is of paramount importance (Enkerlin 2021). Sterilisation can be achieved by irradiation or genetic manipulation. Irradiating *B. minax* pupa two days before emergence with a dose of 90 Gy is recommended to ensure adult sterility (Zhang and Li 1990).

Due to the serious damage in major citrus production regions in China, a massrelease of sterilized *B. minax* flies was carried out in Guizhou, China in the late 1980s and early 1990s. In these pilot projects, mature  $3^{rd}$  instar larvae were collected in fruit from the field and then allowed to pupate in the lab (Wang et al. 1990; Wang and Luo 1995). Although the SIT trials in Guizhou Province resulted in a significant reduction of the pest population (Wang et al. 1990; Wang and Luo 1995), the technology is so far not integrated into *B. minax* control programmes in China. This is mainly related to problems with the development of mass-rearing methods due to technical barriers such as the extreme long pupal diapause period. This is the reason that there is currently no foreseeable plan for an SIT approach against *B. minax*, although the selection of non-diapausing strains is being explored.

### 4.2.5. Natural Enemies

Knowledge on the parasitoids associated with the Chinese citrus fly remains very scarce. Only one parasitoid, *Diachasmimorpha feijeni* van Achterberg, has been associated with this fly in Bhutan (van Achterberg 1999) and in China (authors' unpublished data). However, there is still no detailed information available on the parasitoid's interactions with Chinese citrus fly or the potential for parasitoid manipulation.

#### 4.2.6. From Pilot Trials to Area-Wide Management in China

Since 2009, the Ministry of Agriculture and National Agro-Tech Extension and Service Center of China has promoted and organized annual nation-wide conferences and training courses on tephritid control for local technicians and farmers, with the aim to educate and transfer new developments and technologies. Over the years, pilot trials of AW-IPM approaches against *B. minax* have been implemented in the provinces of Guizhou, Hubei, Hunan, Shanxi, and Sichuan where *B. minax* is a serious pest. The validated demonstration practices in the main citrus producing regions led to the establishment of a *B. minax* AW-IPM programme in China.

In the beginning of every year, the Ministry of Agriculture announces and issues online management programmes for *B. minax* control, along with that for other major agricultural pests, to facilitate the sustainable management of the pests. In the case of *B. minax*, the integrated environmental-friendly measures can be summarized as follows:

- First, population monitoring in April using field traps and pupal emergence cages, aimed at accurately identifying the timing of pupal developmental state and adult emergence, is carried out by local plant protection stations annually: this is critical to guide the implementation of control practices.
- Second, "attract and kill" strategies are applied from May to July including the systematic use of spherical green sticky traps, protein bait sprays and sugar-vinegar-wine liquid in bait stations or spot sprays.
- Third, field sanitation of habitats/orchards by removing the fallen and infested fruits and weeds from September to November has proved to be an important and effective population control measure. Recyclable plastic bags are widely adopted to keep-and-kill the mature larvae by a combination of hypoxia and heat.

Thus, through years of continuous efforts in AW-IPM of *B. minax*, significant progress has been achieved in suppressing *B. minax* populations to a level resulting in less than 5% of infested commercial fruit and a reduction in the use of synthetic pesticides by 60-80%. The *B. minax* AW-IPM programme will now be complying with national standards for 'green control', which results in notable direct and indirect economic, ecological and social benefits for China.

## 5. FUTURE PERSPECTIVES

With increased understanding of the biology and behaviour of *B. minax*, effective operational AW-IPM strategies against this pest have been established in China and Bhutan. However, SIT and natural enemies have not yet been exploited due to various biological and physiological obstacles. Great efforts are required to overcome these gaps for the future sustainable management of *B. minax*:

1. Mass-rearing of *B. minax* still remains a big challenge considering its univoltine and oligophagous traits. Future work should be focused on improving (a) understanding of egg hatch and (b) artificial diet formulation for the newly hatched young larvae.

2. Little information has been published on the insect-plant interactions of *B. minax*. It is widely known that the adults have a close relationship with *Citrus* spp., but how these adults utilize visual, olfactory and tactile cues to orientate to host plants for mating and oviposition has received very little attention.

3. Symbiotic organisms, including *Wolbachia* (Stouthamer et al. 1999), that affect biology and reproduction of *B. minax* should be characterized. A thorough screening of microorganisms by culture-dependent and high-throughput technology, in combination with related functional studies, will help to better understand the complex relationship between symbionts and *B. minax*. Such knowledge may lead to potential development and application of the Incompatible Insect Technique (IIT) (Zabalou et al. 2004).

4. Natural enemies have not been used as a component of AW-IPM against *B. minax* most likely because very little knowledge is available about them and their effectiveness. The possible synchronised diapause of parasitoids and *B. minax* pupae needs to be investigated. In addition, other agents such as predators or fungi causing pupal mortality deserve to be further investigated.

5. There are currently no effective semiochemical or plant derived volatile lures to attract *B. minax* males or females available. The volatile chemicals from the host fruits, as well as a sex attractant for monitoring and mass-trapping the Chinese citrus fly, urgently needs to be identified and exploited.

6. The growing published online resources on transcriptome, proteome, and genome of *B. minax*, RNA interference and CRISPR-Cas9 technologies targeting specific gene functions, will facilitate further investigations of molecular mechanisms responsible for the biology, behaviour, physiology and evolution of the Chinese citrus fly. The comprehensive understanding of *B. minax* is the most promising way to develop sustainable management of this economically important citrus pest in the long run.

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# AREA-WIDE FRUIT FLY PROGRAMMES IN LATIN AMERICA

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### SUMMARY

The chapter presents an overview of fruit fly (Tephritidae) pests and their economic impact in the Latin America and Caribbean (LAC) region, with a focus on the damage they inflict to horticultural production, as well as national and international commercialization. It reviews global trends that have favoured the establishment of several invasive fruit fly species in the region and the need to avoid further transboundary movement of invasive species. It also discusses the opportunities to increase fruit and vegetable production in the region despite the fruit fly problem and how integrated fruit fly management approaches within the framework of the International Plant Protection Convention (IPPC) can be applied for effective fruit fly control and to facilitate the international commercialization of horticultural commodities. The need for increased consumption of fruit and vegetables worldwide and in the Latin America and Caribbean region to mitigate the growing incidence of non-communicable diseases is discussed, as well as the trends in human population growth that will require increased provision of adequate diets. It also discusses the opportunities for Latin America and Caribbean countries to commercialize produce taking advantage of the global trend towards healthier food and less animal protein consumption. It presents available mechanisms for technical cooperation that facilitate technology transfer for more sustainable area-wide fruit fly management. It also provides case examples in the Latin America and Caribbean region of successful areawide fruit fly programmes that have increased production, opened markets and generated significant return on investment, as well as job opportunities. Future perspectives and challenges to address the fruit fly problem in the Latin America and Caribbean region are described.

*Key Words:* Tephritidae, *Anastrepha, Bactrocera, Ceratitis, Rhagoletis,* Caribbean, Central America, South America, invasive, horticultural exports, losses, pest free areas, fruit and vegetable demand, economic impact

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### 1. INTRODUCTION

Among the most important Sustainable Development Goals (SDGs), established by the United Nations in 2015, are: No Poverty, Zero Hunger, Good Health and Well-Being, and Life on Land (UN 2015).

Food is a common thread linking all 17 SDGs, given the interconnected economic, social and environmental dimensions of food systems (EIU 2018). Policy makers, politicians, health officials, and entrepreneurs are currently faced with the need to end hunger, achieve food security and improve nutrition, which are key steps toward sustainable development (UN 2016).

Food insecurity is one of the main challenges the world is facing to achieve the 2030 set milestones for at least these SDGs. Food insecurity is defined by the Food and Agriculture Organization of the United Nations (FAO) as the:

"situation when people lack secure access to sufficient amounts of safe and nutritious food for normal growth and development and an active and healthy life" (FAO/IFAD/UNICEF/WFP/WHO 2017).

Globally, the total number of people defined as moderately or severely food insecure was nearly 1.8 billion in 2015. On the other hand, over the next four decades, the world's population is forecast to increase by 2 billion people to exceed 9 billion by 2050 (Worldometers 2019).

Recent estimates indicate that to meet the projected food demand, global agricultural production will have to increase by 25-70% from its 2005-2007 levels, while nutrient losses and greenhouse gas emissions from agriculture must drop dramatically to restore and maintain the functioning of ecosystems (Hunter et al. 2017).

No one can question that pests, in particular insects, are significantly contributing to food insecurity worldwide. On average, insect pests are responsible for substantial pre- and post-harvest losses of horticultural products estimated according to some sources at least 18 to 20% at an annual estimated value of USD 470 billion (Sharma et al. 2017).

Losses are considerably higher in the developing world, especially, in the tropics of Africa, Asia and Latin America where pest control practices are much less effective and where most of the human population increase is expected. Moreover, the human population is facing an epidemic increase of non-communicable diseases due to insufficient consumption of fruit and vegetables among others (Hall et al. 2009). Treating these diseases between now and 2030 will cost USD 30 trillion globally (Nierenberg 2018).

Due to the climatic and soil conditions, the countries of Latin America could produce fruit and vegetables in larger quantities and at improved quality, taking advantage of the increasing demand from an increasing human population in the region and the world. The increased production of fruit and vegetables would further develop the horticultural industry creating diversification of the region's income. However, to take full advantage of the opportunity, a number of insect pest problems need to be addressed and overcome.

From the list of so-called key pests of agriculture, some fruit fly species are considered among the most devastating insects pests. They cause direct damage to horticultural production by reducing yields and indirect damage by disrupting national and international trade (White and Elson-Harris 1992). For example, the devastating impact of fruit flies in Africa, including direct damage to horticultural production and bans on trade, result in estimated losses of at least USD 2 billion annually on this continent alone (Ekesi et al. 2016). By preventing and significantly reducing fruit fly damage by means of effective and environment-friendly pest control measures, additional food supply would be made available, contributing to alleviating the large global deficit projected by 2050 (Hunter et al. 2017).

To reduce the transboundary risks and global burdens caused by invasive pests and at the same time continue facilitating international trade of horticultural products, the World Trade Organization (WTO) provides an instrument through its Agreement on Sanitary and Phytosanitary Measures (SPS). The SPS agreement is enforced by the International Plant Protection Convention (IPPC), based at FAO, and its contracting parties. The IPPC drafts and adopts *International Standards for Phytosanitary Measures* (ISPMs), aimed at providing a framework to the contracting parties for best practices in pest control and for mitigating pest risk in international trade. This includes a number of ISPMs specifically for fruit flies that have been recently harmonized in one suit of ISPMs to facilitate interpretation and use by IPPC contracting parties, as will be presented and discussed later.

This chapter presents a summary of the tephritid fruit fly pests and their economic impact in the Latin America and Caribbean (LAC) region. The main focus is on the damage they inflict to horticultural production and commercialization in the region, as well as on area-wide integrated pest management (AW-IPM) strategies for fruit flies that are applied, based on the international phytosanitary framework, for effective fruit fly control and to facilitate the commercialization of horticultural commodities.

### 2. FRUIT FLIES AND THEIR ECONOMIC IMPACT WORLDWIDE

### 2.1. Numbers and Current Distribution

To date, worldwide, approximately 4223 fruit fly species have been identified belonging to around 500 genera (Norrbom et al. 1998). From the total recorded so far,

250 species (less than 6%) are of some economic importance and from these, only some 78 species (1.8%) in 11 genera can be classified of major economic and quarantine significance (FAO/IAEA 2019). Nevertheless, the damage inflicted by these species to the horticultural industry worldwide is devastating, amounting to billions of US dollars every year.

These pests are found in almost all fruit and vegetable growing areas in all continents with the exception of the Antarctic (White and Elson-Harris 1992). The most important genera include: *Anastrepha* (Schiner), *Bactrocera* (Macquart), *Ceratitis* (MacLey), *Dacus* (Fabricius), *Rhagoletis* (Loew), *Toxotrypana* (Gerstaecker) (recently synonymised with *Anastrepha*) and *Zeugodacus* (Hendel). The genus *Anastrepha* and corresponding species are indigenous to the Americas, the *Bactrocera* to Asia and Oceania/Australia, the *Dacus* and *Ceratitis* to Africa, *Rhagoletis* to more temperate areas of Europe, North and South America, and the *Zeugodacus* to Asia.

Some fruit fly species belonging to these genera inflict serious economic damage to the horticultural industry in the countries and regions of endemism. With some exceptions (some temperate species), fruit fly pests are polyphagous, infesting a wide range of host species including some of the fruits and vegetables with the highest commercial value. Fruit flies have a high reproductive rate; thus, can produce several generations per year. These characteristics place fruit flies among the most important group of insect pests affecting horticultural production and trade worldwide. Fruit flies cause damage by laying eggs inside horticultural crops after which larvae hatch and feed on the mesocarp. As a result, infested fruits will prematurely drop from the tree or remain on the tree until harvest.

Depending on the fruit fly species and host, damage can range from 10 to nearly 100% of the crop when effective control practices are not implemented. In addition to the direct damage on fruits and vegetables, and significant yield reduction, fruit fly presence may seriously disrupt trade by quarantine restrictions imposed by importing countries which are free of the pests. This is illustrated by the recent case in the Dominican Republic where the Mediterranean fruit fly *Ceratitis capitata* (Wiedemann) was detected in 2015 and ten months later USD 40 million had been lost due to an immediate ban to Dominican exports imposed by importing countries (Zavala-López et al., this volume).

Fruit flies are effective invaders capable of spreading and establishing in regions outside their natural distribution range. For example, the oriental fruit fly *Bactrocera dorsalis* (Hendel) is an Asian species of great economic importance, infesting some 200 species of fruits and vegetables and causing direct losses estimated in millions of US dollars/year. Over the last decade, this species has invaded sub-Saharan Africa, causing losses ranging from 30 to 100% in some fruit crops such as mango, closing trade routes and triggering the loss of export opportunities that are vital for both the smallholder farmers and the more commercial fruit industry in the continent (Ekesi et al. 2016).

Other classic examples of fruit fly tephritid pest species spreading outside their natural distribution range and causing significant damage include the Mediterranean fruit fly, spreading from Central and North Africa to Europe, the Americas, Indian Ocean and Oceania (Gutiérrez-Samperio 1976); the melon fly *Zeugodacus cucurbitae* (Coquillet) spreading from Southeast Asia into Central Africa, and the Indian and Pacific Oceans (FAO/IAEA 2019); the peach fruit fly *Bactrocera zonata* (Saunders) spreading from Central Asia into North Africa and the Indian Ocean (FAO/IAEA 2019); the olive fruit fly *Bactrocera oleae* (Rossi) invading California and northern Mexico (Yokohama 2015; and the carambola fruit fly *Bactrocera carambolae* (Drew & Hancock) spreading from Asia to Suriname (Malavasi et al. 2000).

### 2.2. Factors that Contribute to Pest Movement and Establishment

Global trends including increased travel and trade, human movement and climate change are positively correlated with the significant increase in transboundary movement of invasive insect pests. Hulmes (2009) estimates that in the past 200 years the rate of non-native species introductions has increased 76-fold. The way these factors affect the spread of invasive pests is briefly described below.

### 2.2.1. Global Trade and Transport

With open market economies developing further through the integration of countries into new economic blocks and the expansion of current economic regions, transboundary trade and transportation of goods, including agricultural commodities, has been increasing significantly and is expected to increase further. The commercial movement of goods results in effective pathways for invasive pest species such as fruit flies, which are moving along with the commodities that they infest. For example, an analysis to assess the risk of Mediterranean fruit fly incursions into California, USA, indicated that the pathways with the highest probability for pest introduction were air passengers and crew baggage from foreign countries, express mail carriers from Asia and Hawaii to California containing packages with small amounts of fruits sent to relatives living in communities around Los Angeles Basin and cargo ships from Central America and other foreign countries (USDA/APHIS 1992).

### 2.2.2. Human Movement and Travel

Humans crossing borders to escape from violence, hunger and lack of opportunities in their countries, is undeniably increasing. Unintentional movement of insect pests by migrating humans is also an effective pathway for disseminating and introducing invasive pests. This is particularly true for fruit fly pests, that are often moved in fruits and vegetables that migrants carry as they travel for long distances across borders. Therefore, a rise in human movement and travel also results in an increased movement of invasive insect pest species that are not present in the country of transit or destination.

Humans are not only moving more frequently and in larger numbers because of social pressure or economic reasons, but also, as standards of living increase, travel has been increasing for segments of the population who travel long distance for leisure. For example, the number of tourists arriving to the Americas from all over the world has increased from 109 million in 1995 to 192.6 million in 2015, i.e. an increase of more than 75% in 20 years. Tourists often carry small amounts of fruits and vegetables to and from the site of destination, becoming effective pathways for invasive pests as well (Statista 2019).

#### 2.2.3. Climate Change

Climate change plays an increasingly important role in the survival and establishment of invasive pests (Pimentel 2002). Some pest species which are of tropical and subtropical origin, are now able to survive in regions of the world where climate has been gradually changing from cold winters with freezing temperatures to milder winters. This has in many cases, allowed pest binvasion and expansion of their distributions into new territories. For example, in the past 25 years, the Mediterranean fruit fly has been expanding its geographic distribution from North Africa and South Europe, to Central and East Europe as average winter temperatures are raising (Bjelis et al. 2016).

In addition, other factors related to climate change such as the increasing frequency of the El Niño Southern Oscillation (ENSO) in the tropics of Central and South America. Average temperatures increase, affecting biological cycles of pests by reducing development time and increasing the yearly numbers of generations and population density, thereby exacerbating pest problems. For example, the effects of the El Niño on Mediterranean fruit fly populations was assessed in Guatemala (Lira and Midgarden, this volume). This was done through modelling the increase by fractions in the average temperatures and observing the effect on population growth rates. A rapid and steep population growth exerts greater pressure over the containment barrier in southern Mexico that protects the Mediterranean fruit fly-free areas from infested areas in Central America. A positive correlation was observed during periods characterized as El Niño years, with increasing numbers of outbreaks in the Mediterranean fruit fly-free areas in northern Guatemala and the free areas in the state of Chiapas bordering Guatemala (Enkerlin et al. 2015).

Another relevant factor contributing to invasive pest introductions is the increase in the frequency and intensity of tropical storms, with high speed winds easily disseminating invasive pests over very long distances (Bhattarai and Cronin 2014). Calamitous events, such as hurricanes, tidal waves/tsunamis, droughts, floods, civil strife, often result in mass-displacement of people that can promote pest movement. Aid and assistance brought in as relief can pose risks if perishable provisions are brought in from countries infested with non-native fruit fly species.

#### AREA-WIDE FRUIT FLY PROGRAMMES IN LATIN AMERICA

### 3. FRUIT FLIES AND THEIR ECONOMIC IMPORTANCE IN THE LAC REGION

### 3.1. Fruit Flies in the Americas

Of the total number of tephritid fruit fly recorded, roughly, 23% (ca. 977 species) occur on the American continent. Most of them are present in the Neotropical region, extending from Mexico to Argentina (Norrbom et al. 1998). From this number of endemic species, around 15 (1.5%) are of economic significance.

Endemic to the more subtropical and tropical regions of the Americas is the genus *Anastrepha* with nine species known to be of economic significance, as follows: Mexican fruit fly *A. ludens* (Loew), West Indian fruit fly *A. obliqua* (Macquart), South American fruit fly (*A. fraterculus* Wiedemann) (a complex of about seven species with different hosts and behaviours), guava fruit fly (*A. striata* Schiner), sapote fruit fly *A. serpentina* (Wiedemann), inga fruit fly *A. distincta* Greene, Caribbean fruit fly *A. suspensa* (Loew), papaya fruit fly *A. curvicauda* (Gerstaecker) (formerly *Toxotrypana curvicauda*), and South American cucurbit fruit fly *Anastrepha grandis* (Macquart) (Weems Jr. et al. 2017; FAO/IAEA 2019).

In most of the countries in the Latin American region, these fruit fly species are still largely responsible for reduced horticultural production yields and for commercial production being mostly marketed domestically (Enkerlin et al. 1989). Exports of fruit and vegetable commodities are limited as many markets, including lucrative foreign ones, maintain restrictions from countries where major fruit fly pests are known to occur. Where horticultural trade does occur, it is largely among countries within the region that share similar pest problems or must undergo post-harvest commodity treatments.

For example, Mexico has more than one million hectares (ha) planted to fruit crops which are affected by fruit fly pests with an estimated annual production value of more than USD 4850 million (Gobierno de México 2018). The fruit industry is significantly hindered by fruit fly species, in particular by *A. ludens, A. obliqua, A. striata* and *A. serpentina*. In 1991, the annual direct damage caused by these indigenous fruit flies was estimated at more than USD 230 million despite control activities (Reyes et al. 1991). To address this serious constraint in a more systematic and coordinated way, the National Plant Protection Organization (NPPO) of Mexico established in the early 1990s the National Fruit Fly Campaign, which has effectively controlled fruit fly pests on an area-wide basis in large regions of the country, generating a very significant return on investment as discussed in Section 6.1.2 of this chapter (Reyes et al. 2000; IICA 2010; Gutiérrez-Ruelas et al. 2013).

The genus *Rhagoletis* is also endemic to the Americas but is generally present in more temperate regions. The species of economic concern include: the eastern cherry fruit fly *R. cingulata* (Loew), the walnut husk fly *R. completa* Cresson, the blackbodied cherry fruit fly *R. fausta* (Osten Sacken), the western cherry fruit fly *R. indifferens* Curran, the blueberry maggot *R. mendax* Curran, and the apple maggot *R. pomonella* (Walsh).

All these species inflict serious economic damage to fruit production and commercialization. For example, a report has indicated that the overall domestic and export cost of *R. pomonella* could be USD 392.5 million annually in the state of Washington, USA (DEFRA 2018). The total reduction in net returns to producers from *R. completa* injured walnuts amounts from 50 to 75 % (Boyce 1934).

In addition to the endemic fruit fly species, a number of non-native invasive species have invaded and become established in some parts of the Latin America and Caribbean region and are causing severe direct and indirect losses to the horticultural industries in the region (Fig. 1).

From these species, the Mediterranean fruit fly is the most devastating as it is capable of infesting more than 300 species of fruits and vegetables (USDA/APHIS 2019) and it is subject to rigorous quarantine restrictions by importing countries free from the pest (Fig. 2). This species is responsible for economic losses estimated at USD 242 million/year in Brazil alone (Oliveira et al. 2013). Moreover, citrus in Central America covers an area of approximately 84 000 ha; the damage without control has been estimated at 28% in orange, 50% in tangerine and 24% in grapefruit. The combined damage of the endemic *A. ludens* and *A. obliqua* and the non-native *C. capitata* in mango, *Mangifera indica* L. amounts from 15 to 20% when left without control actions (Daxl 1978; Rhode et al. 1971; Vo et al. 2003).

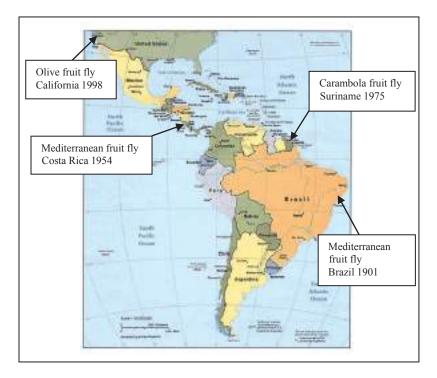


Figure 1. Introductions, establishment and spread of non-native tephritid fruit fly species in the Americas.



Figure 2. Coffee berries are a host of the Mediterranean fruit fly. The coffee belt extends from southern Mexico to Brazil (photo from W. Enkerlin).

Despite this serious fruit fly problem, as will be presented in Section 7, some countries in the Latin America and Caribbean region have been able to effectively control fruit fly pests and overcome trade barriers by establishing and maintaining fruit fly free and low prevalence areas through the systematic implementation of phytosanitary strategies, including AW-IPM strategies (Hendrichs et al. 2007), that in certain situations also integrate the Sterile Insect Technique (SIT).

### 3.2. Current Situation of Horticultural Production in the LAC Region

The Latin America and Caribbean region has a surface area of 19.2 million  $\text{km}^2$  that represents nearly 13% of the Earth's surface and is currently home to 640 million inhabitants. The region has a wide range of climatic conditions, ecological zones, soil types and an overall positive balance in water supply (Peel et al. 2007).

The favourable subtropical and tropical conditions of Latin America and Caribbean countries allow them to produce fruits and vegetables for their inhabitants as well as for export markets. The Latin America and Caribbean region is a net exporter of agricultural commodities to the world, with ca. 16% of global food and agriculture exports between 2012 and 2014. The Latin America and Caribbean region has always maintained a strong comparative advantage in agricultural and in particular in horticultural production (World Bank 2013).

The sustainability of food production is a desirable path that is being promoted, and the production of seasonal vegetables, and in particular fruits in orchards, not only allows regenerative agricultural processes (soil recovery, protection from erosion, CO<sub>2</sub> capture) to take place, but also results in a low ranking with respect to ecological footprints among foods (Nierenberg 2018).

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Many Latin America and Caribbean countries have steadily increased their fruit and vegetable exports during the past years and have enjoyed increasing investments in the production of fruit and vegetables (Prensa Libre 2019). Experts have especially increased from fruit fly free and low prevalence areas, where investments of the horticultural industry are protected from the presence of native and non-native fruit fly species (SARH/DGSV-USDA/APHIS 1990; SAG 1996; SAGAR 1999; Braga Sobrinho et al. 2004; Noe-Pino 2016).

### 3.3. LAC Region – The Need to Increase Horticultural Production

Despite the progress made in the past decades in fruit and vegetable production in the Latin America and Caribbean region, it is certainly insufficient to face the challenges ahead, including satisfying the demand of an increasing population. The projected increase in human population in Latin America alone will require additional production of fruit and vegetables in order to provide it with adequate diet options (Fig. 3). The countries of the Latin America and Caribbean region are suffering in different degrees the 'triple burden' of malnutrition, which consists of:

- Undernourishment, affecting 5.5% of the population in the region is (ca. 35 million)
- Micronutrient deficiencies, and
- Overweight and obesity.

Malnutrition results in an increase of non-communicable diseases, the incidence of which needs to be reduced (FAO 2017a). Fruits and vegetables are loaded with vitamins, minerals, antioxidants and fibre, which are considered to reduce many health problems, including cancer. However, fruit and vegetable consumption, even though less acute than in some other subtropical/tropical regions of the world, remains generally low (Hall et al. 2009).

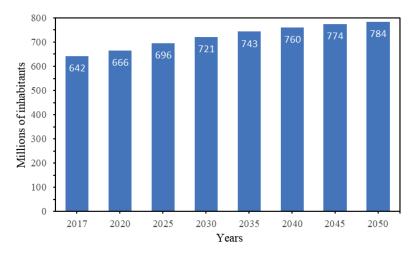


Figure 3. Projected human population increase in Latin-America 2017-2050 (based on data available at Worldometers 2019).

Fresh fruits and vegetables consumption could be increased by:

1. Subsidies to reduce the fruit and vegetable prices

2. Expanding access to healthy diets and income generation strategies (FAO 2017a), and

3. Introducing new educational programmes regarding nutrition, and modification to policies and best practices, which range from the development of eating guidelines to imposing taxes that discourage unhealthy consumption patterns (EIU 2018). Combining all or parts of these strategic actions might be needed to address the projected increase in human non-communicable diseases, which stresses the need for visionary and timely decision-making, including policies to reduce food losses and waste.

### 3.4. Opportunities for Horticultural Exports

As mentioned in previous Sections, the Latin America and Caribbean region produces and exports large amounts of produce for international markets. Currently, due to the awareness of the effects of food choices on human health, more customers are changing their preferences towards more healthy food. This is also favoured by rising incomes in some countries (i.e. per capita vegetable consumption has significantly risen in China).

World-wide, there are a number of countries with a population that enjoys increased life expectancy and improved health due to the increased consumption of healthy foods. This mega-trend provides the opportunity for producers, entrepreneurs and existing companies to offer their services for these expanding export markets. Also, countries need to review their internal policies and nutrition guidelines, which combined with education and school lunches, could open additional opportunities for sustainable horticultural production.

Food safety is a key concern for production and handling of fruit and vegetable produce, as unsafe food remains a major cause of disease and death (WHO 2015). There is also an increased need for safe non-residual pest control tactics which include the area-wide application of environment-friendly and therefore sustainable tactics (e.g. the EU food safety public standards are established in the General Food Law or Regulation (EC 2002).

The Latin America and Caribbean region needs to strengthen its business sector, while embracing sustainable farming. Such actions will build up agricultural diversification, increase fruit and vegetable production and exports, and further job creation, i.e. factors that contribute towards a prosperous and stable economy.

For the past 100 years or more, the international trade of horticultural products has been subjected to phytosanitary regulations. Given the presence of fruit fly pests, postharvest disinfestation treatments were, since the 1920's, the only alternative to overcome export barriers. Trade was prohibited for fruit and vegetable commodities for which no post-harvest treatment was available, or it was too costly, or reduced the quality of the product, or for which there was no feasibility for the establishment of a pest free area, the first of which was only recognized in 1988 in Sonora, Mexico (SARH/DGSV-USDA/APHIS 1990; SAGAR 1999; USDA 2018). Given this situation, and with the aim of facilitating agricultural international trade where the phytosanitary risk is mitigated to an acceptable level, an alternative is the SPS agreement of the WTO that includes the possibility to combine areas of low pest prevalence with other pest mitigating measures in a "systems approach" that provide a negligible risk to the importing countries. Section 5 presents a brief description of the phytosanitary approaches available to reduce pest risk and increase the potential for exporting horticultural products that are fruit fly hosts, following ISPMs of the IPPC.

## 4. CHALLENGES TO PREVENT THE INTRODUCTION AND ESTABLISHMENT OF PESTS AND DISEASES AND THEIR CONTROL

In order to maintain and increase productivity and food security, in particular for middle- and low-income countries, national and regional plant protection organizations (RPPOs) need to actively participate in the prevention of transboundary movement of pests and diseases. Prevention has proven to be the most cost-effective strategy, minimizing the use of insecticides, negative environmental impacts and reducing high costs associated with remedial control practices (FAO 2017a). Prevention and rapid response might be the only way to protect crops, while other options may be limited or more expensive (i.e. desert locust prevention in western and north-western Africa is estimated at USD 3.3 million per year, while a control campaign during 2003-05 was close to USD 600 million, or equivalent to 170 years of prevention (FAO 2014).

Prevention, suppression and eradication of pests and diseases will require greater coordination at the international and regional levels to understand the risks and strategize how to deal with them. One example of such coordination and cooperation is the Practical Arrangement subscribed between the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture and the regional plant protection organization for Central America, namely the Organismo Internacional Regional de Sanidad Agropecuaria (OIRSA). This collaboration allows training of plant protection staff from the Central American region, through the implementation of surveillance practices and emergency response drills against incursions of invasive species of quarantine significance such as the oriental fruit fly and other major fruit fly pests of economic importance.

Other challenges include the timely detection and reporting of pest presence that can prevent the national or regional spread of pests. This is feasible through the development and adoption of regional surveillance databases that include pest alert systems and discussion platforms.

Late reporting has had unfortunate consequences, such as the carambola fruit fly introduction in Suriname in 1975 which spread from there to other areas in French Guiana and other countries in the region (Marchioro 2016).

### 5. APPROACHES TO OVERCOME TRADE BARRIERS

Countries need to aim at effective fruit fly surveillance and control to increase quality and production of horticultural products, and to foster opportunities for trade in international markets. To achieve these goals, the initial step is to apply basic IPPC standards; these include:

- *ISPM No. 6 "Surveillance"* that includes general surveillance aimed at providing NPPOs with elements such as phytosanitary import/export requirements, documentation on pest free areas, pest reporting and eradication strategies, and specific surveillance aimed at providing technical information such as pest detection and population dynamics in an area (FAO 2018).
- *ISPM No.* 8 "*Determination of a Pest Status in an Area*" aimed at providing information on the presence or absence of a pest (FAO 2017b), and
- *ISPM No. 11 "Pest Risk Analysis for Quarantine Pests"* aimed at providing details to conduct pest risk analysis (PRA) to determine if pests are of quarantine importance. It describes the integrated processes to be used for risk assessment as well as the selection of risk management options (FAO 2019a).

If it has been determined that the pest is absent from the target area (ISPM No. 8) or if the commodity of interest is not a fruit fly host, then the commodity should not be subjected to quarantine regulations for trade. Host status is assessed through applying *ISPM No. 37 "Determination of host status of fruit to fruit flies (Tephritidae)"* (FAO 2019b). For example, by applying a research protocol mutually agreed upon by the NPPOs of Mexico and the USA, findings showed that Hass avocado produced in Mexico could be recognized as a non-fruit fly host of *Anastrepha* species of concern. As a result, a quarantine that had been imposed for 82 years by USDA on Hass avocado exports was revoked in 1997, resulting in the opening of the USA market (Enkerlin et al. 1993; Aluja et al. 2004; Gutiérrez-Ruelas et al. 2013). Over one million tonnes of Hass avocado are exported annually to the USA under a bilateral workplan subscribed by the NPPOs of Mexico and the USA, generating over two billion USD per year, creating thousands of jobs and a high demand for materials and services.

If, on the contrary, the regulated pest of concern is present in the area and the commodity is a host (even if only a conditional host), then pest management strategies need to be applied against the pest to mitigate the risk posed by it to the importing country.

In the case of fruit fly pests, the IPPC has adopted a suite of fruit fly-specific ISPMs (IPPC 2017). Depending on the objective of the fruit fly control programme and the situation of the pests and hosts in the area, the following ISPMs may be applied:

1. ISPM No. 37 on determination of host status as described above,

2. ISPM No. 26 "Establishment of Pest Free Areas (PFA) for Fruit Flies (Tephritidae)" (FAO 2015), and

3. *ISPM No. 35 "Systems Approach for Pest Risk Management of Fruit Flies (Tephritidae)"* (FAO 2019c). This international standard combines the application of monitoring and control practices to mitigate pest risk that may or may not include a post-harvest treatment.

These fruit fly ISPM's have a number of technical annexes which are considered to be part of the standards, such as *Annex 1 of ISPM No. 35 "Establishment of Areas of Low Pest Prevalence (ALPP) for Fruit Flies (Tephritidae)"* and *Annex 3 of ISPM No. 26 "Phytosanitary Procedures for Fruit Fly Management"*.

In addition, the fruit fly ISPMs refer to a series of (non-binding) appendices which provide additional information such as the *Appendices of ISPM No. 26 "Fruit Fly Trapping"* and *"Fruit Sampling"*.

Through its standard implementation facility, the IPPC promotes the provision of technical assistance to contracting parties with the objective of facilitating the implementation of the international standards. This can be done by the NPPOs together with stakeholders through a number of technical cooperation mechanisms, as will be presented in the following Section.

### 6. TECHNICAL COOPERATION MECHANISMS

Effective implementation of SPS measures requires capabilities and competencies in the public and private sectors of each country, as well as good communication and collaboration between the various public sector organizations involved and with the private sector. Typically, governments are responsible for the establishment and oversight of an enabling regulatory framework for food safety, animal health, veterinary services, plant health and/or trade, and for ensuring the compliance of agrifood exports with SPS requirements of trading partners. Ultimately, it is the private sector that plays the leading role in food and agricultural production and trade, and that is responsible for meeting SPS requirements in export markets.

The IAEA and FAO support Member States in creating capacities for implementation of SPS measures, including transferring technologies for fruit fly prevention and control through technical cooperation projects. Technology transfer regarding the area-wide application of the SIT is carried out with the support of professional staff of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture.

In the Latin America and Caribbean region, this mechanism for technology transfer has been used in support of SIT technology transfer since the late 1970's when it was transferred to southern Mexico for the eradication of the invading Mediterranean fruit fly. This represented the first large-scale use of the SIT technology for fruit flies and resulted in the eradication of the pest from 800 000 ha in the state of Chiapas, Mexico (Hendrichs et al. 1983). Previously, in 1975, a small infestation of this pest in Los Angeles, California, USA, was eradicated using sterile flies reared in the USDA-ARS Hawaiian Fruit Fly Laboratory and shipped to California (Harris 1977).

Other more recent mechanisms that support capacity building and technology transfer to Member States are the Standards and Trade Development Facility (STDF 2019), as well as the Collaborating Centres (CC) scheme, through which the IAEA officially recognizes the technical capacity of specific institutions in Member States. One relevant CC is the Programa Nacional de Moscas de la Fruta in México. Through this CC, international training courses on fruit fly AW-IPM are regularly offered, expert advice is provided and technology for fruit fly surveillance and control is advanced through research and development.

Regarding the STDF, it provides a platform for organizations to come together to discuss SPS capacity building needs, share experiences and good practice, leverage additional funding, and work on coordinated and coherent solutions, including solutions to fruit fly problems. The goal of the STDF is to increase the capacity of developing countries to implement international SPS standards, guidelines and recommendations, and hence the ability to gain and maintain market access.

### 7. CASE STUDIES OF FRUIT FLY AW-IPM PROGRAMMES IN THE LAC REGION

Some cases of successful fruit fly area-wide programmes, several of which have been supported through the above mechanisms and international standards, are presented and described in this Section.

### 7.1. Guatemala-Mexico-USA Moscamed Programme for the Containment and Eradication of the Mediterranean Fruit Fly

After invading Costa Rica in 1955 and gradually spreading through Central America, most likely in infested fruits carried in small amounts by migrants moving north looking for better living conditions and through commercial trade of horticultural products throughout the region, the Mediterranean fruit fly was first detected in Guatemala in 1975. The establishment of the pest in Guatemala posed a significant threat to high value fruit and vegetable industries, as well as neighbouring countries of Belize, Mexico, and the Caribbean. It also posed a serious threat to producers in the USA.

To address this threat, the NPPOs of Guatemala, Mexico and the USA, established bilateral cooperative agreements and, in 1977, created the Mediterranean fruit fly eradication programme (Moscamed Programme). The Moscamed programme based its control strategy on the area-wide integration of the SIT with other methods to contain and eradicate the pest. The SIT technology was transferred to the Moscamed Programme through technical cooperation projects with the IAEA and FAO, and the technical guidance of the Joint FAO/IAEA Division as well as with the support of USDA-ARS in Honolulu, Hawaii. A large Mediterranean fruit fly mass-rearing and sterilisation facility (with a production capacity of 500 million per week) was constructed (1977-78) in southern Mexico at Metapa, Chiapas (a state on Mexico's southern Pacific coast bordering Guatemala) with the first sterile flies released in 1979 (Fig. 4).



*Figure 4. Fruit fly mass-rearing and sterilisation facilities at Metapa, Chiapas, Mexico SENASICA-SAGARPA (photo from Moscamed Programme; reproduced with permission).* 

Four years later (in 1982), after releasing billions of sterilized flies, the Mediterranean fruit fly was declared eradicated from approximately 800 000 ha in Chiapas (Hendrichs et al. 1983; Enkerlin et al. 2015).

A second rearing facility funded by USDA producing Mediterranean fruit fly standard strain (San Miguel Petapa) was inaugurated in 1983 in Guatemala. In 1985, a modular section was added at this location (Tween 1986).

The Petapa facility maintained its production of standard (non-genetic sexing) strains until it was superseded years later by a second and much larger mass-rearing facility in El Pino, Guatemala (Fig. 5), which was designed and constructed (1994-95) using modular design for the production of a genetic sexing strain, i.e. based on a temperature sensitive lethal (*tsl*) mutation. The mass-production capacity of this facility is up to 2000 million sterile males per week.



Figure 5. El Pino Mediterranean fruit fly mass-rearing and sterilisation facility in Guatemala (photo from Moscamed Programme; reproduced with permission).

This regional programme has greatly contributed to maintaining the biological containment barrier and the goal of protecting the Mediterranean fruit fly-free areas in Petén, Guatemala and preventing northern spread of the pest (Fig. 6) (Enkerlin et al. 2017).

To continue to maintain the barrier today, the Mediterranean fruit fly mass-rearing facilities in Mexico and Guatemala currently rear over 1.5 billion insects per week (Figs. 4 and 5). The El Pino and Metapa facilities also shipped sterile pupae to support SIT programmes in a number of countries, including preventive release programmes in California and Florida, USA; eradication programmes in Chile, Dominican Republic, Mexico (Tijuana and Manzanillo), and the USA; and, for some periods suppression programmes in Argentina (Patagonia), Israel, and more recently in Ecuador (DIR-SIT 2018). This has been done through bilateral arrangements between the NPPOs of interested countries, and some also received technical support within the framework of IAEA technical cooperation projects.

Keeping the USA and Mexico Mediterranean fruit fly-free has created favourable conditions for the development of multi-billion dollar horticulture industries in these countries and paved the way to increase production and export of fruits and vegetables from Guatemala and Belize (IICA 2013). The return on investment measured in macroeconomic terms through a benefit-cost analysis, gives an extremely favourable 150 to 1 benefit-cost ratio (BCR) in spite of the programme's annual operational cost of ca. USD 35 million (Enkerlin et al. 2015, 2017; Enkerlin 2021).

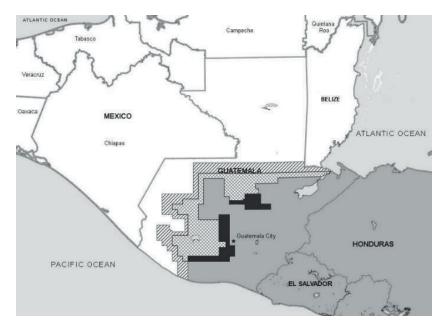


Figure 6. Location of the Mediterranean fruit fly containment barrier in Guatemala in 2015 (reproduced from Enkerlin et al. 2017).

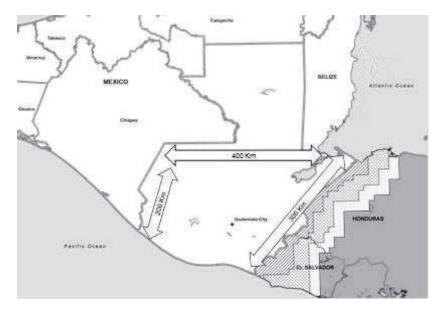


Figure 7. Potential, more sustainable, containment barrier at the El Salvador and Honduras border with Guatemala (reproduced from Enkerlin et al. 2017).

A prospective benefit-cost analysis (IICA 2013) projected for the period 2012 to 2021, with the corresponding investment, presented a scenario where using current improved technology the Mediterranean fruit fly could be eradicated from Guatemala and the containment barrier moved to the border with El Salvador and Honduras, where the Central American Isthmus begins and the length of the containment barrier would be reduced by half (Fig. 7).

Moving the containment barrier would make it more sustainable in economic terms, but also in technical terms, in view of the easier topography for programme activities, significantly reduced host areas, and quarantine measures in place at land border crossings, seaports, and airports (IICA 2013).

### 7.2. The Mexican and West Indian Fruit Fly Suppression and Eradication Programme – The National Fruit Fly Programme in Mexico

In 1992, the Mexican federal government (SENASICA-SAGARPA) approved the National Fruit Fly Programme for the control of indigenous fruit fly species, primarily the Mexican fruit fly and the West Indian fruit fly. The programme applies an AW-IPM approach including the SIT (Reyes et al. 2000; Gutiérrez-Ruelas et al. 2013). Strategic alliances between federal and state governments, and the horticultural industry, proved to be an effective way to operate a national programme aimed at suppressing and eradicating populations of fruit flies of economic significance for the establishment of ALPP and PFA.

For the area-wide application of the SIT against these two major *Anastrepha* species, a multi-species mass-rearing and sterilisation facility was built in Metapa, Chiapas, Mexico (Fig. 4). The federal government supplies the sterile flies and provides the infrastructure for their processing and release, the state governments contribute financial resources for operations, and the horticultural industry implements activities in the commercial orchards, including trapping and fruit sanitation in orchards. In 1997, these two fruit fly species were eradicated from more than 35 000 ha of commercial plantations of citrus, mango, apple, and peach in northwestern Mexico, completely freeing the states of Chihuahua, Sonora, Baja California, and Baja California Sur of these pest insects (SAGAR/IICA 2001).

In 2001, after fruit fly eradication in north-western Mexico was officially declared and PFAs established, the direct benefits (reduced fruit fly damage and increased yield) amounted to USD 25 million per year. In addition, during the same period, the benefits obtained from the price differential paid by export markets and savings in post-harvest treatments, totalled approximately USD 35 million. Thus, the total benefits from these fruit fly PFAs over four years amounted to USD 60 million, with a total programme implementation cost of USD 4 million over this period, resulting in a benefit-cost ratio (BCR) of 7.5 to 1 (SAGAR/IICA 2001).

An economic study covering the period from 1994 to 2008, shows that the return on investment of the National Fruit Fly Programme for the mango industry resulted in a BCR of twenty-two dollars for each dollar invested (22 to 1) and a net revenue at present value (NPV) of USD 1.1 billion, and for the citrus industry a BCR of 19 to 1 and a NPV of USD 2.0 billion (IICA et al. 2010).

### 7.3. Chile's National Fruit Fly Programme

Chile's fruit fly-free status has allowed the development of one of the most important export-oriented horticultural industries in the world. To obtain this status and protect this valuable asset, the Government of Chile, through the Agricultural and Livestock Service (SAG) of the Ministry of Agriculture, created Chile's National Fruit Fly Programme in 1980. Its objectives have been to free Chile of the Mediterranean fruit fly, established in the north, and to prevent the introduction and establishment of any fruit fly species of economic importance, including the Mediterranean fruit fly and other pest species of the genera *Anastrepha* and *Bactrocera* anywhere else in Chile (Olalquiaga and Lobos 1993; Rodríguez et al. 2016).

The National Fruit Fly Programme in Chile operates through a centralised organizational structure under the Ministry of Agriculture, that includes a mass-rearing facility in Arica near the border with Peru (Fig. 8).



Figure 8. Mediterranean fruit fly mass-rearing and sterilisation facility in Arica, Chile (photo from R. Rodríguez SAG Chile; reproduced with permission).

In addition, as part of a regional approach to the fruit fly problem, the Government of Chile has also subscribed bi-national agreements with neighbouring Argentina and Peru (Wedekind 2007). Chile has achieved and maintained its fly-free status by implementing two major strategic activities:

- An effective national and international quarantine system (including interprovincial quarantine road stations when there is an outbreak and international quarantine at ports of entry), and an extensive and highly sensitive fruit fly-trapping network to detect fruit fly introductions at an early stage. Numerous outbreaks of non-native fruit flies, mainly the Mediterranean fruit fly, have been eradicated through the effective execution of an emergency eradication plan based on detecting and eradicating infestations (McInnis et al. 2017; Shelly et al. 2017). A *B. dorsalis* outbreak on Easter Island was eradicated in 2011 at a cost of USD 100 000 (AGROMEAT 2011; FAO/IAEA 2011).
- In Arica province, at the border with Peru, a containment barrier that integrates the release of sterile males (Fig. 8) to avoid the natural or artificial spread of Mediterranean fruit fly populations into northern Chile, in order to protect the main fruit and vegetable production areas in the central and southern parts of the country.

After six years of an intensive integrated area-wide programme based on the SIT, the Mediterranean fruit fly was eradicated from Arica province in 1995, and all of Chile was declared a fruit fly-free country (SAG 1995, 1996). USDA-APHIS recognized Chile as a pest free area for Mediterranean fruit fly in 2010 (Federal Register 2010).

As a result of the fruit fly-free status, exports have grown to 320 million boxes of fruits per year, mainly table grapes, apples, stone fruits, kiwis, and avocados, valued in 2016 at USD 4000 million (ASOEX 2018).

### 7.4. Patagonia, Argentina – Mediterranean Fruit Fly PFA

A programme to eradicate Mediterranean fruit fly from fruit production areas in northern Patagonia was launched in late 1996 by the Programa de Control y Erradicación de Mosca del Mediterráneo (PROCEM-SENASA) and the Fundación Barrera Zoofitosanitaria Patagónica (FUNBAPA) (Borges et al. 2016). Mediterranean fruit fly eradication activities started in 2001 and concluded in 2004 with the official declaration of Patagonia as a Mediterranean fruit fly-free area (Guillen and Sanchez 2007).

Trading partners, including Chile, Mexico and the USA, recognized Patagonia as a Mediterranean fruit fly-free area (Borges et al. 2016). Eradication was achieved through an intensive area-wide programme integrating the SIT. Sterile flies were shipped from the mass-rearing and sterilisation facility located in the Province of Mendoza, Argentina (De Longo et al. 2000). As a result of the eradication of Mediterranean fruit fly, costly quarantine treatments could be eliminated for most of the three million boxes of quality pears and apples that this region exports yearly. Other mayor benefits included gaining access to previously closed markets (Villareal et al. 2018). Of fundamental importance to protect this pest free area is the extensive quarantine barrier that is effectively operated by FUMBAPA (Fig. 9) (Wedekind 2007).



Figure 9. Inspection at a FUNBAPA quarantine road station in Patagonia, Argentina (photo from E. Rial, PROCEM Patagonia, reproduced with permission).

### 7.5. Mediterranean Fruit Fly-Free Places and Sites of Production, Honduras, Central America

Through a careful review of international phytosanitary standards, the National Health and Agrifood Safety of Honduras (SENASA) determined that the pest risk mitigation scheme that could apply to the melon (*Cucumis melo* L.) production sites of Montelibano (400 ha) and Santa Rosa (800 ha) was ISPM 10 "*Pest free places of production and pest free production sites*". Following international fruit fly trapping guidelines (Appendix 1 of ISPM 26; IAEA 2013), SENASA established a fruit fly surveillance network for these sites in July 2011 (Noe-Pino 2016). Trapping results clearly confirmed the absence of fruit fly pests in the areas of interest. These results, and the fact that melon is recognized as a conditional host of the target fruit fly species, were the critical technical factors used in the bilateral negotiations between the national phytosanitary authorities of Honduras and Taiwan that resulted in an agreement to export melons using a pest risk mitigation scheme based on pest free production sites (Fig. 10).

A major advantage of this pest risk mitigation scheme is that no internal quarantine checkpoints are required, and places and sites of production need to be fruit fly free only during the entire fruit production and harvest period (FAO/IAEA 2017a).



Figure 10. Melons from fruit fly free places of production in Honduras (photo from SENASA Honduras, reproduced with permission).

### 7.6. Establishment of Fruit Fly ALPP and PFA in Central America

For the past decades, countries in Central America have been affected by low international prices of the traditional export crops coffee, banana and sugarcane. The governments of these countries and Panama have therefore been seeking new alternatives for international trade through production and export of non-traditional fruit and vegetables. To assists them in this task, from 2001 to 2006, IAEA and FAO provided support through a regional technical cooperation project to strengthen the countries' phytosanitary framework to allow them to establish fruit fly ALPP and fruit fly PFA using an AW-IPM approach that included, in some cases, the SIT (Reyes et al. 2007).

To achieve the goals, an approach was proposed to overcome existing constraints by integrating three main elements: 1) the development of a multi-institutional strategic alliance, 2) the use of pilot areas as a territorial strategy for suppression and eradication of fruit flies, and 3) a focus on promoting the export of fruits and vegetables.

The project outcome included: 1) the establishment of a number of fruit fly ALPP and PFAs in each of the participating countries, 2) investment by the fruit and vegetable industries in Costa Rica, El Salvador, Guatemala, and Nicaragua and of around USD 150 million in support of establishing and maintaining areas of fruit fly

low prevalence through a systems approach for exports of tomatoes and bell peppers to the USA, and 3) exports of papaya from Mediterranean fruit fly-free areas in the Department of Petén, in northern Guatemala, without the need for quarantine treatments (Fig. 11).

This project demonstrates that exports of non-traditional fruits and vegetables are a viable economic alternative to the traditional crops in the region by establishing fruit fly PFAs and ALPP integrated with a systems approach and creating more rural jobs than traditional crops (Reyes et al. 2007). Attempts to establish more of these areas throughout the region would be successful if:

1. The Ministries of Agriculture are the driving force of any such area-wide initiatives

2. The horticultural industry is convinced of the potential benefits that these areas can bring and is an active partner in the activities, and

3. Alliances are established between technical and financing organizations present in the region and they commit to working together sharing a common vision.

Strategic approaches for fruit fly control, which focus on specifically selected horticultural production areas, are in some instances easier to implement and more realistic than approaches which aim to initiate extensive and costly suppression and eradication programmes.



Figure 11. Papayas from Mediterranean fruit fly-free area in Petén, Guatemala (photo from Moscamed Programme; reproduced with permission).

### 7.7. Mediterranean Fruit Fly Eradication from the Dominican Republic

The presence of the Mediterranean fruit fly in the Dominican Republic was officially reported in March 2015. The pest had already spread to 2053 km<sup>2</sup> in the eastern part of the country, constituting a major outbreak in the Caribbean that up to then had been free of the Mediterranean fruit fly. An immediate ban on most exports of fruit and vegetables was imposed by trading partners, causing a loss of over USD 40 million for the remaining nine months of 2015.

Given the emergency situation, the Ministry of Agriculture established the Moscamed-DR Programme, with adequate financial resources and an effective organizational structure for its coordination and operations. The Guatemala-Mexico-USA Moscamed Programme and international organizations, including the FAO, IAEA, IICA (Inter-American Institute for Cooperation on Agriculture) and OIRSA, joined efforts with the Ministry of Agriculture to address the Mediterranean fruit fly outbreak. A technical advisory committee of experts provided oversight throughout the eradication campaign. An AW-IPM approach, including the application of the SIT as a final component, was used to eradicate the pest.

Official eradication was announced in July 2017 after six Mediterranean fruit fly generations of zero catches (Zavala-López et al., this volume). The Dominican Republic is now on the list of countries that have successfully eradicated the Mediterranean fruit fly, thereby avoiding the establishment of a major pest in the Caribbean, and substantially strengthening its fruit fly surveillance system and emergency response capacity (Fig. 12).



Figure 12. Plaque presented in July 2017 by the IAEA to the Minister of Agriculture of the Dominican Republic as a recognition for his leadership in eradicating the Mediterranean fruit fly (photo from W. Enkerlin).

As a spin-off of the successful eradication of the Mediterranean fruit fly, the Ministry of Agriculture of the Dominican Republic established a national fruit fly programme to manage native fruit flies, and to maintain the surveillance and response capacities for invasive fruit flies and other pests.

### 7.8. Carambola Fruit Fly Containment Programme in Guyana, Surinam, French Guiana and Brazil

The carambola fruit fly *B. carambolae* is native from Southeast Asia. It is known to infest a wide range of fruits and vegetables including carambola, guava, mango and others. The pest was first detected in the Americas in Suriname in 1975. Given its invasiveness and the risk it represented to the horticultural industry in Suriname and neighbouring countries, including Brazil, an eradication programme based on the male annihilation technique (MAT) was launched (Malavasi et al. 2000). The programme was funded by the International Fund for Agricultural Development (IFAD), France, the Netherlands, and the USA, and officially started in 1998 (Midgarden et al. 2016). In addition, during 1994-95, FAO provided capacity building assistance to the Brazilian Ministry of Agriculture (MAPA) to strengthen exclusion, detection and emergency response to new pest introductions. This enabled MAPA to train personnel, install detection traps within the state of Amapa, and take emergency response measures whenever detections of CFF occurred.

Considering the transboundary nature of this pest, programme operations were implemented on a regional level, including activities in Suriname, French Guiana, Guyana, and states in north-eastern Brazil. As a result, by 2001, the distribution of *B. carambolae* was reduced to limited areas of Suriname and French Guiana (Midgarden et al. 2016). Containing the spread of the pest resulted in 1) important economic benefits in reduced direct damage to fruits and vegetables, 2) social benefits by protecting important jobs associated to the horticultural industry, and 3) environmental benefits by preventing the massive use of insecticides that would be needed to control the pest.

Despite these results, in 2002 some of the donors significantly reduced their funding and the programme had to close in 2003. As a result, the pest reinvaded areas that had already been freed. Since then, only Brazil has continued implementing and financing the programme to contain its advance into Brazil. Nevertheless, in the following years *B. carambolae* expanded its distribution with detections as far southeast as Curralinho in the state of Pará in Brazil (Fig. 13). The presence of the pest in Guyana means that continuous incursions into both northern Brazil and Venezuela are likely, if not inevitable (Godoy et al. 2020).

Closing this programme before its completion has resulted in increasing costs to South American agriculture, and increased risk to Central and North America and the Caribbean. A specimen of *B. carambolae* was identified in Puerto Rico in 2015, and two putative *B. carambolae* specimens were trapped in Orlando, Florida in 2008 (Midgarden et al. 2016).



Figure 13. Presence of carambola fruit fly in South America up to 2016 (red dots present, blue dots detections eradicated) (reproduced from Midgarden et al. 2016).

A recent economic assessment shows a BCR of up to 37 to 1 for eradicating *B. carambolae* from the currently infested areas and from preventing further spread and invasion of the free areas in Brazil (IDB 2018). Based on this, in 2018, the Government of Brazil jointly with the Interamerican Development Bank (IDB) commissioned the preparation of a regional project for the control and eradication of the carambola fruit fly (IDB 2018). A coordinated programme amongst infested countries could still mitigate the risk of the spread of *B. carambolae* in the region (Midgarden et al. 2016).

Area-wide fruit fly control programmes in Latin America that apply an AW-IPM approach are listed in Table 1.

### 8. FUTURE PERSPECTIVES

• Given its devastating effects in the region, the fruit fly problem should receive high priority on the agendas of the Ministerial Agricultural Organizations, namely the Comité Internacional Regional de Sanidad Agropecuaria (CIRSA), the Consejo Agropecuario del Sur (CAS), and the Caribbean Agricultural Health and Food Safety Agency (CAHFSA).

| Strategic<br>Objective           | Area-wide Programme  | References  |
|----------------------------------|--|---|
| Prevention                       | Chile's National Fruit Fly Programme, 1980 – present   | Olalquiaga and Lobos 1993;<br>Rodríguez et al. 2016   |
|                                  | Mexican Fruit Fly Preventive Release Programme<br>(Rio Grande Valley, Texas, Mexico – USA border),<br>1980s – present                            | Holler et al. 1984  |
|                                  | Mexican Fruit Fly Preventive Release Programme<br>(Tijuana, Baja California, Mexico – USA border),<br>1960s – present                            | Lopez 1970; Dowell et al. 2000  |
|                                  | Bi-national Chile-Peru Programme for Mediterranean<br>Fruit Fly Eradication, 1996 – present  | Wedekind 2007; Rodríguez<br>et al. 2016   |
|                                  | Carambola Fruit Fly Containment Programme in<br>Surinam, French Guiana, Guyana, and Brazil, 1998 –<br>present                                    | Malavasi et al. 2000;<br>Midgarden et al. 2016; IDB<br>2018   |
|                                  | <i>A. grandis</i> prevention programme in north-eastern<br>Brazil to protect PFA, 1990 – present   | Razera Papa 2019  |
| Eradication                      | Guatemala-Mexico-USA Moscamed Programme for<br>the Containment and Eradication of the<br>Mediterranean Fruit Fly, 1975 – present                 | Hendrichs et al. 1983;<br>Enkerlin et al. 2017  |
|                                  | Mediterranean Fruit Fly Eradication Programme<br>"PROCEM" (Patagonia – Mendoza – San Juan,<br>Argentina), 1992 – present                         | De Longo et al. 2000;<br>Guillen and Sanchez 2007;<br>Wedekind 2007; Borges et<br>al. 2016; Quiroga et al. 2016 |
|                                  | Mediterranean Fruit Fly-Free Places and Sites of<br>Production in Honduras, 2017 – present   | Noe Pino 2016   |
|                                  | Mediterranean Fruit Fly Eradication Programme<br>(Altagracia, Dominican Republic), 2015 – 2017   | Zavala et al., this volume  |
| Suppression                      | Mediterranean Fruit Fly Control Programme (Peru),<br>1970s – present   | Guillen and Quintanilla<br>2008; Rivera-Tejada 2011;<br>Manrique and Rivera 2016                                |
|                                  | Fruit fly control programme in Ecuador in localized production sites, 2018 – present   | Vilatuña 2018   |
|                                  | Plan Nacional Moscas de la Fruta (PNMF) in Colombia, 2008 – present  | Arevalo 2016  |
|                                  | Programa Nacional de Control y Erradicación de<br>Mosca de los Frutos in north-eastern Argentina –<br>PROCEM NEA, 2015 – present                 | Morilla et al. 2016   |
|                                  | Moscasul programme pilot project to suppress the<br>South American fruit fly in southern Brazil, 2014 –<br>present                               | Kovaleski and Mastrangelo,<br>this volume   |
|                                  | Mediterranean fruit fly suppression on table grape<br>export areas along Rio San Francisco irrigation zone<br>(Bahia/Pernambuco), 2015 – present | Baronio et al. 2018   |
| Suppression<br>or<br>Eradication | The Mexican and West Indian Fruit Flies Suppression<br>and Eradication Programme – The National Fruit Fly<br>Programme in Mexico, 1991 – present | Reyes et al. 2000; Gutierrez<br>et al. 2013; Liedo et al., this<br>volume                                       |
|                                  | Establishment of Fruit Fly ALPP and PFA in Central<br>America, 2007 – present  | Reyes et al. 2007   |
|                                  |  |   |

Table 1. Fruit fly AW-IPM programmes in the LAC region

- These regional government organizations and their institutions should show strong political will by mobilizing and committing national resources and establishing regional phytosanitary policies that enable the enforcement of phytosanitary strategies such as the establishment of fruit fly PFAs and ALPP.
- The policies and actions should be enforced through instruments such as "Regional Fruit Fly Strategic Plans" against endemic fruit fly species, as well as against invasive species of quarantine significance. This would provide a systematic framework and guidance on the necessary actions required to achieve the objectives in controlling fruit fly pests in the Latin America and Caribbean region.
- Given the transboundary nature of the fruit fly problem in the Latin America and Caribbean region, RPPOs should have a more proactive role in preventing the introduction and establishment of invasive fruit fly pests and providing guidance on the phytosanitary approaches available for effective control of endemic fruit flies. They should promote the implementation of the ISPMs and propose/coordinate specific actions together with the NPPOs and main beneficiaries and stakeholders, including the growers and exporters.
- To address climate change and globalization that induce new pest problems, current legislation and policies need to be amended. Among these is the need to support the development of innovative control approaches to mitigate and manage these biological incursions, and to contain the geographic expansion of non-indigenous pest populations.
- The surveillance systems and emergency response capacities need to be strengthened on a regional basis to detect early introductions of non-native species still not present and that are of quarantine significance.
- The strategic approach against endemic fruit flies of economic significance should be, in most cases, the establishment of carefully selected areas free of pests or at low pest prevalence levels from which horticultural products can be produced and sold, rather than major eradication or suppression programmes that extend over very large areas, sometimes entire countries.
- Countries from the Latin America and Caribbean region should consider the possibility of promoting increased production and consumption of horticultural products in order to reduce the incidence of non-communicable diseases, plan for expected population growth, and take advantage of the trends in consumer preference to generate commercial opportunities to strengthen their economic performance.
- Countries interested in applying area-wide the SIT should take advantage of the existing sterile fly production capacity in the region (over 2.5 billion sterile flies per week) rather than each country aiming at building their own rearing and sterilisation facility. The investment should be focused on building sterile fly emergence and release facilities (FAO/IAEA 2017b), to hold and feed adult flies emerged from sterile pupae purchased from reliable external sources. The capital investment of such an approach would be much lower, as well as the risks of successfully integrating the SIT.

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# AREA-WIDE MANAGEMENT OF FRUIT FLIES IN A TROPICAL MANGO GROWING AREA INTEGRATING THE STERILE INSECT TECHNIQUE AND BIOLOGICAL CONTROL: FROM A RESEARCH TO AN OPERATIONAL PROGRAMME

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#### SUMMARY

The Sterile Insect Technique (SIT) has been successfully used for the control of fruit flies in a number of places in the world. One requirement for its successful application is that wild populations should be at low densities to achieve effective sterile to wild fly overflooding ratios. This has been an important reason that has limited its integration in fruit fly management in tropical fruit growing areas, where climate conditions and the availability of hosts all year-round results in high population densities. Here we report the results of a project where SIT integration into fruit fly management was evaluated under the tropical conditions of the mango growing area in the Soconusco region of Chiapas, Mexico. The basis for the area-wide integrated pest management (AW-IPM) approach was the knowledge of the population dynamics of the pest fruit flies in the region and of the fruit phenology. The main commercial mango growing areas are in the lowlands, where fruit fly populations are very low outside of the mango production season. Population densities are higher in the midlands and highlands, where alternate hosts are common in backyards and as part of the natural vegetation. We call these refuge areas, and the AW-IPM approach aimed at establishing a biological barrier with releases of parasitoids and sterile male fruit flies to suppress the fruit fly populations and prevent or minimize the dispersal of wild flies from the refuge areas to the mango orchards. In 2014, after two years of releases, fruit fly population densities were suppressed more than 70% in the release area and 65% in the entire area, including the lowlands with the mango orchards. With the support of fruit growers, state and federal governments, this project was continued and established as an operational AW-IPM programme. In 2016, after 4 years of programme implementation, the detection of wild flies was significantly reduced, and the number of batches of fruit that were rejected at the packing houses due to the detection of infested fruits was the lowest in the past 12 years, since the recording of these data was initiated. These indicators declined even further in 2017. The results obtained demonstrate that AW-IPM integrating the SIT can be applied successfully against fruit flies under tropical conditions with naturally high pest densities, providing there is adequate knowledge on the population dynamics of the fruit fly species present in the region.

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J. Hendrichs, R. Pereira and M. J. B. Vreysen (eds.), Area-Wide Integrated Pest Management: Development and Field Application, pp. 197–214. CRC Press, Boca Raton, Florida, USA. © 2021 IAEA Key Words: Integrated pest management, autocidal control, SIT, augmentative biological control, Anastrepha fruit flies, Tephritidae, Anastrepha ludens, Anastrepha obliqua, Diachasmimorpha longicaudata, Soconusco, Chiapas, Mexico

#### 1. INTRODUCTION

In Mexico, mango represents one of the most important fruit production and export value chains, with more than 180 000 ha of cultivation, giving an annual production of approximately 1.8 million tons. Mexico ranks 6<sup>th</sup> by area and 4<sup>th</sup> by production in the world, and on the international market Mexico and India are among the most important exporting countries by volume (SIAP 2015).

Among the factors that limit or affect mango production and marketing are insect pests, and within these, fruit flies are among the most devastating. In view of their importance, these are considered of public interest and for this reason the National Campaign against Fruit Flies was established in 1992. The Campaign has succeeded in achieving fruit fly pest free areas (FF-PFA, FAO 2016) in 52.8% of the national territory, and fruit fly areas of low pest prevalence (FF-ALPP, FAO 2008) in another 10.4%, while the remaining 36.8% is considered a zone under phytosanitary management (Liedo 2016; Ramírez y Ramírez et al. 2019). Due to their agro-ecological requirements, mango producing areas are located mostly in the subtropical and tropical zones of the country, most of which are in the area under phytosanitary management. Under these favourable ecological conditions that promote fruit fly abundance, the development of technologies and the design of pest management strategies are required to allow the production of fruits free of fruit fly damage.

There are modern and appropriate and more sustainable technologies to deal with these pests, such as the Augmentative Biological Control (ABC) and the Sterile Insect Technique (SIT), among others (Montoya and Toledo 2010; Enkerlin et al. 2021). These technologies, integrated with other control methods have been applied successfully in Mexico to prevent for over 35 years the invasion of the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), into southern Mexico along the border with Guatemala (Enkerlin et al. 2015) and for the implementation of FF-PFA for native *Anastrepha* fruit flies in the north of the country (Reyes et al. 2000; Liedo 2016). However, under the tropical conditions in which commercial mango is extensively grown, native *Anastrepha* fruit flies have high rates of population growth and therefore the effective application of these technologies is much more challenging (Montoya et al. 2000).

The characteristics of the SIT and the ABC, as well as the high mobility of fruit flies, make it necessary to adopt an area-wide integrated pest management (AW-IPM) approach, which considers the management of the total population of the pest and its spatial distribution (Hendrichs et al. 2007; Montoya et al. 2007).

In the mango producing region of Soconusco, Chiapas in southern Mexico, there is ample knowledge about the population dynamics of *Anastrepha* fruit flies (Aluja et al. 1996; Celedonio-Hurtado et al. 1995; Montoya et al. 2000). In the case of *Anastrepha ludens* (Loew) and *Anastrepha obliqua* (Macquart), which are the species that infest mango, it is known that their populations are high in the midlands and in the highlands (120-600 m elevation) with little temporal fluctuations.

In the commercial mango production areas of the lowlands (0-120 m elevation), the *Anastrepha* populations are low most of the year and only increase during the mango fruiting season, when growers have to apply repeated ground bait sprays to minimize fruit fly infestation.

Based on the available background information, and with the objective to reduce the bait sprays and fruit fly infestation in mangoes, a project was submitted to validate the use of the ABC and the SIT as elements of an AW-IPM approach, for the management of these native fruit flies in mango. We proposed to carry out this project in the Soconusco region in Chiapas State, considering the available knowledge on fruit fly populations, their hosts and their seasonality, and taking advantage of the infrastructure of the National Fruit Fly Campaign (Mexican Plant Protection Organization SENASICA, and IICA) in the region with respect to mass-rearing and release of sterile fruit flies and parasitoids. This 4-year project was funded by the National Council of Science and Technology (CONACYT) and the Ministry of Agriculture (SAGARPA) sectorial fund. In view of the increasing support and interest of the mango growers, the project was converted into an operational programme during the fourth and last year of the project.

#### 2. MATERIALS AND METHODS

#### 2.1. Strategy

It was assumed that the *Anastrepha* populations are maintained by year-round host availability in the refuge sites of the midlands and highlands outside the mango season. Thus, the designed strategy consisted of implementing a "biological barrier" based on the release of sterile flies and parasitoids in the intermediate zone between the high and the lowlands (ca. 100-200 m elevation), seeking to suppress the populations there and avoiding or minimizing their movement to the lowlands, where most of the commercial mango orchards are located. To facilitate achieving favourable sterile to fertile male ratios, releases of sterile insects were initiated at the end of November or early December 2012 and continued to 2015, i.e. the period when historically the lowest population levels have been observed (Aluja et al. 1996).

In the first year of the project (2011-2012), two trapping transects were established from the highlands to the lowlands, to monitor the populations of fruit flies along an altitudinal gradient (Fig. 1). Before initiating the releases in the second year, a third trapping route was established with the objective of monitoring the populations of sterile flies in the release polygon and adjacent areas.

The initial experimental design for the 4-year project (2011-2015) was: (a) to monitor populations during the first year without the application of the ABC and the SIT, (b) to apply the ABC and the SIT in the intermediate zone during the second and third years, and (c) depending on results, to transfer the technology to mango growers during the fourth year. Based on the results obtained during the second and third years, when the control methods were applied on an area-wide basis, it was decided to continue with the releases for another year. During this fourth year, the technology was transferred, and from week 10 of 2015, the fruit growers' union was in charge of

funding and coordinating the parasitoid and sterile fruit fly releases through the Local Board of Plant Health and the State Committee of Plant Health of Chiapas. During this fourth year, the research project was converted into an operational action programme, recognized by the National Fruit Fly Campaign.

Based on the monitoring of the first year and the availability of biological material, a 15 000 ha polygon (21.0 x 7.14 km) was established for the release of parasitoids and sterile fruit flies. This polygon was located between the cities of Mazatán and Tapachula, in the transition zone between the high and the lowlands. The location of the release polygon and the traps deployed are shown in Fig. 1.

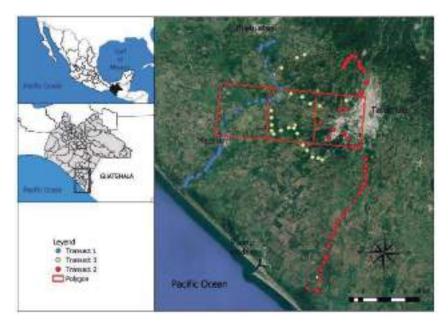


Figure 1. Location of the sterile fly release polygon in the Soconusco region of the state of Chiapas, covering 15 000 ha, and the three subquadrants for the release and assessment of parasitoids of 5000 ha each, as well as the location of the traps for population monitoring. The blue dots indicate the location of the traps deployed along transect 1, the red dots correspond to transect 2, and the yellow dots to transect 3.

For the evaluation of the ABC, the polygon was subdivided into 3 quadrants of 5000 ha each in which to make and assess the release of parasitoids. Initially, it was planned to release the parasitoids only in the sub-quadrant to the East -the one with higher trap captures- and leave the central one as the buffer area and the one on the West as a control, with the intention of alternating the quadrants in the third year and thus be able to make comparisons in time and space. However, based on the results obtained in the second year, it was decided to repeat the release area in the third year (East quadrant) in order to evaluate the effect of the modifications in the release method (see below) and also to contribute to the suppression of pest populations in the area of highest infestation.

## 2.2. Monitoring of Populations

For the monitoring of the *Anastrepha* populations, Multilure® traps baited with Biolure® (ammonium acetate + putrescine) were used. Propylene glycol was used for the retention and conservation of the trapped specimen. The traps were checked weekly and the trapped flies identified by species and sex. In the case of *A. ludens* and *A. obliqua*, the dye marking used for the sterile flies was used to discriminate between the released sterile and wild flies. Along transect 1, 35 traps were deployed from the town of Huehuetán in the highlands to Barra de San Simón in the lowlands. Fortyfour traps were deployed along transect 2 from Canton Pumpuapa to the northwest of the city of Tapachula, in the highlands, to the Ejido Conquista Campesina, in the lowlands. Transect 3 contained 30 traps of which 15 were deployed within the release polygon, 10 were located in the area adjacent to the north of the polygon and another 5 to the south of it (Fig. 1).

# 2.3. Biological Material

Every week, the project received 7.5-8.5 million sterile males of *A. ludens* Tapachula-7 genetic sexing strain, 5-10 million sterile males and females of *A. obliqua*, as well as 5 million of the parasitoid *Diachasmimorpha longicaudata* (Ashmead). With these quantities and based on the experience of the National Campaign, the target release densities were 533 male *A. ludens* / ha, 333 male *A. obliqua* / ha, and 1000 parasitoids / ha. The weekly amounts varied slightly depending on the weekly production of the Moscafrut facility in Metapa, as well as the needs of the National Campaign. For some time during the second year of releases, batches of the bisexual *A. ludens* strain (males and females) were also received, increasing the number of sterile insects released. The opposite was true for *A. obliqua* as the needs of the National Campaign resulted in smaller quantities being received sometimes (see Table 1 and Fig. 4), reducing the release densities.

|                        | 2012-13     | 2013-14     | 2014-15*    |
|------------------------|-------------|-------------|-------------|
| A. ludens Tap-7 males  |             |             |             |
| Total                  | 499 771 956 | 436 226 000 | 269 320 000 |
| Average / week         | 9 610 999   | 8 388 961   | 9 974 815   |
| A. ludens bisexual     |             |             |             |
| Total                  | 85 729 000  | 517 932 000 | 241 365 000 |
| Average / week         | 1 617 528   | 9 772 302   | 7 785 968   |
| A. ludens sum of males |             |             |             |
| Total                  | 542 636 456 | 695 192 000 | 390 002 500 |
| Average /week          | 10 419 763  | 13 275 112  | 13 867 799  |
| A. obliqua             |             |             |             |
| Total                  | 470 633 000 | 505 576 006 | 244 436 000 |
| Average / week         | 8 879 868   | 9 520 302   | 8 147 867   |

Table 1. Number of sterile flies received during three release seasons

\* In the 2014-15 season, data are included only until week 22 (June 6, 2015)

During the three release seasons, a total of 1205 million sterile *A. ludens* Tap-7 strain (only males), 845 million sterile *A. ludens* bisexual strain (males and females), 1219 million sterile *A. obliqua* (males and females) and 385 million *D. longicaudata* parasitoids were released. The number of sterile flies received in each of the three release seasons, as well as weekly averages, are shown in Table 1.

The quality of these sterile insects is shown in Table 2. From the series of quality control tests that were applied, we selected the percentage of flying flies after chilling as a parameter representing quality of this biological material. In the quality control manual this test is known as "absolute-post-chill flyers" (FAO/IAEA/USDA 2019).

| Strain             | 2012-13 | 2013-14 | 2014-15* |
|--------------------|---------|---------|----------|
| A. ludens Tap-7    | 81.05   | 86.14   | 85.42    |
| A. ludens bisexual | 87.47   | 89.05   | 85.23    |
| A. obliqua         | 88.46   | 89.39   | 88.26    |

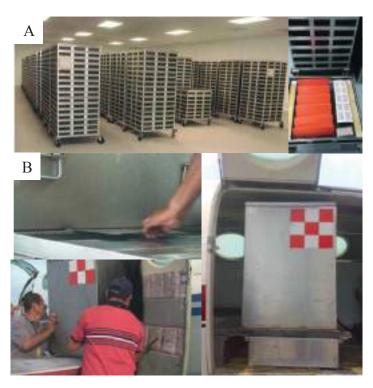
 Table 2. Quality of the sterile flies received during three release seasons, as percentage of flying flies after chilling

\*For the 2014-15 season, data are included only until week 22 (June 6, 2015)

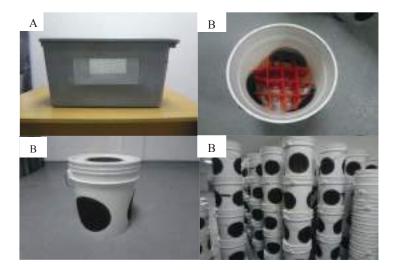
The mass-production facility in Metapa delivered the biological material to the Mediterranean Fruit Fly Emergence and Release Facility (CEMM by its initials in Spanish), located near the Tapachula airport. All biological material was received as irradiated pupae under hypoxic conditions. The CEMM staff placed the pupae in "Mexico" type emergence towers that were provided with water and food. The food was a mixture of sugar and enzymatic hydrolysed yeast in a 24:1 ratio. The towers were kept in the emergence rooms at  $23 \pm 1$  °C and  $65 \pm 5\%$  relative humidity. Adults emerged 2-3 days later and were released once they were sexually mature, i.e. 5 days later in the case of *A. ludens* and 4 days in the case of *A. obliqua*. On the release day, the towers were placed in specially designed refrigeration boxes and transported to the airport for chilled aerial release (Hernández et al. 2010). Images of the emergence towers and the release box are shown in Fig. 2.

The services of the Mubarqui<sup>®</sup> company were contracted to implement the aerial releases of the sterile flies. This company has adapted aircrafts, appropriate release machines, and support infrastructure to implement these releases. For each release flight, a report was generated indicating the time of the flight, the route followed, the number of sterile insects released and the corresponding density. Samples were taken from each batch of sterile insects to assess standardized quality parameters (FAO/IAEA/USDA 2019).

Parasitoids were released from the ground with the support of the staff and vehicles of the State Committee for Plant Health. During the first year of releases, the parasitized pupae were placed in PARC<sup>®</sup> boxes (Plastic Aerial Release Container) with a mixture of honey and paper as a food source (Fig. 3A).



*Figure 2. Towers type "México" used for the emergence of sterile adults (A) and chilled release box being loaded into the aircraft (B).* 



*Figure 3. PARC box (A) and "R2D2" devices (B) used for the emergence and feeding of parasitoids and their subsequent ground release in the field.* 

After eight days, the emerged parasitoid adults were released. With the aim of improving the performance of these parasitoids, in the following year the release was implemented using "R2D2" devices, which are 20 litres plastic containers with mesh windows on the wall and cover. To increase the surface area, a corrugated plastic honeycomb was placed inside the containers. Each device contained approximately 2000 parasitized pupae (Montoya et al. 2012). The "R2D2" devices are shown in Fig. 3B. These "R2D2" devices replaced the PARC boxes in the second and third release years.

#### 3. RESULTS

#### 3.1. Biological Material

Weekly variations in the number of sterile males released during the three release seasons are shown in Fig. 4.



Figure 4. Number of sterile males released per week in each of the three release seasons.
A) A. ludens males of the Tapachula-7 and bisexual strains. B) A. obliqua males.
Season 2012-13 is shown in blue, 2013-14 in orange, and 2014-15 in grey.

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For the third season (2014-15), data are shown only up to week 22 when, based on the number and locations of wild fly detections, it was decided to modify the release polygon (see Section 4). It should be noted that since week 10 of this last season, the association of fruit growers took over the funding and coordinating of field activities, fulfilling the goal of technology transfer, and thus transforming a research project into an operational programme.

The number of *A. ludens* sterile males released ranged between 10 and 20 million per week for most of the time for the three release seasons. Numbers for the 2013-14 and 2014-15 seasons were consistently higher. With these quantities, the average densities released were 696, 891 and 849 males per ha for the seasons 2012-13, 2013-14, and 2014-15, respectively. The highest density was 2084 males / ha in week 46 of 2013 and the lowest was 314 males / ha in week 1 of 2013. Considering the natural population dynamics of this species, in the first weeks of the calendar year it is key to have high sterile fly densities in the field, which was achieved in the 2014-15 season.

For *A. obliqua* the quantities of sterile flies were smaller and the variation greater. The average densities released were 302, 323 and 299 males / ha for the seasons 2012-13, 2013-14, and 2014-15, respectively. The highest density was 543 males / ha in week 13 of 2014 and the lowest was 148 males / ha in week 40 of the same year.

As of week 34 of 2014, there was a significant reduction in *A. obliqua* sterile fly availability and from week 49 to 52 of 2014 no biological material was received. This was attributed to production problems at the Moscafrut facility and due to the demand for this species by the National Campaign.

## 3.2. Sterile Fly Densities

The releases of sterile flies were monitored with Multilure traps deployed inside the area of the release polygon. Out of the total of 109 traps were deployed as part of the three trapping transects, 30 were located inside this polygon. Average fly per trap per day (FTD) values for trapped sterile males in these 30 traps for each season and species are shown in Table 3.

| Season   | A. ludens | A. obliqua |
|----------|-----------|------------|
| 2012-13  | 0.766     | 0.643      |
| 2013-14  | 1.363     | 0.531      |
| 2014-15* | 0.472     | 0.135      |

 Table 3. Average of sterile males captured per trap per day (FTD) in the release polygon in each of the three release seasons for A. ludens and A. obliqua

\*Only until week 22, since the release polygon was modified afterwards

Another parameter used to monitor sterile fly releases was the percentage of traps that trapped sterile flies, regardless of the amount captured. This parameter informs about the uniformity of the releases. Fig. 5 shows how this percentage varied throughout the year for the two species in the three seasons. The average percentage of traps that trapped sterile flies in each season ranged from 53 to 66% for *A. ludens* and 27 to 60% for *A. obliqua*. The lower value in *A. obliqua* corresponded to the 2014-15 season and was related to the suspension of releases from week 49 to 52 due to lack of biological material.

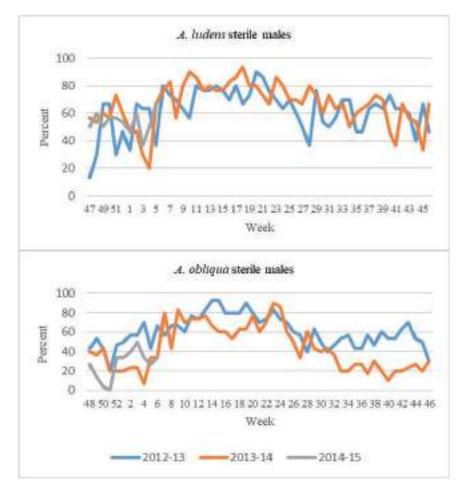


Figure 5. Percentage of traps with capture of sterile flies in each of the three release seasons for A. ludens and A. obliqua.

The sterile: fertile ratio refers to the number of sterile males compared to the number of fertile or wild males in the monitoring traps. This relationship is the basis of the Knipling model (Knipling 1955), which in turn is the foundation of SIT

application. The ratios achieved for each species and in each season are shown in Table 4. In the case of *A. ludens*, considering the total catches throughout the complete season, the sterile: fertile ratios were always higher than 200:1 and in the 2013-14 season the ratio was 519:1. In *A. obliqua* these ratios were much lower, even in those cases where the number of sterile males trapped was similar to *A. ludens* (i.e. season 2012-13).

| Season   | A. ludens |         | A. obliqua |         |         |     |
|----------|-----------|---------|------------|---------|---------|-----|
|          | Sterile   | Fertile | S:F        | Sterile | Fertile | S:F |
| 2012-13  | 7 936     | 31      | 256        | 6 956   | 618     | 11  |
| 2013-14  | 15 058    | 29      | 519        | 5 929   | 145     | 41  |
| 2014-15* | 2 131     | 10      | 213        | 575     | 25      | 25  |

Table 4. Number of sterile and fertile or wild males captured in traps and the corresponding sterile: fertile ratio for A. ludens and A. obliqua in the three seasons

\* Only includes the first 22 weeks, since the release polygon was modified afterwards

Empirical evidence indicates that to obtain good suppression, the sterile: fertile ratio must be greater than 30:1, but ideally greater than 100:1 (Flores et al. 2014, 2017). The achieved sterile: fertile ratios were satisfactory in the case of *A. ludens*, particularly in the 2013-14 season, when the average release density was 891 males / ha. While in the case of *A. obliqua*, the lower release densities (averages <350 males / ha) together with the larger wild populations resulted in sterile: fertile ratios much lower than the target of 100:1.

#### 3.3. Wild Population Densities

To make an estimate of wild population levels we used wild female catches. During the four monitoring seasons, considering all the traps, a total of 3792 *A. ludens* and 37 445 *A. obliqua* females were trapped. This represents a 9.8 times greater catch for *A. obliqua*. To assess the effect of the sterile fly releases, only the traps located in the release polygon were considered (Fig. 6).

The wild populations in the first 2012-13 season were higher in the release polygon than in the preceding year without them. In the subsequent 2013-14 and 2014-15 seasons, the density of sterile flies was increased, and this situation was reversed. The populations were significantly suppressed. The suppression effect ranged from 76 - 81% for *A. ludens*, while for *A. obliqua*, despite the low sterile: fertile ratio, suppression ranged from 89 to 91%. The suppression is confirmed by a comparison of the mean numbers of females per trap per two seasons between the release polygon and the traps located at the north of the polygon (Fig. 7), where no control measures were applied, and south of the polygon, where most commercial mango orchards are located and which was the protected area.

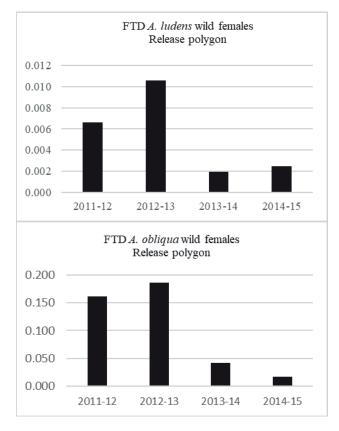


Figure 6. Capture of wild females, expressed in FTD (flies per trap per day), for A. ludens and A. obliqua during four seasons in the release polygon. The 2011-12 season without and the other three seasons with ABC and SIT releases.

Releases of *D. longicaudata* parasitoids resulted in a significant increase in the parasitisation rate. Table 5 shows these rates in the area of the polygon where parasitoids were released and in the control area where no releases were made. Parasitism by *D. longicaudata* in the area where releases were made was 15.12%; the other 1.38% was by native parasitoids. Montoya et al. (2017) provide more detailed information on the effects of the *D. longicaudata* releases.

 Table 5. Number of flies, parasitoids and parasitisation rate in the zone with augmentative releases of D. longicaudata and the control zone without releases

| Zone             | Flies | Parasitoids | Parasitism<br>(%) | Fruit samples with parasitoids (%) |
|------------------|-------|-------------|-------------------|------------------------------------|
| Without releases | 468   | 5           | 1.05              | 3.8                                |
| With releases    | 5 271 | 947         | 16.50             | 27.7                               |

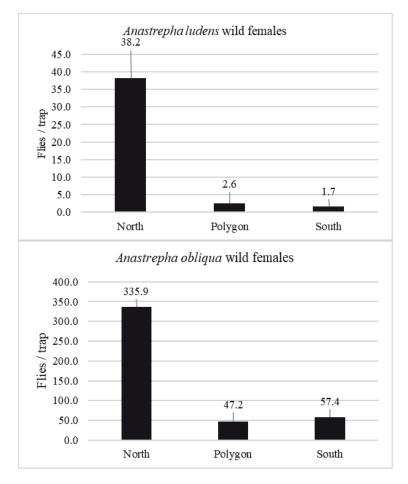


Figure 7. Mean (SE) capture of wild females expressed in flies per trap for A. ludens and A. obliqua in the 2012-13 and 2013-14 seasons within the release polygon and in the traps located north and south of the release polygon.

# 3.4. Infested Batches and the Species of Concern

The differences in abundance observed between the two species - *A. ludens* and *A. obliqua* - were consistent and in agreement with what was previously reported (Aluja et al. 1996). However, this difference did not match the detection of infested batches of mango in the packing houses: more than 94% of the infested lots were by *A. ludens* (information provided by the State Committee for Plant Health of Chiapas) (Table 6).

In 2014, Aluja et al. (2014) reported that mango cultivar Ataulfo was not infested by *A. obliqua* when fruits were still on the trees. Only the fruits known as "mango niño" (baby mango) were infested. These are small fruits of the same cultivar that do not grow as the normal fruits. The Ataulfo cultivar is the one that occupies the largest surface area in Chiapas and represents more than 90% of mango exports. This observation made us conclude that *A. ludens* was the important species to suppress to minimize or prevent fruit infestation. This new knowledge was fortunate as the programme could receive sufficient sterile male *A. ludens* to achieve densities of 800 to 1000 males per ha, reaching sterile: fertile ratios of more than 100:1.

| Season | A. obliqua | A. ludens | % A. ludens |
|--------|------------|-----------|-------------|
| 2012   | 21         | 390       | 94.9        |
| 2013   | 9          | 401       | 97.8        |
| 2014   | 6          | 236       | 97.5        |
| 2015   | 3          | 241       | 98.8        |

 Table 6. Number of infested batches at the packing houses per species and the percent of those infested by Anastrepha ludens

## 4. FROM A RESEARCH PROJECT TO AN ACTION PROGRAMME

Considering the four seasons of the research project, we were able to verify and validate the effectiveness of the SIT and the ABC for the suppression of populations of these two species of fruit flies. The strategy of establishing a "biological barrier" between the midlands and highlands with high fruit fly populations, and the lowlands, where the largest area planted with commercial mango is located, seemed to be appropriate to reduce fruit fly infestation in mango from 76% to 91%. Nevertheless, for an action programme, monitoring should be expanded so that the location of this "biological barrier" is dynamic and can be adapted to the situation of the pest. Also, the importance of achieving high sterile to fertile ratios before the start of the mango season should always be kept in mind.

At this stage of the research project, growers were very interested in continuing the releases of sterile insects and parasitoids. They were willing to contribute the needed funds, and to fulfil the requirements to be considered for participation in the National Campaign. It was decided to focus only on *A. ludens*, and the National Campaign offered to provide the programme with 15 million sterile males (Tapachula-7 genetic sexing strain) and 5 million *D. longicaudata* for the ABC.

A technical group to follow up the programme was established with participants from the mango growers' union, the State Plant Health Committee, the National Fruit Fly Campaign and ECOSUR (El Colegio de la Frontera Sur, public research centre). This technical group meets every two weeks or weekly, depending on the time of the year. The technical group agreed on the design of a new release polygon of 15 000 ha based on the pest situation and the availability of biological material. Releases of sterile flies and parasitoids have not been stopped since then. As an example of this follow-up, the situation for week 25 in 2016 is shown in Fig. 8, with the location of the modified release polygon (40 x 3.75 km), the aerial release lines, and the location of the monitoring traps indicating flies captured.

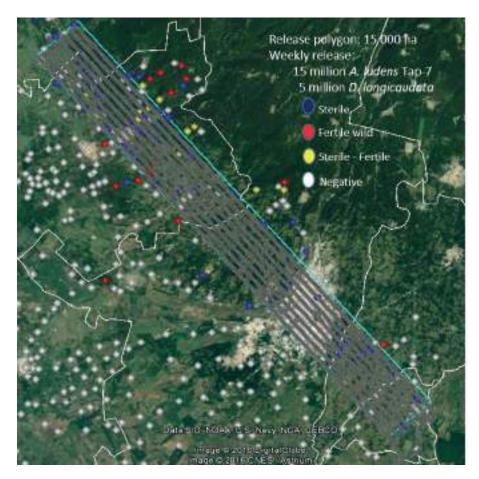


Figure 8. Modified release polygon of the action programme showing flight lines and the location of traps. Colour codes indicate type of trapped flies; sterile - fertile means that both types of flies were captured in the trap; these data correspond to week 25, 2016.

Although there was no immediate suppression effect of sterile flies and parasitoids releases, the continued releases from 2012 to 2017 resulted in a gradual suppression of wild *A. ludens* populations and as a result the number of infested mango batches detected at the packing houses has been greatly reduced. The number of infested batches per season, and the corresponding index of infested batches per ton of exported mangoes are shown in Fig. 9. These results are encouraging and demonstrate the long-term effects of area-wide integration of the SIT and the ABC. They also show that with good knowledge of the dynamics of fruit fly populations it is feasible to design AW-IPM programmes, integrating the SIT and the ABC in the management of fruit flies under tropical conditions where pest populations are normally high.

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The challenge now is to maintain and refine the releases of sterile flies and parasitoids and assess whether their synergistic effect will further decrease the pest populations, and in the long term minimize or avoid bait insecticide sprays in the region.

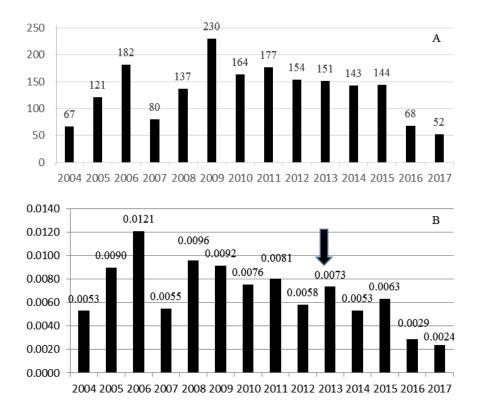


Figure 9. Number of infested batches detected at packing houses per season (A) and index of infested batches per ton of exported mangoes (B). The arrow shows when releases of sterile flies and parasitoids were initiated.

# 5. ACKNOWLEDGMENTS

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# MOSCASUL PROGRAMME: FIRST STEPS OF A PILOT PROJECT TO SUPPRESS THE SOUTH AMERICAN FRUIT FLY IN SOUTHERN BRAZIL

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#### SUMMARY

In Brazil, 99% of the apple growing areas are concentrated in the southern region, with an annual harvest of more than 1.2 million tons of fruits and a supply chain amounting to USD 1900 million. Despite the occurrence of several species of tephritid fruit flies in the region, the South American fruit fly, Anastrepha fraterculus (Wiedemann) (Tephritidae) represents 98.5% of the flies captured in commercial apple orchards. The gross value of yield losses and the cost of associated chemical control of this pest were estimated at close to USD 8 million per year. Moreover, the infestation rate by A. fraterculus has increased during the past four years, as the most commonly used insecticides have been banned. Brazilian researchers, along with state institutes and the Brazilian Association of Apple Producers (ABPM) have been promoting environment-friendly alternatives to insecticide application, such as sterile insects and parasitoids, to suppress the pest, and created the Moscasul Biological Control and Integrated Fruit Fly Management Center. After receiving the support of the Ministry of Agriculture at the end of 2014, almost USD 600 000 have been invested in constructing a rearing facility for A. fraterculus at the Agricultural Experiment Station of EMBRAPA (Brazilian Agricultural Research Corporation), Vacaria, Rio Grande do Sul, Brazil. The first containerized laboratory modules were installed in May 2016. During the pre-operational phase of the project, pilot trials were planned in apple orchards at Vacaria. As the Center for Nuclear Energy in Agriculture (CENA) has a 250 m<sup>2</sup> facility to produce fruit flies, including radiation sources, the sterile flies for the pilot trials will be initially provided by CENA. Both sterile flies and Diachasmimorpha longicaudata (Ashmead) parasitoids will be released in the surrounding non-commercial vegetation located within a 50-100 m buffer zone in the periphery of the target orchards, as these areas are the native breeding sites and repositories of the wild flies. Based on the wild population densities of the in the target orchards (114 ha) and the surrounding forested areas (111 ha), about 150 000 sterile flies per week will be required for the first pilot phase. After the fine-tuning of all rearing and sterilisation procedures in the beginning of 2017, CENA will ship more than 200 000 irradiated pupae weekly by air to Vacaria for 6 months, starting in September when the level of the wild fly population is lower. The results may influence the direction of future control tests and benefit the area-wide management of A. fraterculus involving hundreds of apple producers in the region and other temperate fruit growing farmers from southern Brazil.

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J. Hendrichs, R. Pereira and M. J. B. Vreysen (eds.), Area-Wide Integrated Pest Management: Development and Field Application, pp. 215–230. CRC Press, Boca Raton, Florida, USA. © 2021 IAEA Key Words: Anastrepha fraterculus, Diachasmimorpha longicaudata, parasitoids, SIT, sterile insects, areawide, apple orchards, wild hosts, Rio Grande do Sul, Santa Catarina, Paraná

#### 1. INTRODUCTION

Refuting USA technical reports from the 1960s claiming that producing apples on a commercial scale would be impossible in Brazil, the apple industry has become one of the most recent success stories of fruit producers in the country (Klanovicz 2010). The joint efforts of industry, research institutions and extension services has resulted in an increase in apple yield from 2-4 to 28-30 tons per ha (Klanovicz 2010). In 1986, the First Santa Catarina Apple Festival was celebrated at Fraiburgo, where the Brazilian Association of Apple Producers (*Associação Brasileira dos Produtores de Maça* – ABPM) launched the campaign "The Brazilian Apple: the sin that worked out right". By 1989, commercial apple production had become firmly established in Brazil (Brazilian Apple Yearbook 2017).

Recently, in only 30 years, Brazil almost reached self-sufficiency in apple production, with a total planted area of 34 399 ha, a yield of 38.9 tons/ha and a total of 1 247 088 tons produced in 2016/2017; the supply chain of the sector amounts to USD 1900 million per year (Brazilian Apple Yearbook 2017). More than 4300 growers are distributed throughout the three southern states of Brazil (Rio Grande do Sul, Santa Catarina and Paraná), where 99% of Brazil's apple production is located (GAIN Report 2016).

The municipalities of São Joaquim (Santa Catarina state) and Vacaria (Rio Grande do Sul state) have taken turns leading the production volume (ca. 400 000 tons in each one, depending of the year) (Brazilian Apple Yearbook 2017). These localities present the most favourable weather conditions, with colder winter temperatures (i.e. more than 900 hours below 7.2°C), mean annual temperature of 15.2°C, altitudes higher than 800 m, as well as mild summer and autumn days, but with cold nights (10-15°C), ideal for the physiological processes of temperate fruit trees (Petri et al. 2011).

Despite all the technological advances that allowed the successful establishment of the apple production in the highlands of the states of southern Brazil, the orchards are constantly under threat of important pests like the European red spider mite (*Panonychus ulmi* Koch), woolly apple aphids (*Eriosoma lanigerum* Hausmann), Brazilian apple leafroller (*Bonagota salubricola* Meyrick), oriental fruit moth (*Grapholita molesta* Busk) and the South American fruit fly (*Anastrepha fraterculus* Wiedemann).

In October 1991, the codling moth, *Cydia pomonella* L., was detected in urban areas of four municipalities, but never invaded the commercial farms. Brazil was declared free of this pest in May 2014 after a successful eradication programme that was mainly based on host-tree removal in household backyards of urban and suburban areas (Kovaleski and Mumford 2007).

In 2015, the South American fruit fly became the target of another integrated pest management (IPM) programme.

# 2. IMPORTANCE OF THE SOUTH AMERICAN FRUIT FLY IN SOUTHERN BRAZIL

The South American fruit fly is a complex of cryptic species that comprises at least eight different morphotypes under the same species designation (Hernández-Ortiz et al. 2012). This complex is distributed from Texas to Argentina and can attack more than 80 species of host fruit trees (Steck 1998; Norrbom 2004). The flies develop within the range of 15-30°C, and the main biological characteristics at 25°C are: pre-oviposition period of 7-14 days; oviposition until 46-62 days (fecundity can reach 40 eggs/female/day, with an average of 25.2 eggs/female/day, and one female can lay up to 979 eggs during her lifetime); embryogenesis lasts 1-3 days; larval development of 10-14 days and a pupal period of 11-21 days (Machado et al. 1995; Salles 1993, 2000; Vera et al. 2007; Cladera et al. 2014).

The damage caused by *A. fraterculus* on apples occurs soon after the beginning of fruit development (ca. 2 cm diameter) and is caused by the piercing of the fruit skin by the female ovipositor, causing lesions in the fruit that result in fruit deformations (Magnabosco 1994; Sugayama et al. 1997). In case fully developed fruits get infested, the external symptoms usually do not appear, but the pulp may be destroyed by larvae (Kovaleski et al. 2000).

The pomiculture in southern Brazil has suffered heavy losses due to the attacks of *A. fraterculus* over several years, and the annual yield losses and associated annual costs of chemical control of this pest alone have amounted to more than USD 7.9 million (Salles 1998; Nora and Hickel 2002; Kovaleski and Ribeiro 2003).

Despite the presence of the Mediterranean fruit fly *Ceratitis capitata* (Wiedemann), *Rhagoletis* spp., and more than 10 other fruit fly species of the *Anastrepha* genus in southern Brazil (Kovaleski et al. 2000), *A. fraterculus* represents 98.5% of the flies captured in commercial apple orchards (Canal Daza et al. 1994; Nora et al. 2000; Santos et al. 2017).

Pre-harvest management of fruit flies has been implemented almost exclusively with chemical methods, and the control effort has been guided by population monitoring with McPhail traps with a solution of 25% grape juice or hydrolysed proteins (McPhail 1937; FAO/IAEA 2018) (Fig. 1).



Figure 1. Weekly field monitoring of Anastrepha fraterculus populations in commercial apple orchards with McPhail traps baited with fruit juice or hydrolysed proteins.

The number of traps deployed depends on the size of the orchards, i.e. 4 traps/ha for areas up to 2 ha, and 2 traps/ha for areas between 2 and 5 ha (Kovaleski et al. 2000). Growers mostly have used a solution of 25% grape juice as the common attractant with the traps, but there are concerns with respect to their efficiency for fruit fly monitoring in apple orchards (Bortoli et al. 2016). Recent studies have demonstrated that protein-based lures of plant or animal origin (e.g. BioAnastrepha<sup>TM</sup> and Ceratrap<sup>TM</sup>, respectively) are better attractants for *A. fraterculus* (Scoz et al. 2006; Rosa et al. 2017).

As the *A. fraterculus* populations usually invade apple orchards from the surrounding areas, where they develop on non-commercial preferred hosts (Kovaleski et al. 1996; Sugayama et al. 1997; Santos et al. 2017), growers have traditionally applied weekly toxic bait sprays at the periphery of the apple orchards. The use of these baits (composed of water + insecticide + attractant such as hydrolysed proteins or 5-7% sugarcane molasses) is intensified in the first months of fruit growth, when the introduction of ovipositor may lead to external fruit deformations (Kovaleski et al. 2000).

When the number of adult flies captured inside the orchard exceeds a threshold level of 0.5 flies per trap per day (FTD) (a threshold level adopted by Brazilian growers since the 1980s), insecticides with systemic action and long residual effects are applied as cover-sprays, usually requiring 8 to 10 applications per season at a cost of about USD 240/ha/year.

Due to their low cost and residual properties (Harter et al. 2015), organophosphate insecticides have been heavily used by Brazilian growers for more than 20 years (Puzzi and Orlando 1957; Salles and Kovaleski 1990; Kovaleski and Ribeiro 2003). Many organophosphate insecticides, however, are being gradually banned or the maximum tolerable residue levels have been drastically reduced for exported fruits and derivatives like juice, concentrates and purees (Rawn et al. 2006; Eddleston et al. 2012). For example, fenthion was the most commercialized insecticide to control *A. fraterculus* in Brazilian apple orchards until 1997, when the growers stopped using it due to the risk of rejection of the fruits on international markets (Kovaleski et al. 2000). In addition, there is the risk of insecticide resistance developing in *A. fraterculus* as reported for *C. capitata* (Couso-Ferrer et al. 2011).

In order to meet the requirements of international markets for low residues on fruit, growers are increasingly being pushed to avoid insecticide applications against *A. fraterculus* during longer periods before harvest. Consequently, many apple orchards are being left unprotected from this pest during periods when the fruits are most susceptible.

In the absence of chemical control, the yields of apple orchards can be reduced with up to 30% due to *A. fraterculus* damage (Kovaleski et al. 2000). The shrinking choice of insecticides available to control *A. fraterculus* in Brazil, coupled with public demand for sustainable alternatives, have created a significant opportunity for promoting the area-wide augmentative biological control using sterile insects and parasitoids.

# 3. POPULATION DYNAMICS OF THE SOUTH AMERICAN FRUIT FLY IN APPLE ORCHARDS OF SOUTHERN BRAZIL

Tephritid pest population dynamics are largely affected by climatic features and availability of hosts (Aluja et al. 2012). This was confirmed by the general dynamics of the *A. fraterculus* populations of the highlands of the midwestern plateau of Santa Catarina and the mountainous region of Rio Grande do Sul, where dynamics of this fly have remained relatively constant for the last 20 years (Salles 1995; Garcia et al. 2003; Calore et al. 2013; Santos et al. 2015, 2017).

Trap captures over a period of 10 years showed that *A. fraterculus* population densities were higher between November and February (spring-summer), but between May and September (autumn-winter) they practically disappear from the commercial apple orchards when mean daily temperatures drop below 15°C and host availability is very low (Fig. 2) (Salles and Kovaleski 1990). This pattern of population fluctuation has remained unchanged in most apple orchards to date (Santos et al. 2017).

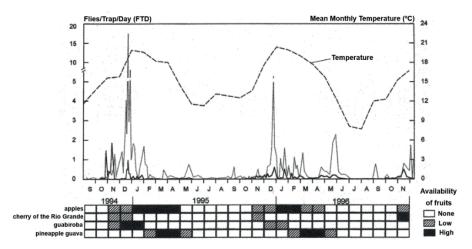


Figure 2. Mean monthly temperatures and population fluctuation of Anastrepha fraterculus (FTD) from commercial apple orchards (represented by a dark solid line) and from adjacent native forest (by a lighter broken line), together with the availability of apples and wild hosts at Vacaria, Rio Grande do Sul from September 1994 to November 1996 (from Kovaleski et al. 2000).

The presence of natural hosts from the family Myrtaceae (Malavasi et al. 1980) in the forested areas, bordering the apple orchards, provide an opportunity for *A*. *fraterculus* populations to be maintained throughout the year in the region. The natural hosts are mainly the cherry of the Rio Grande (*Eugenia involucrata* DC), guabiroba (*Campomanesia xanthocarpa* Berg) and pineapple guava (*Feijoa sellowiana* Berg) (Fig. 2), which bear fruits in November, December-January, and February-May, respectively (Kovaleski et al. 2000).

In the beginning of November, the cherries of the Rio Grande can be infested by A. fraterculus females who are residuals from the previous autumn season. When the adults that emerged from the infested cherries become sexually mature, they infest the guabirobas, but available mature apples can also be attacked. After January, pineapple guavas are preferably infested. As the temperatures start to drop in April, the larval development in these guavas can be prolonged until 80 days, and the pupal period can last up to 120 days (Kovaleski 1997; Kovaleski et al. 2000). The flies that emerge in August and beyond May survive until the appearance of new cherries, guabirobas and apples in November (adult overwintering). Thus, autumns with high availability of pineapple guavas and mild winters can be followed by high A. fraterculus populations in the spring. The late infestation of apples during March-April may also produce a certain amount of pupae, whose adults can emerge at the end of winter (or overwintering as immatures) (Kovaleski et al. 2000). Therefore, the 4-5 months (June - October), when mean temperatures are below 15°C and no host fruits are available, cause a natural decline in A. fraterculus populations each year, creating a perfect window of opportunities for the initiation of the release of parasitoids and sterile flies in the forested areas surrounding apple orchards.

The area-wide management of *A. fraterculus* in the commercial apple orchards in southern Brazil is facilitated by the absence of resident populations in the apple orchards, as the natural breeding sites of this pest are the native forests (Kovaleski et al. 1996, 1999; Santos et al. 2017). In the sierra region, where apple orchards and forested areas intermingle, traps located closer to the forested areas generally catch much more flies (e.g. values can even reach 20 FTD) than those deployed inside the orchards (Fig. 3).

Almost 80% of the damage to fruits occurs in the periphery of the first lines of the apple orchards, because of the prevalence of the foraging behaviour of *A. fraterculus* for oviposition sites and food resources rather than migratory movements (Sugayama 1995; Kovaleski et al. 1996, 1999).

Sugayama et al. (1997) described the diel pattern of *A. fraterculus* in Brazilian apple orchards. When an apple orchard is located closely to forests, most of the flies do not remain in the orchard during the night. The females mostly oviposit their eggs between 16 and 17 h and at nightfall they return to the forested areas. The fact that apple trees in Brazilian orchards do not form dense canopies that could serve as shelter can contribute to this behaviour. Furthermore, apples can be considered alternative hosts for *A. fraterculus* (Salles 1995) as most varieties behave as poor hosts. Immature apples are unsuitable hosts, with less than 1% of survival to pupal stage, and the reproductive rates of *A. fraterculus* in mature apples is usually low (e.g. under field conditions, infestation levels reach 600-800 pupae/kg of apple) (Sugayama 1995; Sugayama et al. 1998; Sugayama and Malavasi 2000).

A decade-long study (Kovaleski et al. 2000) that included fly monitoring, releaserecapture trials and host surveys has demonstrated that *A. fraterculus* populations are primarily present in the forested areas that contain native hosts surrounding the apple orchards (Fig. 3). Consequently, these areas should be the primary targets for suppression and the release of sterile flies and parasitoids starting in September. Maintaining sufficient overflooding ratios throughout the final winter months and spring-summer should theoretically suppress the wild populations.

# 4. THE STERILE INSECT TECHNIQUE AGAINST THE SOUTH AMERICAN FRUIT FLY

The Sterile Insect Technique (SIT) is an effective and environment-friendly control technology that relies on inundative releases of mass-reared insects, sterilized by ionizing radiation (Dyck et al. 2021). This technique has been applied as a component of many area-wide integrated pest management (AW-IPM) programmes against fruit flies, moths, screwworms and tsetse flies (Vreysen et al. 2007).

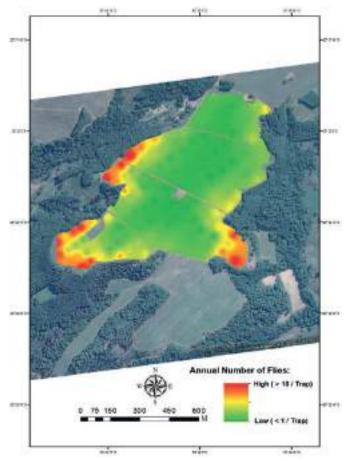


Figure 3. Annual trap catches of wild Anastrepha fraterculus in McPhail traps baited with grape juice in a commercial apple orchard largely surrounded by forest in Vacaria, Rio Grande do Sul, during the 2016-2017 harvest.

For example, almost 300 million sterile flies are produced per week at the Moscafrut facility for the control of several *Anastrepha* species of economic importance in southern Mexico (Orozco-Davila et al. 2017). In British Columbia, Canada, the wild populations of *C. pomonella* are being kept at minimum levels since

1997 through the release of sterile moths from the Okanagan-Kootenay Sterile Insect Release (OKSIR) Programme, with populations reduced by 94% and damage reduced to less than 0.2% of fruits in more than 90% of the orchards in the programme area (Judd and Thompson 2012; Simmons et al. 2021; Nelson et al., this volume).

Despite all the knowledge gathered on the biology and genetics of *A. fraterculus* (Cladera et al. 2014), AW-IPM programmes integrating the SIT against this pest are still not being implemented. The SIT has several technical components and major requirements that need to be met: (1) availability of accurate baseline data on the target wild population (e.g. population density in space and time, dispersal patterns etc.); (2) methods available to mass-rear the insect at reasonable cost; (3) irradiation procedures for proper sterilisation of large batches of the mass-produced insect; (4) a reliable quality control management system of the sterile insects that is applied routinely; (5) transport, fly emergence, handling and release technologies available; (6) adequate sterile to wild male overflooding ratios in the field in order to guarantee a significant induction of sterility in the wild population (Dyck et al. 2021). Most of these issues have been addressed by studies conducted with *A. fraterculus*, especially by researchers from Argentina and Brazil (Ortiz 1999).

Morphological and genetic studies have revealed that *A. fraterculus* is actually a complex of cryptic species (Morgante et al. 1980; Steck 1991), and as a consequence, each morphotype should be treated separately for the successful implementation of the SIT (Whitten and Mahon 2021). At least eight morphotypes have been recognized so far based on integrative taxonomy and their geographic distribution in Latin America has been defined (Hernández-Ortiz et al. 2012, 2015; Devescovi et al. 2014; Hendrichs et al. 2015; Prezotto et al. 2017). The results obtained by Dias et al. (2016) showed the existence of full mating compatibility among *A. fraterculus* populations from southern Brazil (populations from Vacaria, Pelotas, Bento Gonçalves and São Joaquim). Thus, southern Brazilian populations and Argentinean morphotypes are likely to belong to the same species within the *A. fraterculus* complex (Rull et al. 2012, 2013).

Significant progress has been made with the domestication and artificial rearing of *A. fraterculus* since the FAO/IAEA Workshop held in Viña del Mar, Chile, in November 1996 (Ortiz 1999), and large colonies have been successfully established in Argentina and Brazil (Salles 1992, 1999; Jaldo et al. 2001; Walder et al. 2006; Vera et al. 2007; Oviedo et al. 2011; Nunes et al. 2013). Walder et al. (2014) developed an artificial rearing system that allows rapid colony built-up and production of enough sterile insects for use in pilot-programmes. The available rearing system can still be optimized to increase insect yields. For example, the rearing costs at the Center for Nuclear Energy in Agriculture from the University of São Paulo (CENA/USP) were reduced by half in 2016 when agar for the larval diet of Salles (1992) was replaced with carrageenan. Ninety litres of pupae (ca. 3 million pupae) of a strain from Vacaria were produced by the F<sub>3</sub> to the F<sub>12</sub> generation with mean values of 77.4%, 77.0% and 0.49 for egg hatch, adult emergence and sex ratio (Q/Z+Q), respectively (Mastrangelo, unpublished data).

Radiation experiments with gamma and X-rays have shown that treating pupae of *A. fraterculus* with a dose of 40-60 Gy can induce 99% sterility in adult male flies (Bartolucci et al. 2006; Allinghi et al. 2007; Mastrangelo et al. 2010). Although the recommended sterilisation dose for treating pupae 48 h before emergence has been 70 Gy in Argentina (Cladera et al. 2014; Alba et al. 2016), radiation studies with both gamma and X-rays and field cage tests carried out at CENA/USP with the Vacaria strain in 2016 demonstrated that treating pupae 72 h before emergence with 40 Gy is sufficient to produce 99% sterile flies (that are competitive against wild flies). This dose is sufficient as doses higher than 15 Gy induce complete atrophy of the females' ovaries, and a sterile:wild male overflooding ratio of 45:1 induced more than 95% sterility in wild populations (Mastrangelo et al. 2018).

The sterile flies can be released by static ground-based devices (such as cardboard, plastic or PVC boxes), mobile ground-based systems (such as bags or cardboard containers being released from a mechanical device) (Dominiak et al. 2010; Bjeliš et al. 2013), or as chilled adult flies delivered from small aircrafts or even drones (Tan and Tan 2011; Mubarqui et al. 2014; FAO/IAEA 2017).

Despite all the advances made, some issues must be addressed yet, like automation of rearing processes, strain management and sex separation. Progress has been made to study the cuticular hydrocarbons and the chemical composition of the volatiles emitted by *A. fraterculus* males (Vanickova et al. 2012; Milet-Pinheiro et al. 2015), but no specific lure is so far commercially available that would increase the accuracy of field-monitoring. Overall, however, nearly all technical problems have been solved by scientists, and the implementation of the SIT against *A. fraterculus* can already be performed at pilot scales.

Most of the AW-IPM campaigns that include the SIT are composed of three phases of implementation: a preparatory pre-operational phase, a population reduction phase applying suppression measures and, then, the sterile insect release phase (Hendrichs et al. 2021). The pre-operational phase includes obtaining the commitment of all stakeholders, the development of funding mechanisms, of physical infrastructure (the establishment of mass-rearing, sterilisation, packing, fly emergence and release facilities) and securing appropriate management and human resources (i.e. strong leadership, dedicated full-time staff, development of institutional capacity, flexible and independent management structure), collection of baseline data on the distribution and population dynamics of the target species, public awareness, pilot trials in the field, continuity of the implementation of the critical components of the project, and independent reviews of it. Although Cladera et al. (2014) stated that most of these human and managerial components were still missing in Argentina, the case in southern Brazil is different. The Brazilian apple industry is strongly organized and competent to support an endeavour against another pest, as demonstrated by the successful eradication of C. pomonella achieved in 2014 after a 17-year campaign (Kovaleski and Mumford 2007; Capra 2014).

In north-eastern Brazil, some progress has also been made in the past few years with the management of fruit flies and mosquitoes after the establishment of Moscamed Brasil in the San Francisco River Valley. This programme has focused on the suppression of populations of *C. capitata* in the valley with the integration of sterile males with other suppression methods (Malavasi et al. 2007). More than 20

million sterile males *C. capitata* were produced for a pilot project in 2006-2007, that successfully suppressed wild populations in more than 2000 ha of mostly mango orchards (Moscamed Brasil 2007). However, most *C. capitata* populations from the San Francisco River Valley are still very high (FTD > 2) due to the presence of alternative hosts almost all year round and an excessive number of neglected orchards (França 2016). In southern states, on the other hand, temperate fruit growers can count on a unique climate advantage (natural suppression during the winter) that makes the management of fruit fly populations less costly.

# 5. HISTORY OF THE MOSCASUL PILOT PROJECT

The *Moscasul Biological Control and Integrated Fruit Fly Management Center* project, including a mass-rearing facility for *A. fraterculus* and its parasitoids, to be established by EMBRAPA Grape & Wine and the ABPM, was first presented to the Federal Government of Brazil on September 18<sup>th</sup> 2013, when it obtained the support of the Ministry of Science, Technology and Innovation. In December 2014, the Ministry of Agriculture, Livestock and Food Supply (MAPA) signed a cooperative agreement that included an investment of ca. USD 630 000 for the establishment of the Moscasul Center, and that was followed by parliamentary amendments from two senators and one congressman that increased that budget by USD 329 000. The total amount of resources obtained from authorities through these efforts by EMBRAPA and ABPM amounted to almost USD 959 000 for the initial phases of the project.

After this initial support, almost USD 600 000 were invested in constructing the mass-rearing facility at the Agricultural Experiment Station of EMBRAPA Grape & Wine at Vacaria, and the first containerized rearing modules were installed in May 2016 (Fig. 4). Twenty-one containers (fifteen larger ones: 12.9 m length x 2.9 m height x 2.4 width, and six smaller ones: 5.9 m x 2.9 m x 2.4 m) were installed in front of the agricultural station.



Figure 4. Containerized rearing modules installed for the establishment of the Moscasul Biological Control and Integrated Fruit Fly Management Center at Vacaria, Rio Grande do Sul, Brazil.

Each stage of rearing (i.e. adult colonies, larval rearing, pupal maturation and holding) will take place in separate containers. The selection of a modular system has several advantages: (1) less costly than building an entire single-unit brick facility,

(2) more species can be reared separately, (3) less susceptible to perturbations of daily operations (Tween 1987), and (4) insect production can be increased depending on the demand and availability of funds.

After the political and economic turmoil in Brazil in 2015, the new Minister of Agriculture visited Vacaria on August 9<sup>th</sup> 2016, when partnership protocols were signed with representatives of research institutes (Universidade Federal do Rio Grande do Sul and CENA/USP) and growers' associations (ABPM and the state farmer's associations AGAPOMI – Associação Gaúcha dos Produtores de Maçã, SINDOCOPEL – Sindicato das Indústrias de Doces e Conservas Alimentícias de Pelotas, and APPRP – Associação dos Produtores de Pêssego da Região de Pelotas), and an additional USD 158 000 were raised from federal funds. In August 2017, EMBRAPA approved an internal project of USD 185 000 to support and implement *A. fraterculus* monitoring and SIT activities.

A gamma or X-ray irradiator still needs to be acquired for the Moscasul Center for the sterilisation of the mass-reared flies. In the meantime, to avoid this constraint and more delays in field tests of the pre-operational phase, the CENA/USP at Piracicaba, São Paulo state, accepted to sterilize the mass-reared flies during the first years of pilot projects. Since the 1970s, research on fruit flies and the SIT has been carried out at this institute, also with the support of the Joint FAO/IAEA Division, and a pilot facility (250 m<sup>2</sup>) is present since 1998 dedicated to the production of sterile insects of several species (e.g. up to 2 million sterile *C. capitata* can be reared per week) and to training (Walder 2002). This centre is equipped with two gamma irradiators, i.e. a GammaCell-220<sup>TM</sup> and a panoramic Gammabeam-650<sup>TM</sup> (MDS Nordion International Inc., Canada), and an X-ray irradiator RS 2400V (RadSource Technologies Inc., Buford, Georgia, USA).

A pilot trial has been planned in three large apple orchards in Vacaria to demonstrate to growers the feasibility of using the SIT against *A. fraterculus*. Sterile flies will be released between the months of September and March only in surrounding zones of forests, covering a 50-100 m periphery of the target orchards, since these areas serve as reservoir of wild flies. Based on the monitoring data from 15 consecutive years of the selected orchards, approximately 150 000 sterile flies per week would be required for the first pilot release phase. The *A. fraterculus* colonies of the Vacaria strain have been well established at both the EMBRAPA and CENA laboratories. After the fine-tuning of all rearing and sterilisation procedures during the first half of 2017, CENA is planning to ship more than 200 000 irradiated pupae weekly by air to Vacaria for 6 months, starting in September when the level of the wild fly population is extremely low in the three pilot areas following the winter (average FTD < 0.5).

Most of the studies on marking and shipment procedures have been completed and the teams have received training in terms of surveillance, distribution of the sterile flies and identification of the caught insects. Depending on the level of production of *Diachasmimorpha longicaudata* (Ashmead) at EMBRAPA and CENA/USP at the time of the trials, this parasitoid is also intended to be released in some of the areas. The feasibility of shipping irradiated *A. fraterculus* eggs for the mass-rearing *of D. longicaudata* and *Doryctobracon areolatus* (Szépligeti) at Vacaria is also being assessed (Nunes et al. 2011; Costa et al. 2016). Arrangements to establish a pilot trial releasing sterile flies and parasitoids to suppress *A. fraterculus* in peach orchards from the region of Pelotas are also being made. The results of these pilot trails will have the potential to influence the direction of future control tests and to lead towards the sustainable management of *A. fraterculus* by hundreds of apple farmers and other temperate fruit-growing farmers from the southern states of Brazil.

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