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

A. KOVALESKI¹ AND T. MASTRANGELO²

¹*EMBRAPA Uva e Vinho, Estação Experimental de Vacaria, BR 285–Km 115, 95200-000, Vacaria, Rio Grande do Sul, Brazil*

²*Center for Nuclear Energy in Agriculture / Universidade de São Paulo, Av. Centenario 303, 13400-970, Piracicaba, São Paulo, SP, Brazil; piaui@cena.usp.br*

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 m² 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.

Key Words: *Anastrepha fraterculus*, *Diachasmimorpha longicaudata*, parasitoids, SIT, sterile insects, area-wide, 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. BioAnastrephaTM and CeratrapTM, 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, release-recapture 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 F₃ to the F₁₂ generation with mean values of 77.4%, 77.0% and 0.49 for egg hatch, adult emergence and sex ratio (♀/♂+♀), 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 18th 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 9th 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 m²) 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-220TM and a panoramic Gammabeam-650TM (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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