Final report

INVESTIGATION INTO CONTROL STRATEGIES OF THE POTATO LEAFMINER ON POTATOES

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ARC • LNR

Excellence in Research and Development

June 2021

CONTENTS

EXECUTIVE SUMMARY

The potato leafminer, *Liriomyza huidobrensis*, is a destructive vegetable pest in most countries of the world. In South Africa, it is considered one of the most important pests of potatoes, causing yield losses of up to 70 %. The majority of potato production regions reported the potato leafminer as always present and always a serious problem. Damage is caused by the larvae that mine in leaves, destroying the foliage in the process. The mystery of the origin of the first leafminers of the season was solved by placing yellow sticky traps at several places in and around new potato fields. Over a four-year study, it was consistently shown that leafminers appeared in previously harvested fields. No leafminers were trapped in other areas, which included virgin veld with weeds and grasses, and fallow areas and old fields surrounding the new potato fields. The appearance of leafminers in the previously harvested fields coincided with the first appearances of flies in the new field. The conclusion is therefore reached that leafminers originate from pupae that remain dormant in harvested fields. After winter, many of these dormant pupae eclose and reach new potato fields where they become available during spring. Because only the lower canopy (older leaves) can sustain leafminer populations in young potato fields, the leafminer reproduces here unnoticed until the top canopy is infested during the second half of the season. The second half of the season always showed a sudden or exponential increase in fly numbers in potato fields. Yellow sticky traps were very effective in monitoring the potato leafminer, but challenges relating to animal and wind damage required modifications to the trap setup. Only when the traps were modified so that the sticky sheet could freely rotate in a vertical as well as a horizontal plane did the challenges subside. The yellow stick traps were less environmentally friendly when non-target organisms were present and had to be replaced often when fly numbers were high and traps became clogged with dirt and mud. Evaluations of several other attractants that are used for fruit fly monitoring showed that they were not suitable for monitoring the potato leafminer. All the registered insecticides evaluated proved to be effective when applied on foliage in the laboratory. Additional evaluations against pupae showed no effect when they were drenched for two minutes in the laboratory. In other experiments, when pupae were buried beneath soil at depths of 16 cm, none succeeded to eclose into adult flies. The results of this research project showed that the use of insecticides is still an effective option but that alternative strategies are also available to enhance control during years of high infestations. In particular, the origin of the first leafminers was identified, and as such, served to broaden and enhance our knowledge about the ecology and epidemiology of one of the more serious pests of potato in South Africa.

BACKGROUND TO THE STUDY

Research team

Project leader

Dr Diedrich Visser

Collaborators

Regional managers of Potatoes South Africa

Participating potato farmers

Duration of the project

Commencing: June 2017

Concluding: June 2021

Chapter 1

INTRODUCTION

The potato leafminer complex

The potato leafminer is one of four leafminer species that are known to attack potatoes in South Africa, namely the potato tuber moth (*Phthorimaea operculella*), the tomato leafminer (**Tuta absoluta*), the potato leafminer (*Liriomyza huidobrensis*), and the American leafminer (*L. trifolii*) (Fig. 1). The potato tuber moth and *T. absoluta* are both micro-moths in the family Gelechiidae. The potato and American leafminers are flies in the family of Agromyzidae. Two of these leafminers, i.e. the potato tuber moth and the potato leafminer, are considered the most important pests on potato in South Africa (Visser, 2005; Visser, 2009). Because control actions differ for the different leafminers, it is important to distinguish between the species and their damage symptoms.

The larvae of both the potato tuber moth and *Tuta* make blotch leaf mines (wide irregular areas), while the two *Liriomyza* species make serpentine (long and thin) leaf mines (Fig. 1). In contrast with the American leafminer, the leaf mines of the potato leafminer usually coalesce, forming chlorotic areas, usually near the mid-rib of the leaf. Because they both make irregular "blotches" on leaves, leaf mines of the potato tuber moth and *Tuta* are more difficult to distinguish from each other.

**Tuta absoluta* was reclassified under its original genus (Chang & Metz, 2021) and should be referred to as *Phthorimaea absoluta* in future reports. We retain the name in this report as *Tuta absoluta* to prevent confusion.

Leafminer flies

Across the larger order of Diptera (true flies), leaf-mining occurs in nine different families, with Agromyzidae having the largest number of species (Mujica & Kroschel, 2011). The genus *Liriomyza* contains more than 300 species worldwide, of which 23 are economically important to agricultural crops (Parrella, 1987). Apart from the potato leafminer, *L. huidobrensis*, two more leafminers in the family, Agromyzidae attack vegetables in South Africa, namely: *L. trifolii* and *Chromatomyia horticola* (Myburgh, 1988; Visser, 2009). More *Liriomyza* leafminer pest species occur in other African countries. Because it is sometimes difficult to distinguish different agromyzid species, it is possible that more than the three species mentioned above occur in South Africa.

Liriomyza huidobrensis (potato leafminer) is a relatively new introduction into the country (in the year 2000), while *Liriomyza trifolii* (American leafminer) has been present in South Africa since the 1970s (Prinsloo & Uys, 2015). The potato leafminer was introduced into many new countries in the 1990s, including Canada, European countries, Indonesia, Israel and Taiwan (Scheffer *et al*., 2001). Potato leafminer is also present in China, India and the USA and presumably originated in South America (Weintraub *et al*., 2017). Potato leafminer was first encountered in South Africa in 2000 in the Sandveld potato production region (Visser, 2001; Visser, 2005). The American leafminer originated in North America and, except for Australia and some parts of eastern Asia, is distributed worldwide (CABI, 2019). Although *L. trifolii* (the American leafminer) was on occasion troublesome on potatoes before the arrival of *L. huidobrensis* (Visser & Barnard, 1992), the extent and yield losses were not as severe. It seems that the potato leafminer has displaced the American leafminer on vegetables in South Africa, but research is needed to verify this assumption. The pea leafminer, *Phytomyza horticola*, is an uncommon and little-studied pest of peas. Note: The potato leafminer (*L. huidobrensis*) is also known as the pea leafminer in the USA, and this may cause confusion between the two species. *Phytomyza horticola* does not occur in the USA.

The adults of *Liriomyza* leafminers are small, yellow-and-black flies approximately 2 - 3 mm in length (Fig. 1). The males of both species are smaller than the females. The American leafminer is also slightly smaller and more yellow than the potato leafminer. The legs of the potato leafminer are dark, whereas those of the American leafminer are yellow. Both species have red eyes and a distinct yellow area on the upper thorax between the wings. American and potato leafminers live for approximately one and two weeks, respectively (Capinera, 2001) and may lay up to 400 eggs. Eggs hatch in about three days and the emerging larvae (maggots) tunnel between the upper and lower leaf surfaces. The cream-coloured to orange larvae are legless and are never found outside their tunnels within leaves. While feeding, the larvae pass through three instars in as few as four days. At maturity, the third instar cuts a slit in the leaf surface, after which it exits the leaf mine. The larvae then usually fall to the ground and pupate on the soil surface, but sometimes they pupate on the surface of a leaf. The pupae are orange to brown (Fig. 2) but turn dark brown to black just before eclosing into flies. It takes approximately 10 days for the adult fly to emerge from the pupa.

Pupae are able to survive very cold field conditions by gradual adaptation as temperatures decline during the autumn months (Chen & Kang, 2004). Cold winters with temperatures as low as minus 11 °C are not enough to kill exposed pupae on the soil surface (Van der Linden, 1993). In fact, it seems that the potato leafminer is adapted to cooler environments. In studies with *L. huidobrensis*, it was found that the upper temperature limit for *L. huidobrensis* was 30 °C (Lanzoni *et al.,* 2002). In other studies, it was found that the potato leafminer reproduced the fastest at 23-25 °C, and decreased with temperatures above 27 °C (Weintraub *et al.,* 2017). In certain countries, the potato leafminer is never a problem during the summer, although they may be a serious pest during the autumn and spring months (Weintraub & Horowitz, 1995).

Potato leafminer

The potato leafminer has been recorded worldwide from 365 host plant species in 49 plant families (Weintraub *et al.,* 2017). The majority of host plants are weedy crops, followed by cultivated crops (32 %) and cultivated flowers (18 %). Indications are that certain potato cultivars may be preferred by female potato leafminer flies (Lopez *et al.,* 2010), but external choices may influence that cultivar choices because similar tests in the laboratory were inconclusive (Fenoglio & Salvo, 2009; Weintraub *et al.,* 2017). Mujica (2016a) eluded that the ecosystem where potatoes are grown, e.g. highland vs. lowland, may dramatically influence the crop's susceptibility to the potato leafminer.

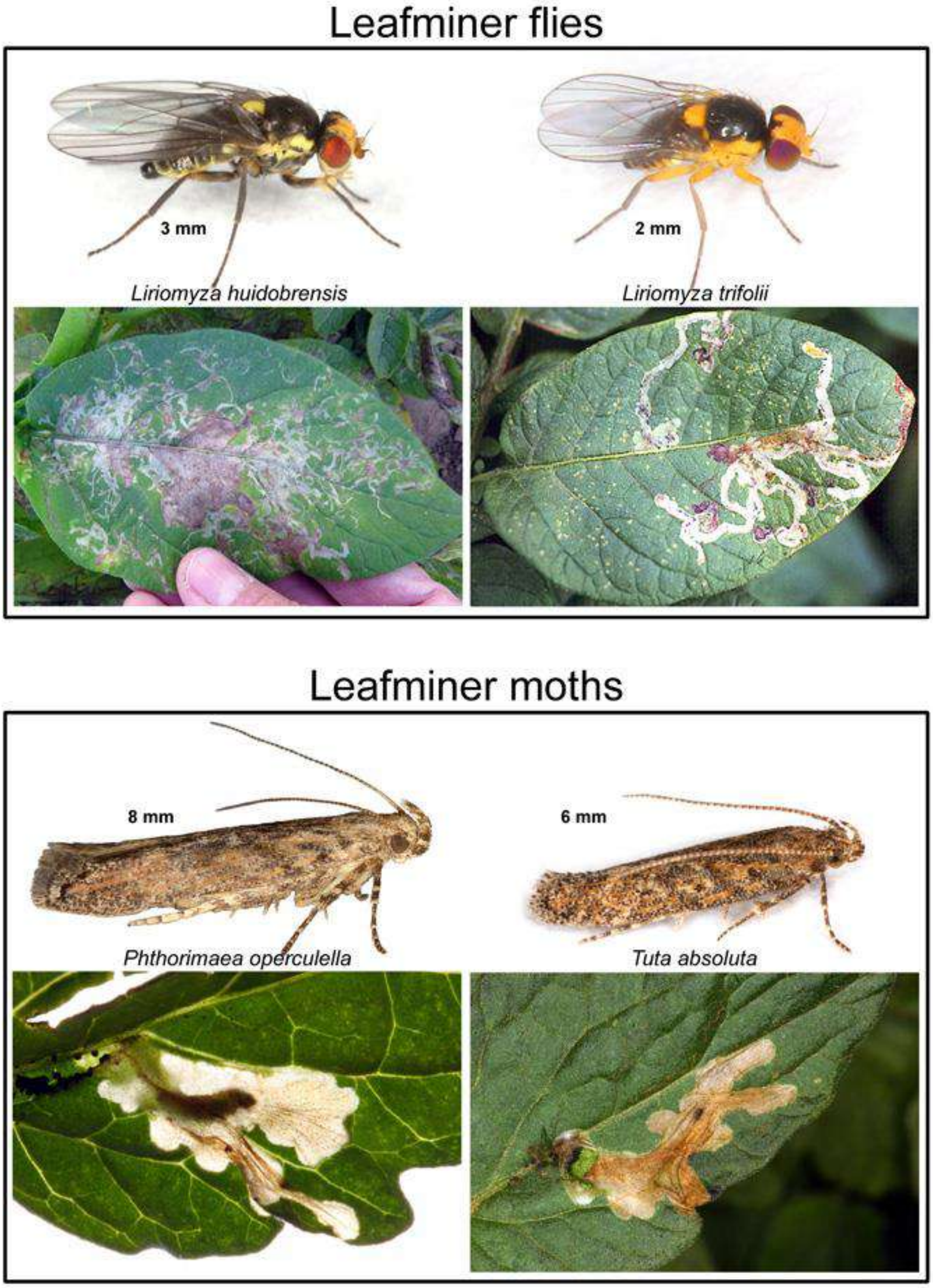

Fig. 1. The four leafminers that attack potatoes in South Africa, with the damage symptoms beneath each photo. The approximate sizes of the leafminers are given.

Fig. 2. Potato leafminer pupae (left), showing the strong outer puparia that protects the pupae inside. On the right are leafminers emerging from their pupae inside the puparia.

The potato leafminer is one of the most important pests of potatoes in South Africa. In a survey to determine the pest status of all pests on potatoes in South Africa, it was found that 13 of the 16 potato production regions reported that leafminers were always a serious problem (PSA, 2017), which makes leafminers the most important pest group in South Africa, followed by the potato tuber moth, reported by 11 regions is always a serious problem. One of the 13 production regions that indicated leafminers as a serious pest, i.e. the Sandveld region, experienced extraordinary high pest pressure during 2016. Even in the colder months of the year, i.e. May to July, fly numbers were higher than normal with very high infestation levels on the leaves. Pest pressure remained high, despite diligent spraying programmes with registered insecticides.

Yield losses due to the potato leafminer can be as high as 70 % (Visser, 2009), and reduction in the photosynthetic ability of foliage up to 62 % (Chabi-Olaye *et al.,* 2008). Although the adult flies may stress the plants by puncturing them with their ovipositors, feeding by larvae results in leaves, and sometimes entire haulms/stems, dying off. Affected growth also looks poor and may resemble diseased plants. Plants usually die off a month earlier than would be normal during senescence. This premature dying-off causes the yield loss associated with this pest on potatoes. Potato plants need the latter part of the season to grow or set the tubers initiated during the first half of the season. The result is that mostly small and medium-sized tubers are produced – the reason for lower yields.

An interesting phenomenon is that the larvae of *Liriomyza* leafminers on potatoes can only survive on older plants or plants/leaves that grow more slowly (e.g. the lower leaves) (Videla & Valladares, 2007). Younger potato plants, although attacked and punctured by females, usually never harbour high numbers of larvae. Therefore, the onset of damage is nearly always sudden and dramatic towards the latter part of the growing season (with the onset of senescence). This dramatic "invasion" of leafminers is exacerbated by the extremely short lifecycle of the larvae, which may be as short as four days. This phenomenon of "delayed but sudden attacks" on potatoes has not been observed in other vegetables (Visser, 2009).

The potato leafminer is a problem wherever potatoes are grown. However, in some countries, the status of the leafminer seems to have diminished over time (Weintraub *et al.,* 2017). According to the authors, this could have been attributed to natural control. In other countries, *Liriomyza* leafminers vary in their significance as an important potato pest depending on different production systems (Foba *et al.,* 2015). The choice of potato cultivar may also play a role in the severity of damage (López *et al.,* 2010; Mujica & Kroschel, 2013). Other studies stressed the importance of the ecology of the leafminer when control strategies are implemented (Mujica, 2016a).

Preventative insecticidal control is the main control strategy used by all potato farmers in South Africa (Visser, 2002; 2011). Thirty insecticides, comprising 11 active ingredients supplied by 18 agro-chemical companies, are currently registered for leafminer control on potatoes in South Africa (Croplife South Africa, 2016). However, despite implementing thorough spraying programmes, leafminers remain the number one pest of potato (PSA, 2017). Some regions are experiencing increasing pest pressure and yield losses, while other regions regard leafminers as a persistent problem that forms a major part of their pest input costs. No resistance has been proven in insecticides used against the potato leafminer in South Africa, and all should presumably be effective. However, some researchers argue that although insecticides are effective, they also kill natural enemies like parasitoids, and this elimination of natural enemies is part of the problem in some countries (Ewell, Fuglie & Raman, 1994).

Parasitoids play an important role in reducing agromyzid leafminer numbers in potato fields (Musundire, Chabi-Olaye & Kruger, 2011). More than 80 different species were reported to parasitise *Liriomyza* leafminers (Liu *et al.,* 2009). *Diglyphus isaea* is an important leafminer parasitoid that occurs worldwide and also in South Africa. In Israel, Weintraub and Horowitz (1995) found the occurrence of this parasitoid, in relation to the leafminer flies, to be 30:2, in favour of the parasitoids, in unsprayed fields. However, when fields were sprayed, the relationship was reversed to 2:49 in favour of the leafminer flies. The efficacy of parasitoids as effective natural enemies is highly variable; reported control figures range between 1 % and 90 % (Rauf, Shepard & Johnson, 2000; Weintraub *et al.,* 2017).

The origin of the first leafminers of the season was mostly unknown when this study was initiated. The small leafminer flies may occur in large numbers in potato fields, sometimes soon after plant emergence. On other occasions, the infestations start much later; late infestations usually result in less severe attacks and reduced yield loss. Occasionally, newly planted crops are attacked by large numbers of flies; in some cases, swarms cover entire plants. Whether the leafminer arrived early or later in the season, the origin was still mostly a mystery. It was thought that the first leafminers entered fields from the surrounding areas where wild solanaceous host plants may sustain viable populations in the off-seasons. However, wild hosts are never abundant, and in the summer rainfall regions, these hosts die off during the winter months.

Therefore, the major objective of this study was to identify the origin of the flies that infest new potato fields early in the season. Other objectives included the potential of traps to monitor the presence and movement of the flies during the early season, and the efficacy of registered insecticides against the mining larvae, in the laboratory. Potential control options to eliminate the pupal stage of the leafminer were also investigated, i.e. the effects of registered leafminer insecticides and the potential to kill the pupae by burying under a layer of soil.

Chapter 2

MATERIALS AND METHODS

Origin of the first leafminers of the season

2017

During the first year of the study (2017), it was still unknown where the first leafminers of the season originated. A trial was therefore designed to monitor a newly planted field, as well as the surroundings of the field. The newly planted field was approximately half a hectare in size and planted with a mixture of cultivars and lines, on a red Hutton loamy-sandy soil, at ARC-VIMP, Roodeplaat, Pretoria. All production practices were according to standard farmer practices, including irrigation and the application of pesticides. Thirteen yellow sticky traps (17 cm X 10 cm) were placed in four transects around the potato field (the four red lines in Fig. 3). The traps were installed as follows; one trap in the middle of the new field, and for each of the four transects, 25 m, 50 m and 100 m from the edge of the field (Fig. 3). The south-eastern transect reached a potato field that was harvested in the previous season just before winter. This transect was later moved slightly to reach a new field that emerged during week nine of the study (Fig. 3, green area indicated as "New field"). The number of leafminer flies was counted on both sides of each trap on a weekly basis. Figure 4 illustrates how leafminers can increase on a trap and why the time intervals could not be extended for longer than a week due to the saturation of the traps.

2018

Leafminer flies were monitored in and around a half-hectare experimental potato field at the ARC-VIMP, Roodeplaat, Pretoria (Fig. 5). The field is marked as "2018" and outlined in green on the figure. The field was planted with a mixture of cultivars and lines on a red Hutton loamy-sandy soil. The emergence date (week one) was 14 September 2018. All production practices were according to the usual farmer practices, including irrigation and the application of pesticides.

In addition to the 2018 field, 11 more yellow sticky traps (numbered 2 to12) were placed in strategic locations across the 15 ha cultivated area (Fig. 5). These locations included previously harvested potato fields (orange rectangles) and a harvested bean field (area 7). Of all the harvested fields in 2017, only areas two and three had significant infestations of the potato leafminer during the growing season of 2017.

The purpose of the 11 additional traps was to establish where the first leafminer flies would emerge in the early season, from September 2018. This was a repeat of the 2017 experiment during which leafminer numbers were monitored around a target field. However, during 2017, no leafminers appeared on traps located in virgin fallow areas around the field. Therefore, for the 2018 season, it was decided to place traps only in previously harvested fields and cultivated areas near the target field.

As for the previous season, the number of leafminer flies was counted on both sides of each trap (17 cm X 10 cm) on a weekly basis for 13 weeks. At week 14, most plants in the target field were dead (late senescence), and the flies were no longer encountered on the sticky traps.

2019

Two newly planted and five previously harvested potato fields were monitored with yellow sticky traps for the emergence of potato leafminer adults. All fields were planted with a mixture of cultivars and lines on a red Hutton loamy-sandy soil. The emergence date (week one) for the newly planted fields was 1 November 2019. All production practices were according to the usual farmer practices, including irrigation and the application of pesticides. The same methods were used as for the first two years for monitoring the emergence of the first leafminers. The experiment was terminated at week 14 because the majority of plants were in a late senescent stage. All the fields were located in the same 15 ha area as indicated in Fig. 5 (fields not highlighted).

2020

During 2020 two new potato fields and two harvested fields were monitored, using the same methods as in the previous three years, on the same 15 ha area. The emergence date was 9 October 2020. The monitoring of the leafminers on the sticky traps continued until week 13 when the plants started to die off naturally.

Investigation into traps for the potato leafminer

Monitoring for pests and diseases is a critical component in control strategies of crop pests (Dara, 2019; McCravy, 2018). No commercial trap is available to monitor *Liriomyza* leafminers. Yellow sticky traps are designed to monitor aphids and thrips. Although they can be used to monitor leafminer flies in fields, they only catch flies in the vicinity of the trap. Pheromone traps attract insects from relative distances and are usually the preferred trap type for pest monitoring. However, the online database, *Pherobase.com*, does not list pheromones for *L. huidobrensis*. A meeting with Dr Marc Bouwer (a scientist in chemistry and a pheromone expert at FABI, University of Pretoria) was arranged for 28 June 2018 to discuss the possibility of developing pheromone lures for the potato leafminer. He indicated that they could investigate if the potato leafminer emits pheromones but that this would be a separate and major project. The ideal would be to develop yellow sticky traps with impregnated leafminer pheromones. Such "impregnated" sticky traps already exist for certain thrips species. Follow-up discussions with Dr Bouwer, and suppliers of traps and lures, may be included in a future, separately funded project.

Due to the lack of commercial traps for *Liriomyza* leafminers, two types of traps were evaluated for monitoring the adults in and around potato fields, namely yellow sticky traps and yellow water pan traps with soap added (Fig. 6). Yellow sticky traps are always commercially available, while yellow water pan traps are always custom-made and are also used to monitor aphids. The yellow sticky traps and water pan traps were evaluated side-by-side to determine their efficacy and practical usability for monitoring adult leafminer flies in potato fields.

Several problems had to be solved while evaluating the yellow sticky traps. Firstly, they are difficult to handle once the outer packaging is removed; the sticky glue used on most commercial traps excessively sticks to anything it comes into contact with. The glue adheres to the operator's fingers and clothing and it is not possible to remove it completely. We experimented with latex and nitrile gloves but found it impossible to detach the part of the glove that came into contact with the glue on the trap to the point that the gloves broke and tore off rather than detached. The same was found with rubber gloves; it was very difficult to remove the part of the glove that was stuck on the glue. The only option was to handle the traps bare handed, but with extreme caution, only touching the upper areas which do not contain any glue.

Adding to the difficulties of handling sticky traps as described above, managing the traps, and counting leafminer flies on the traps produced additional challenges. These challenges included: a) traps were destroyed by wind and/or animals, b) traps were covered by windborne sand and soil, c) traps clogged up too quickly when leafminer numbers were very high, and d) counting leafminers on the traps became difficult when more than 50 flies were encountered on a single side, and when traps were left in the field for longer than seven days.

To prevent wind and animals from damaging the traps, extensive experimentation was undertaken to find the correct design to counter the effects of "blowing" (wind) and "pushing" (animals) against the traps. The final design is illustrated in Fig. 6A. The trap is loosely attached to a bent 4 mm iron rod, using thin cable ties. To prevent the cable ties and trap from sliding to either side, vial stoppers are inserted on both sides. Some commercial sticky trap designs use thin cardboard paper as the base, which tear easily at the holes. For such traps, the areas around the holes were therefore reinforced with extra cardboard, attached with thick masking tape and staples. For some traps, there is enough space to fold the upper area, where the holes are situated, onto itself, to strengthen the area, but then new holes have to be drilled into the folded area. Traps that are made of plastic usually do not tear in the hole areas. Our current design allows the sticky trap to rotate on both the horizontal and vertical axes when forced in any direction by the wind or animals. All other previous designs were either completely torn out and ripped from the iron rod by the wind or pushed over by animals.

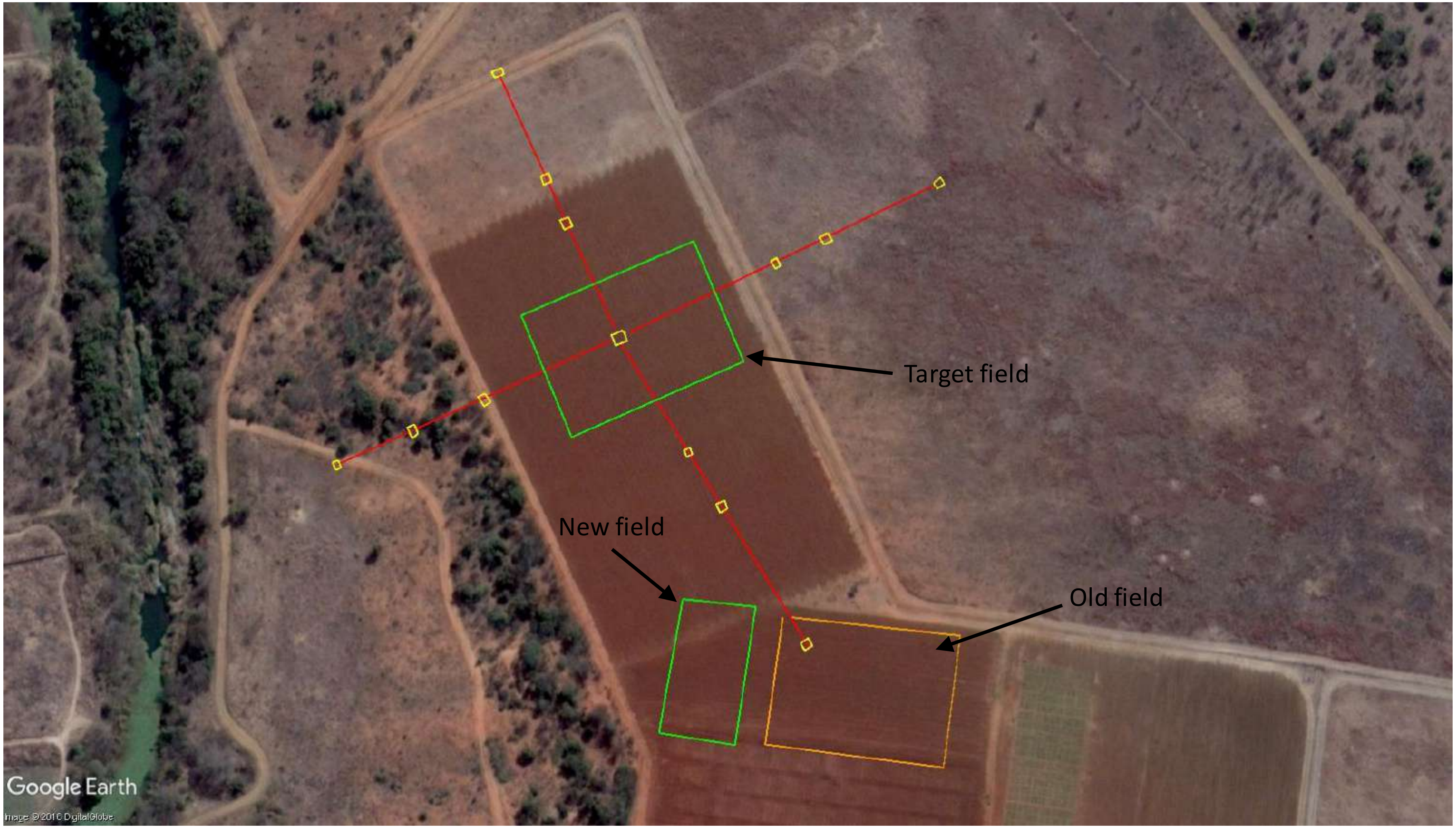

Fig. 3. Field trial plan of the area where leafminer flies were monitored. Yellow squares indicate the positions of the yellow sticky traps. Red lines indicate the transects; the traps were placed on the transect lines, at three distances from the field edge, i.e. 25 m, 50 m, and 100 m. Plants in the target field emerged 6 September 2017 (week 1). Plants in the new field emerged during week 9. The old field was harvested the previous season before winter.

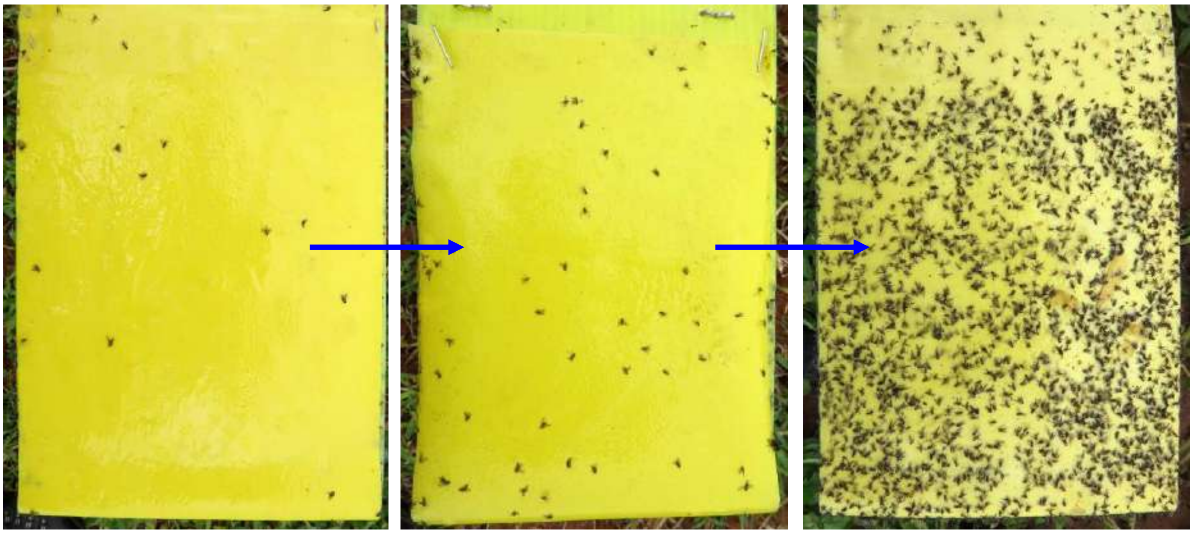

Fig. 4. Examples of leafminer catches on yellow sticky traps, from low density to very high density. To prevent the difficulties associated with counting the small flies on very high density traps, all traps were counted and replaced on a weekly basis.

Traps were inspected frequently and replaced when covered by windborne sand, soil and dust (Fig. 6E). However, when the potato plant grew vigorously and closed its canopy, this problem subsided. During some seasons, leafminer numbers increase so rapidly that sticky traps may become clogged with hundreds of flies in a very short time (Fig. 6C). Such traps do not catch flies any longer and become obsolete. Due to sand and dust that were blown onto the traps, such traps were also replaced when needed. Fortunately, this rarely happened during our experiments, and it may only become a problem when the traps are left in the field for more than a week. All our traps were inspected and replaced on a strict seven-day interval and lifted to canopy level as the plants grew taller (Atakan & Canhilal, 2004).

Counting of leafminer flies on traps was conducted in the field. A hand-held counter was used when numbers increased to more than 10. Both sides of the trap were counted and added together for one trap location. When needed, a head-mounted magnification tool (Optivisor x10) was used to distinguish between *Liriomyza* leafminers and other similar looking midges. However, when fly numbers increased to more than 50, counting became difficult, even while using a counter. Some traps are divided into grids to facilitate and assist in the counting process (Fig. 6D and 6E). These grids simplify the counting process by counting grid by grid or selecting only a few random grids and multiplying the average per grid chosen with the total number of grids afterwards. However, in our study, the leafminer flies were never distributed homogenously on traps, and the latter method was therefore not used – all flies on traps were counted.

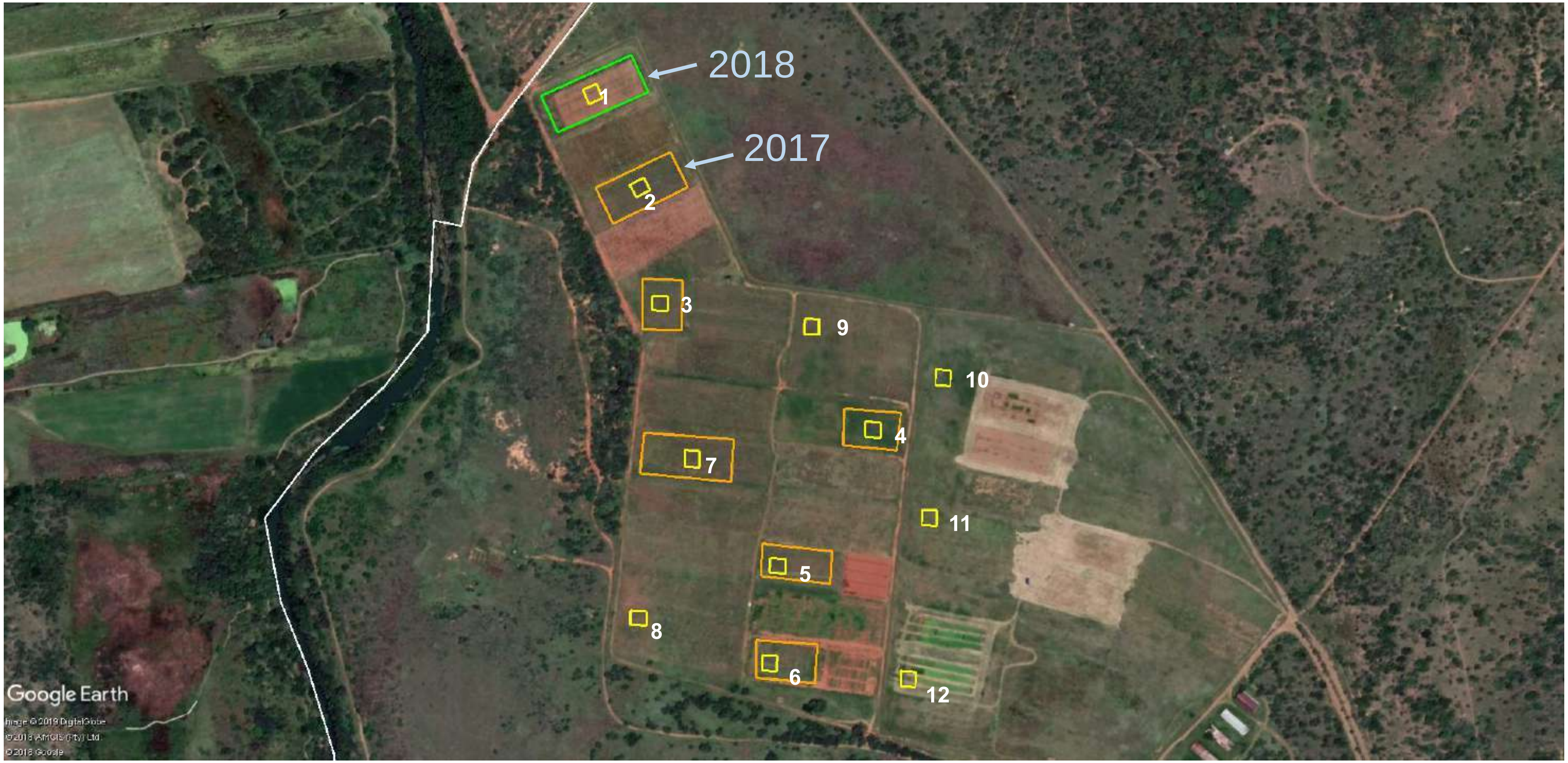

Fig. 5. Field trial plan of the areas where leafminer flies were monitored with yellow sticky traps. Yellow squares indicate the positions of the sticky traps. The target field that was monitored during the 2018 season is indicated in green. The field trial that was monitored as the target field in the previous year is indicated as 2017. Of all the areas that were harvested in the 2017 season (indicated in orange), only areas 2 and 3 had notable leafminers infestations during 2017.

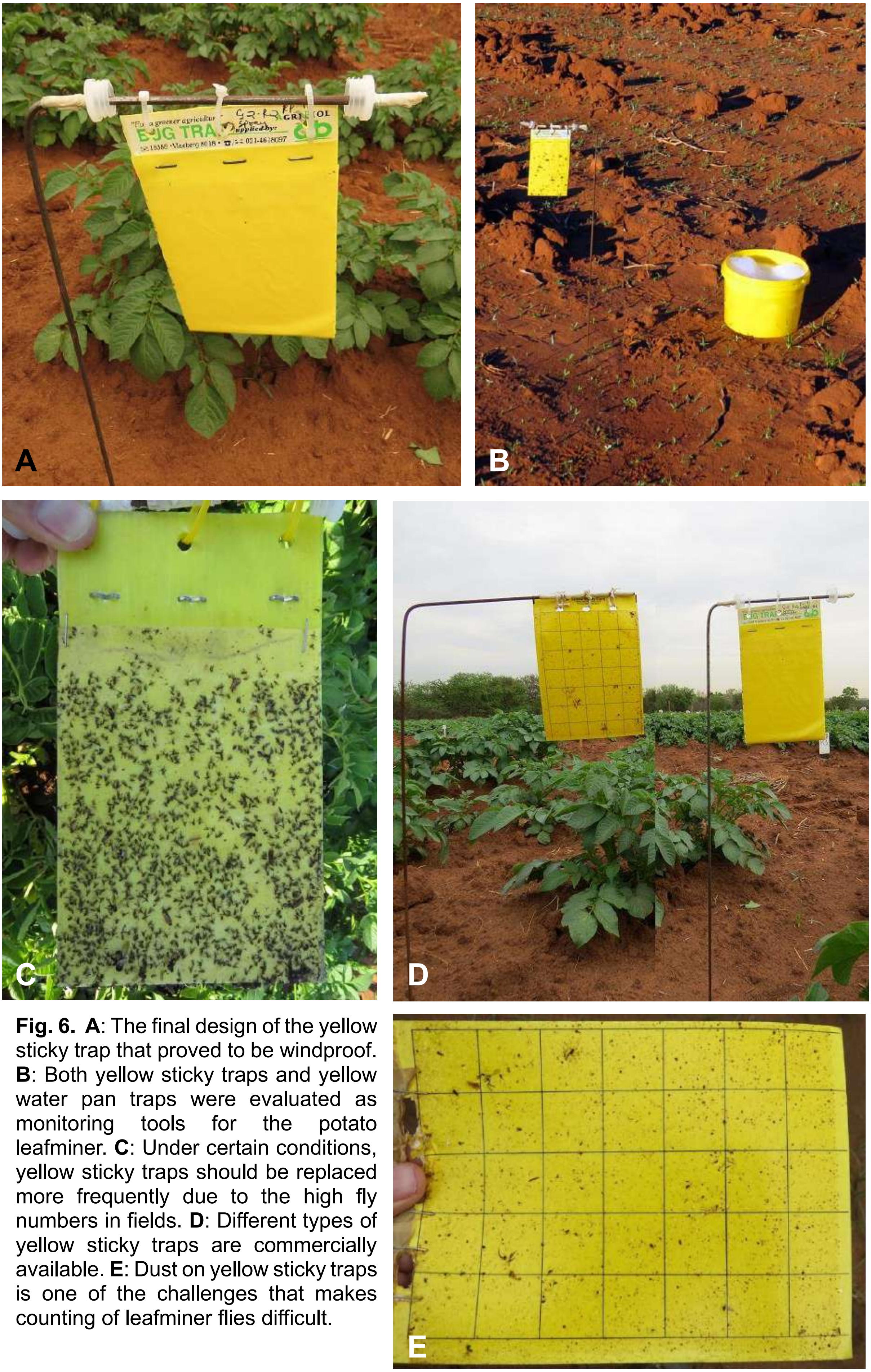

Fig. 6. A: The final design of the yellow sticky trap that proved to be windproof. **B**: Both yellow sticky traps and yellow water pan traps were evaluated as monitoring tools for the potato leafminer. **C**: Under certain conditions, yellow sticky traps should be replaced more frequently due to the high fly numbers in fields. **D**: Different types of yellow sticky traps are commercially available. **E**: Dust on yellow sticky traps is one of the challenges that makes counting of leafminer flies difficult.

We investigated the time that yellow sticky traps could be left in the field before counting flies. It was found that when traps were left for longer than seven days, they not only became clogged with debris and sand/dust but identifying the flies also became difficult. The glue seemed to "cement" the small flies deeper into the glue layer and distort and obscure the vital markings on the flies. Separation of the *Liriomyza* flies and other midges becomes very difficult under such circumstances. Traps were therefore never left in the field for longer than seven days.

When leafminer fly numbers on traps became very high, e.g. when more than 50 flies were present on traps per side, we experimented with taking high-definition close-up photographs of the traps. The photographs were then magnified on a computer screen to do the counting. This method proved to be very effective and easy to use, especially when other midges were also present on the same trap. With this method, a digital record of each trap can be stored on a computer and used later for verification purposes when needed. We did not utilise the "Ziploc method", whereby traps are placed in Ziploc bags for counting in the laboratory. This method is known to "squash" very small insects deep into the glue, after which identification becomes challenging.

Hym-lure is a commercial fly attractant, which is not a pheromone, and therefore attracts flies of both sexes. Hym-lure is a protein hydrolysate liquid concentrate used to prepare bait for the control of fruit flies. The concentrate is mixed with a suitable insecticide and used as an "attract-and-kill" formulation to kill flies attracted to the sprayed solution. Because it can be sprayed on any surface, it is also used to prevent the migration of fruit flies from infested to uninfested orchards. During discussions with role players at a leafminer workshop in the Sandveld, it was mentioned that Hym-lure was added to spraying tanks to attract the leafminer when spraying insecticides. This was presumably done to increase the effectiveness of certain insecticides. Because migration of the leafminer from harvested to new fields seemed to be the main route of new infestations, it was decided to investigate if Hym-lure could be used as an attractant for emerging flies in harvested fields. If an attractant could be found, it could potentially be used to mass capture or "attract and kill" flies before they move out to infest newly planted fields.

An experimental sample of Hym-lure (500 ml) was obtained from Villa Crop Protection. A concentration of 4 ml Hym-lure per litre of water was used to prepare the "bait" solution, which was the maximum concentration prescribed on the label. No insecticides were added – the objective was to investigate if leafminer flies were attracted to the solution and not if it would increase the effectiveness of insecticides.
The experimental layout comprised four delta traps, placed two metres apart in a square formation (Fig. 7). The sticky liners inside the traps were used to catch the flies, and the choice of trap (Delta trap) was purely to protect the sticky liner and Hym-lure inside. Three traps were used to add the Hym-lure solutions, while a fourth was used as a control. Trap one was sprayed with the solution on the insides, trap two contained a small container lid (30 mm in diameter), acting as a saucer, with cotton wool drenched with the attractant, and trap three was placed next to a potato haulm that could reach inside the trap. The distal end of the haulm was dipped into the Hym-lure solution and then placed inside one end of the trap. A white cardboard sticky liner was supplied on the inside floor of each trap. The small container lid (with the cotton wool and attractant), and the haulm, was placed on top of the sticky liner. Enough space was available on the sticky liner to catch flies that may have entered the traps. All traps were removed after two weeks, and the sticky liners were inspected for the presence of leafminer flies.

Apart from Hym-lure, more commercial fly attractants are available to attract fruit flies as a means of pest control. After discussions with a fruit fly expert at the University of Pretoria, namely Prof. Chris Weldon, he kindly supplied the following attractants to include in our evaluations: E.G.O. Phero Lure for *Ceratitis capita*; Chempac Fruit Fly Lure; Fruit Fly Lure (names as appearing on the packaging of the lures).

Fig. 7. The experimental layout for evaluating Hym-lure as an attractant for leafminer. flies.

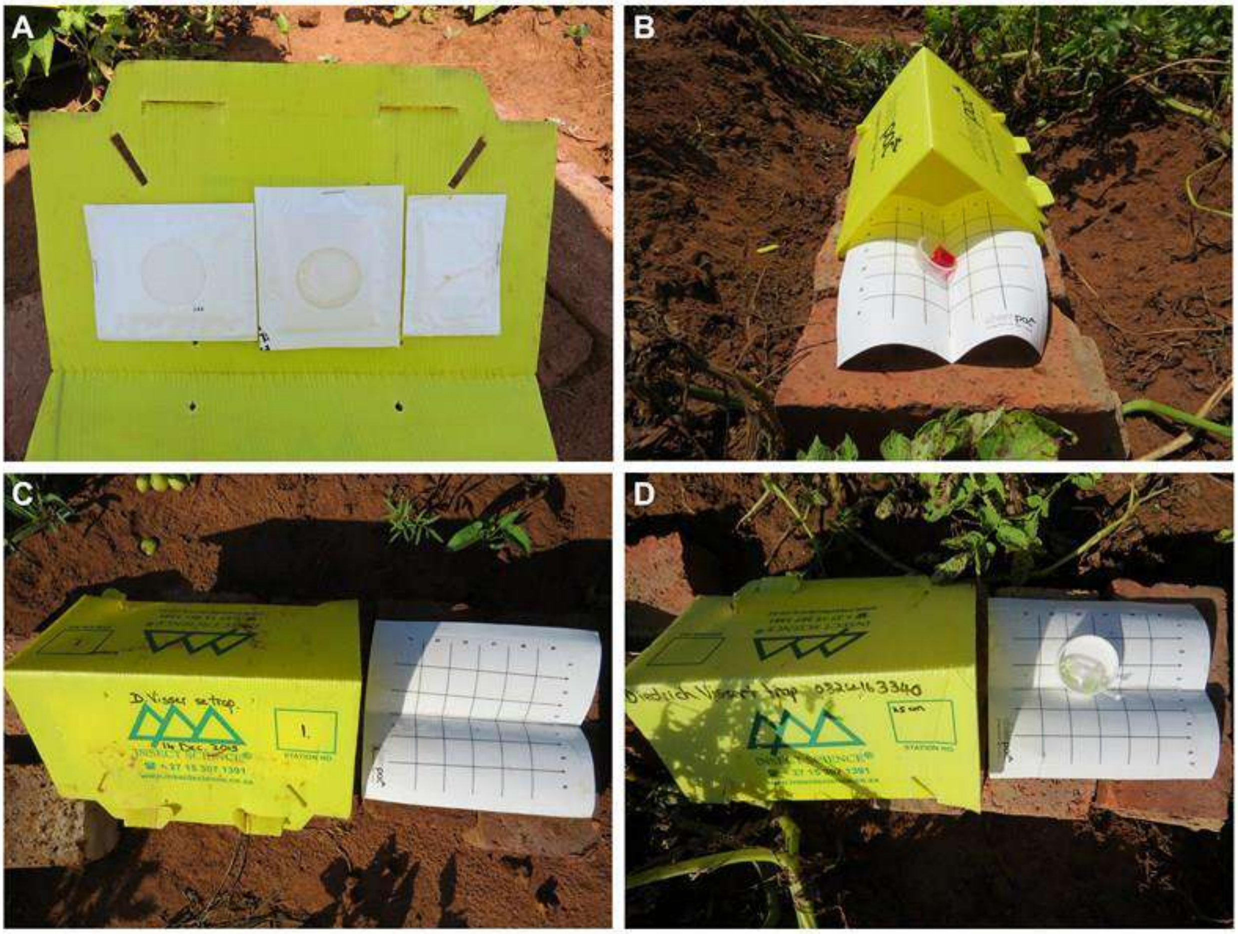

Fig. 8. **A.** Chempac Fruit Fly Lure, in envelopes, attached to the roof of trap; **B.** Fruit Fly lure in a red jelly formulation, place on the sticky liner; **C.** Control; **D.** E.G.O. Phero Lure for *Ceratitis capita,* in a closed bulb, placed on the sticky liner.

The same trial design was used as with Hym-lure, i.e. four Delta traps in a square formation (Fig. 7). The three attractants are formulated in plastic carriers, as illustrated in Fig. 8. The control was a Delta trap with only a white sticky liner.

Rearing programme for laboratory tests

The potato leafminer was reared on potted tomato plants in insect-proof cages (Fig. 9). To ensure a continuous supply of plants of the correct height (approximately 20 cm), a special tomato-rearing programme had to be established in a greenhouse facility at Roodeplaat (Fig. 10). Seeds were planted weekly to ensure a continuous supply of plants of different ages and heights. Leafminer populations were augmented from infested leaves collected in potato fields. Enough leafminer adults and pupae were reared to do laboratory experiments.

Fig. 9. The potato leafminer was reared on potted tomato plants in insect proof cages.

Fig. 10. The greenhouse facility in which the tomato plants were reared (left), and tomato plants at the correct height for use in insect cages (right).

Evaluation of registered insecticides

The insecticides procured for evaluations against the potato leafminer are shown in Fig. 11. The list of insecticides registered against the potato leafminer, and the dosages used for the laboratory bioassays, are indicated in Table 1 (source for registered insecticides: CropLife, November 2018).

Fig. 11. Insecticides procured for laboratory bio-assays for their efficacy against the potato leafminer.

For evaluations against pupae, the insecticide solutions were prepared in translucent one-litre blue screw-cap media bottles. Distilled water with a pH of approximately 6,5 was used, with no additional additives. The control was distilled water only. One treatment replicate consisted of one Petri dish with four pupae placed on a 7-cm filter paper disk (Fig. 12).

Fig. 12. An illustration of the Petri dish bioassays method, in which potato leafminer pupae were evaluated for their susceptibility to insecticides.

Two millilitres of an insecticide solution were added to the filter paper, after which a second filter-paper disk was placed on top. The solution was enough to saturate both the upper and lower filter-paper disks, with the four pupae sandwiched between them. The pupae were exposed to the insecticidal solutions in this manner for two minutes, after which they were removed and placed in clean Petri dishes with dry filter-paper disks. Three weeks later, all treatments were inspected for the emergence of flies.

For the evaluations of insecticides against larvae on foliage, adult flies were firstly introduced into insect-proof cages containing potted tomato plants (variety Floradade) (Fig. 13). These flies were previously collected from a pre-rearing phase (Fig. 9). Flies of the same ages were released into the incubation cages, approximately 15 flies per plant (Fig. 13). The flies were allowed to mate and lay eggs in the leaves of the potted plants.

Fig. 13. Incubation cages with potted tomato plants.

After a few days, when the first leaf mines appeared on leaves, individual plants were chosen randomly from the incubation cages (Fig. 13). Plants were sprayed with treatments until run-off, ensuring 100 % leaf coverage (Fig. 14a). One replicate consisted of one potted tomato plant, and three replicates were used per treatment. The control was distilled water only. After treatment, the plants were isolated in buckets to determine the number of pupae that would emerge from individual plants two weeks later (Fig. 14b). A piece of white carton was placed around the stem of each plant to prevent the pupae that fell off the plants into the bucket from landing in the soil in the pot (Fig. 14b). The pupae were collected from each bucket for two weeks. The room temperature was kept at approximately 25 °C, and humidity was not controlled. The number of pupae that emerged from the control was compared with the number of pupae that emerged from each treatment to establish the efficacy of the different treatments.

Active ingredient	Formulation	Mode of action*	IRAC** group	Dosage on label	Lab dose
1. abamectin	84g/L, SC	TL, Stom, Cont	6	108 ml/ha	0,22 ml/L
2. azadirachtin	3 g/L, EC	Stom	UNE	500 ml/100L	5 ml/L
3. cartap hydrochloride	500 g/kg, SP	Sys, Cont	4c	400 g/100L	4 g/L
4. chlorantraniliprole	200g/L, SC	Stom, Cont	28	200 ml/ha	0,4 ml/L
5. chlorantraniliprole / lambda-cyhalothrin	100 g/L and 50 g/L, SC	TL, Cont, Stom	3 + 28	200 ml/ha	0,4 ml/L
6. cyromazine	750 g/kg, WP	Sys, Cont, Stom	17	200 g/ha	0,4 g/L
7. indoxacarb (to add abamectin)	150 g/L, SC	Stom, Cont	22	250 ml/ha	0,5 ml/L
8. novaluron	100g/L, EC	Stom, Cont	15	350 ml/ha	0,7 ml/L
9. oxamyl	310 g/L, SL	Sys	1A	3 L/ha	6 ml/L
10. spinetoram	250 g/kg, WG	Cont, Stom	5A	150 g/ha	0,3 g/L
11. spinosad	480 g/L, SC	Cont, Stom	5A	300 ml/ha	0,6 ml/L
12. pyridalyldichloropropene – derivative***	500 g/L, EC	Cont, Stom	UN	200 ml/L	0,4 ml/L

Table 1. Insecticides registered against the potato leafminer (Crop Life SA, November 2018). Only the active ingredients are indicated, with the formulations, modes of action, and the IRAC group code. The dosage on the label and the dosage used in the bioassays against the pupae are indicated in the last two columns. A 500 L/ha field dose mixture was used to calculate the laboratory dose. When a dose range was given on the label, the higher dose was selected to calculate the laboratory dose.

* TL = Translaminar action, Stom = Stomach action, Cont = Contact action, Sys = Systemic action.

** IRAC = Insecticide Resistance Action Committee, https://www.irac-online.org. The group is used to alternate insecticides to prevent build-up of resistance.

*** Only indicated on the label that it would result in a reduction in *Liriomyza* leafminers.

UNE and UN: Unknown or uncertain.

Fig. 14. a: Spraying of infested plants with insecticides. **b**: After spraying, plants were isolated individually in buckets to recover any pupae that might have formed.

Survival of buried pupae

This objective was included to investigate a possible solution to eliminate leafminer pupae that were overwintering or oversummering in harvested fields, e.g. by deep ploughing. To establish how deep a leafminer pupa must be buried for it to die, experiments were conducted under laboratory conditions. A sandy and loamy-sandy soil were used to bury leafminer pupae in half-litre plastic containers. Twenty pupae were placed at depths of 1 cm, 2 cm, 3 cm, 4 cm, 8 cm and 16 cm. The control was pupae placed on the surface of the soil. An inverted glass Petri dish was placed on the open end of the containers to prevent the soils from drying out too quickly and to contain any emerging flies (Fig. 15). After four weeks, all containers were inspected, and the number of flies that eclosed from the pupae, and managed to reach the surface, were counted. All treatments were repeated three times.

Fig. 15. Illustrations of the soil types used (sandy and loamy-sandy), in the buried pupae experiments, as well as the containers in which the experiments were carried out. The containers for the 8 cm and 16 cm experiments were similar, but larger, and are not illustrated.

Chapter 3

RESULTS

Origin of the first leafminers of the season

2017

The results of the 2017 field trial are given in Fig. 16 and 17. Fig. 16 shows the first eight weeks with annotations regarding the percentage of leaves infested from week three to week six. The following deductions can be made:

- The first leaf mines appeared on the lower leaves at week three.
- The number of these leaf mines increased exponentially each week to reach 75 % infestation within three weeks.
- A sudden "explosion" in numbers was not observed – rather, a gradual but exponential increase was evident.
- The first leaf mines on the top leaves were observed by week five, most probably on the shorter growers.

Fig. 17 shows the number of adult flies on sticky traps during the entire 12 weeks that the field trial was conducted. The following assumptions can be made from Fig. 17:

- Leafminer fly numbers build up within fields.
- No evidence could be found of the presence of flies in areas adjacent to the field during the early season.
- No evidence could be found of flies migrating *en masse* from areas around potato fields.
- The increase in fly numbers within fields is correlated with the appearance and increase of leaf mines on the lower leaves in the same field.

Figures 16 and 17 demonstrate that leaf mines were present on the lower leaves even before the first flies were caught in the traps. This indicates that the fly numbers (at week 2) were so low that there were not enough flies to reach the traps. At week four, the first flies were trapped, only in the target field (where the leaf mines were noted the previous week) and in the old field (where nothing was growing). No flies were trapped in any of the other 11 traps around the field. This corresponds to Mugala (2021) findings, who found little to no flies in traps in natural vegetation around potato fields in the Sandveld region of the Western Cape. Fly numbers in the "old field" trap stayed low but consistent, while the numbers in the new target field increased exponentially up to week eight (Fig. 17). The graph shows that the only flies present up to week seven were in the old field and the new target field. The other increase visible at week eight was on the trap 25 m from the target field edge, in the direction of the old field; it probably intercepted flies that were moving from the old field to the new target field.

During weeks nine and ten, the new target field started to deteriorate due to early senescence and damage by the leafminer flies and *Alternaria* blight, similarly reported by Deadman *et al.* (2000). Flies started to appear on other traps around the target field as well, even on the traps 100 m away from the field (Fig. 17), which may indicate that the field was so saturated with flies (and in senescence) that they were forced to "migrate" outwards, looking for new host plants. Most of these flies presumably moved to the new field (position of the field indicated in Fig. 3), as indicated by the very high numbers of flies in its trap (brown bars in Fig. 17 at weeks nine, 11 and 12). The reason for the sudden decline in numbers in this field at week 10 is unknown.

2018

The first leafminers of the 2018 season were again caught in a previously harvested field (Fig. 18, area number two). From the second week after plant emergence, leafminers appeared in very low numbers in the new target field (Fig. 10 green bars) and trap number two (Fig. 10 orange arrows). No flies were caught in traps in any other area up to week nine. During week nine, flies were caught in a trap situated in a previously harvested field (Figs. 5 and 18, area number three). All the other previously harvested fields (areas 4, 5, 6 and 7) had limited to no leafminer infestations during 2017, and this may be why no flies were caught in these areas in 2018.

These findings were similar to those in 2017 and confirm that the origin of the first flies of the season will be from previously harvested fields. However, when comparing fly numbers on traps in the new target fields during 2017 and 2018, the timing and tempo with which the flies emerged and increased differed noticeably between the respective years.

Comparison of leafminer fly emergence, 2017 and 2018 (Figs. 17 and 18)

During the 2017 season, the first leaf mines were observed on the lower leaves as early as week three, and the first leafminer flies were encountered on traps at week four. In 2018, the first significant increase in fly numbers on traps was only during week seven, with the first leaf mines only found during week eight. A slow but exponential build-up of fly numbers on traps was noted during 2017, with a peak at week eight (Fig. 17). In 2018, this gradual build-up of numbers was not evident; instead, a sudden peak was found at week 11, with lower and inconsistent numbers in the preceding four weeks (Fig. 18). The first leaf mines on the top (upper) leaves were noted during week five of 2017 and only during week 11 of 2018. The onset of senescence during 2017 was two weeks earlier compared to 2018, a possible consequence of the early and heavy infestation of leafminers during 2017, compared to the mild infestations of 2018.

From the results, it is clear that the time, tempo and severity of leafminer infestations may differ dramatically from season to season. However, no matter if the peak of the infestation is early or late, it is clear that leafminer infestations originate from previously harvested fields and not from other cultivated areas or fallow areas around fields.

2019

Flies emerged in low numbers in previously harvested fields, from week two to week eight, before a spike in numbers in the newly planted fields during week nine (Fig. 19), which differs from the findings of the previous two years, and may be attributed to the late emergence of the plants (two months later). Only in one of the five previously harvested fields (“Harvested January 2019”) did no flies emerge. This was the only field that was harvested in summer, and all flies presumably emerged before the winter. Around week six, 350 mm rainfall was measured over eight days. This excessive rainfall did not seem to have a marked influence on the emergence of the leafminers during later weeks – a peak of more than 600 flies was found during week nine.

The marked increase in the numbers in the field harvested in July (Fig. 19; "Harvested July 2019 B"; purple bars) could have been attributed to volunteer plants emerging in this field during weeks eight and nine. This may be an important additional finding – the emergence of volunteers may equally contribute to infestations of new potato fields.

2020

Two new fields and two previously harvested fields were monitored. Flies started to appear in all traps from week four (Fig. 20). The numbers stayed low until week eight, when a marked increase in the two new fields was observed. Both fields showed the same trend – an exponential increase in numbers up to week 11, with a steady decline during weeks 12 and13. Flies continued to emerge in both the previously harvested fields up to week 11. The gradual and exponential increase in fly numbers during this year was similar to what was found during 2017.

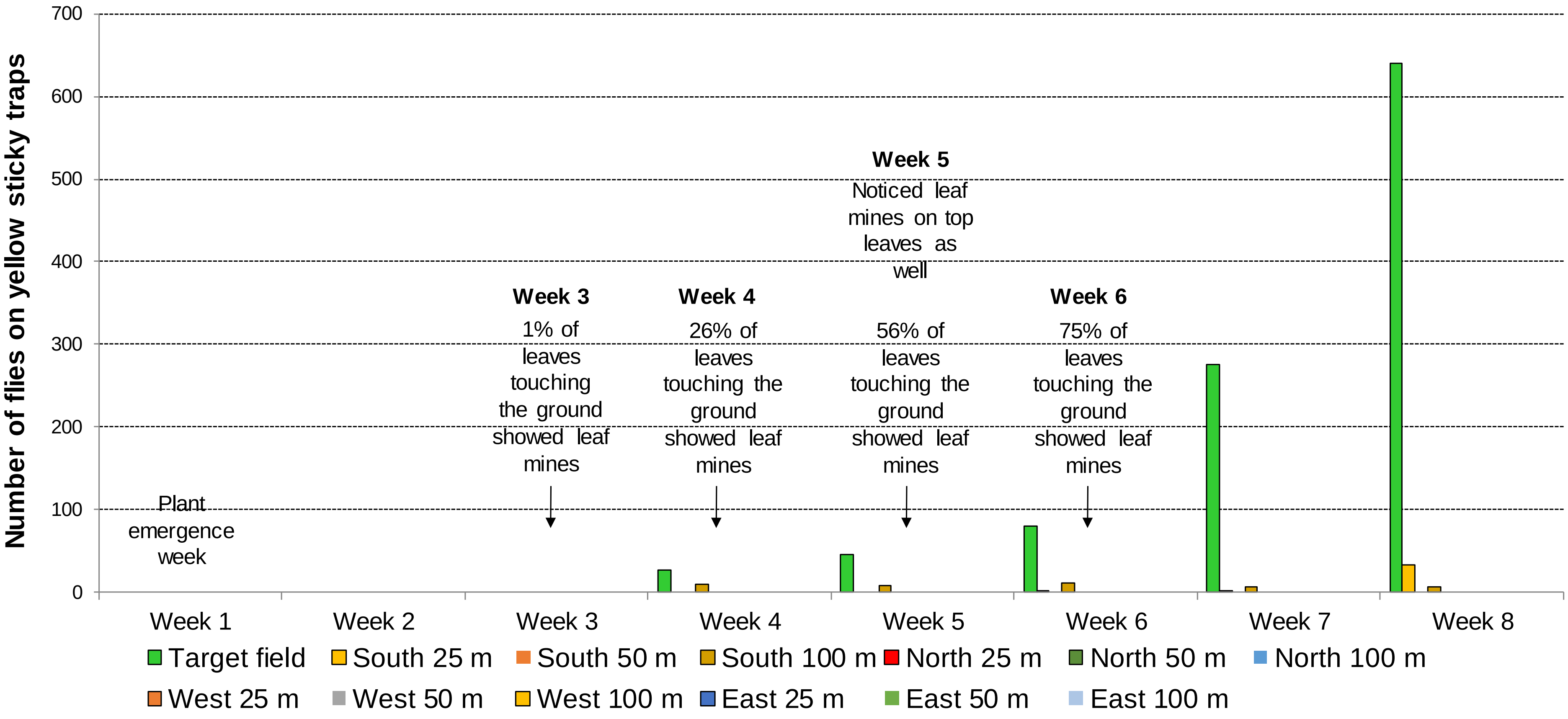

Fig. 16. The number of leafminer flies caught per week on yellow sticky traps in and around the new target field (see Fig. 5). Plant emergence (week 1) was 6 September 2017. Observations on the appearance and increase of leaf mines on the lower leaves are indicated in the text above the relevant weeks.

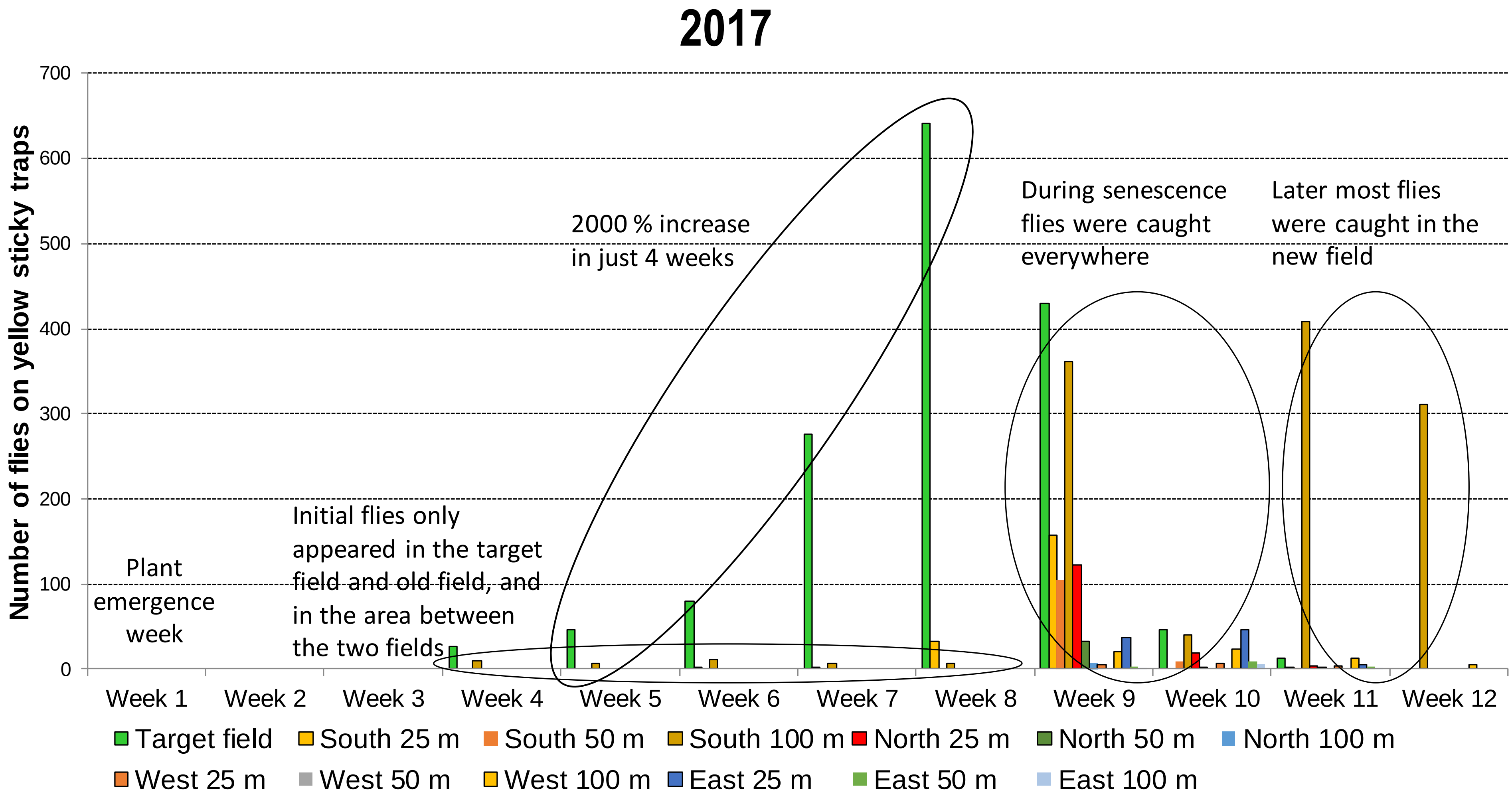

Fig. 17. The number of leafminer flies caught per week on yellow sticky traps in and around the new target field (see Fig. 5). Plant emergence (week 1) was 6 September 2017. Notes on the most important observations are indicated next to the ovals. The trial was terminated when the leafminers and Alternaria blight killed off the foliage in the target field.

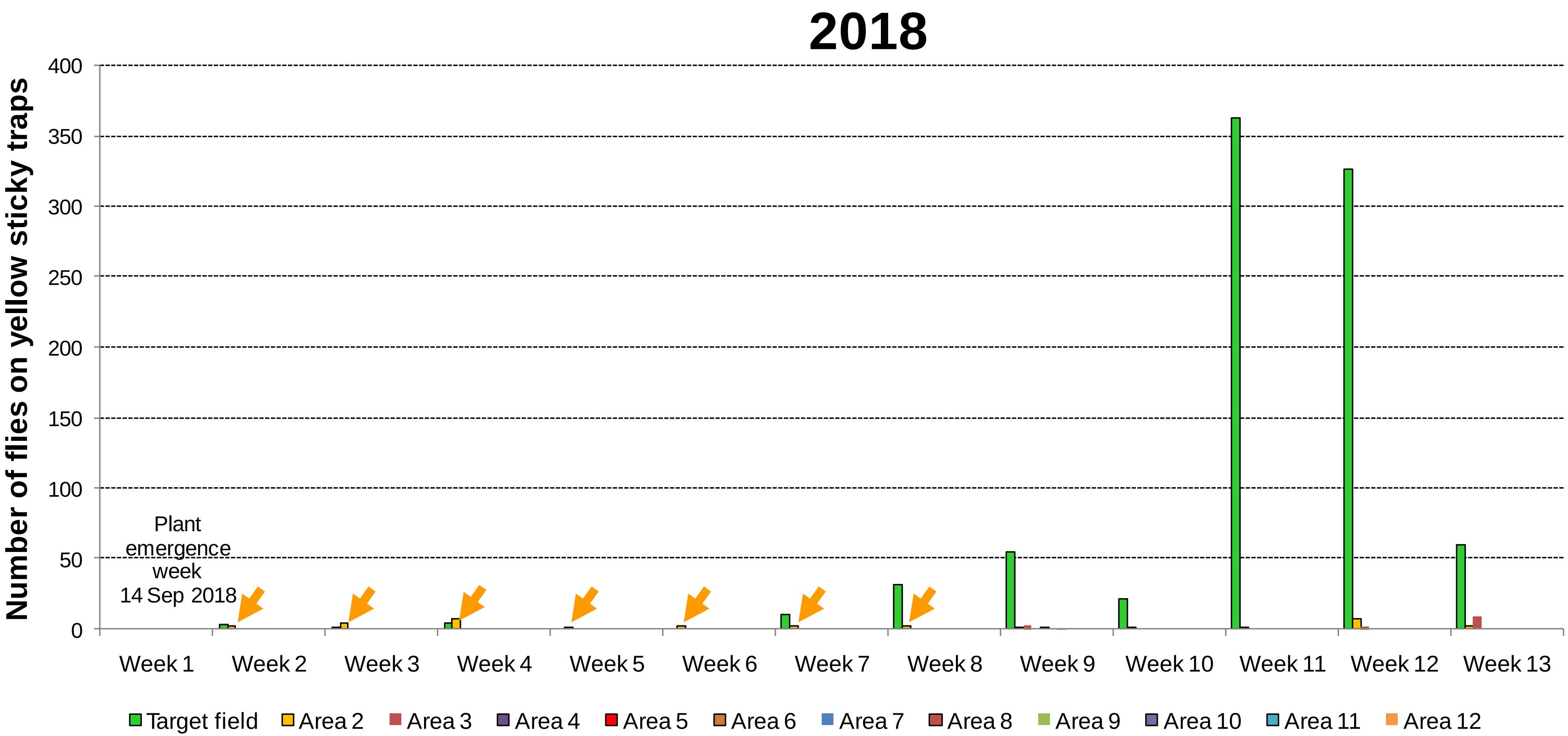

Fig. 18. The number of leafminer flies caught per week on yellow sticky traps in and around the new target field (green bars) of 2018 (see Fig. 7 for area locations). The orange arrows indicate the leafminer numbers in Area 2, i.e. the harvested field of the previous season. No leafminers were caught in any other area up to week 9.

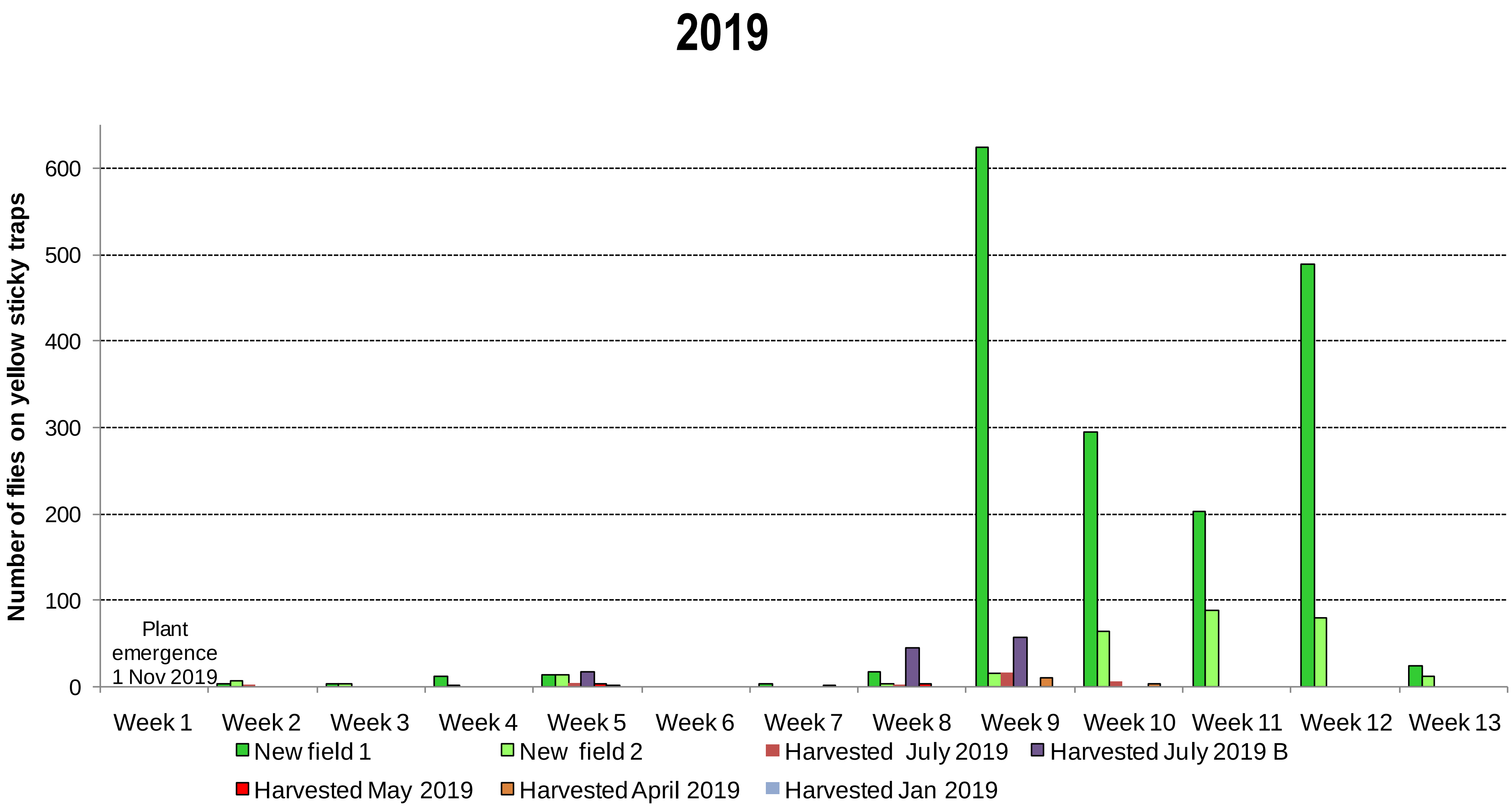

Fig. 19. The number of leafminer flies caught per week on yellow sticky traps in two newly planted fields and five previously harvested fields. During week 6, more than 350 mm rainfall was measured, and no monitoring was possible.

2020

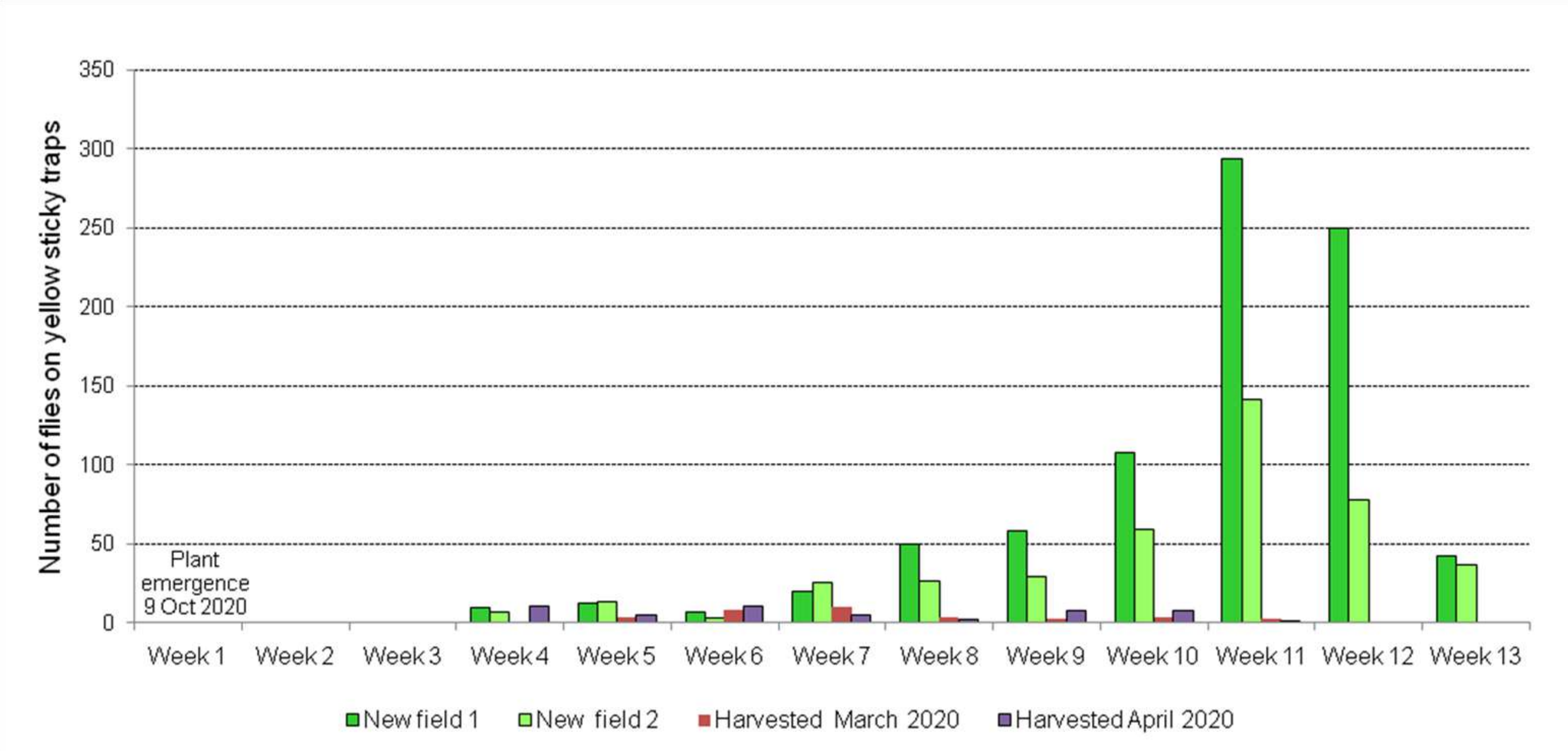

Fig. 20. The number of leafminer flies caught per week on yellow sticky traps in two newly planted fields and two previously harvested fields.

Investigation into traps for the potato leafminer

Sticky traps vs. water pan traps

Very few leafminer flies were trapped in the water pan traps, compared to the yellow sticky traps. Close observations revealed that the majority of flies landed on the sides of the yellow water buckets and that the few that were trapped in the water were most probably blown by the wind onto the water surface accidentally. However, any effort by the flies to land on the sticky taps resulted in entanglement in the sticky glue. Yellow sticky traps were therefore used during the study; this method was very effective in monitoring the number of flies in the vicinity of the traps at any given time.

Disadvantages of using yellow sticky traps

Although yellow sticky traps were found to be very effective and practical for monitoring the potato leafminer, we experienced several challenges during the four years of the study. Apart from the challenges relating to wind and animal damage, which were solved by altering the design of the trap setup (see the Methods section), we identified several other disadvantages that could not be solved with trap design. Several non-target organisms may be caught, sometimes in large numbers, including decomposers like blowflies (Fig. 21A), beneficial natural enemies like ladybirds (Fig. 21B), and even small birds (Fig. 21C). Fortunately, the presence of these non-target organisms in traps was not often encountered and may be seen as exceptions. A fourth challenge (or disadvantage) was the clogging of traps by wind-blown sand or mud splashes caused by rain or irrigation. Therefore after strong winds or heavy rains, traps need to be checked and replaced when needed.

The fruit fly attractant, Hym-lure

No leafminer flies were caught with any of the Hym-lure treatments. Because high numbers of flies were present in the field during the evaluations, it is concluded that Hym-lure is not a suitable agent for attracting potato leafminer flies. The fruit fly attractants, E.G.O. Phero Lure for *Ceratitis capita*, Chempac Fruit Fly Lure, and Fruit Fly Lure (names as appearing on the packaging of the lures), were evaluated for their potential to attract the potato leafminer. No flies were caught in any of the traps containing the attractants. The conclusion is, therefore, that the three products cannot be used to attract leafminer adults.

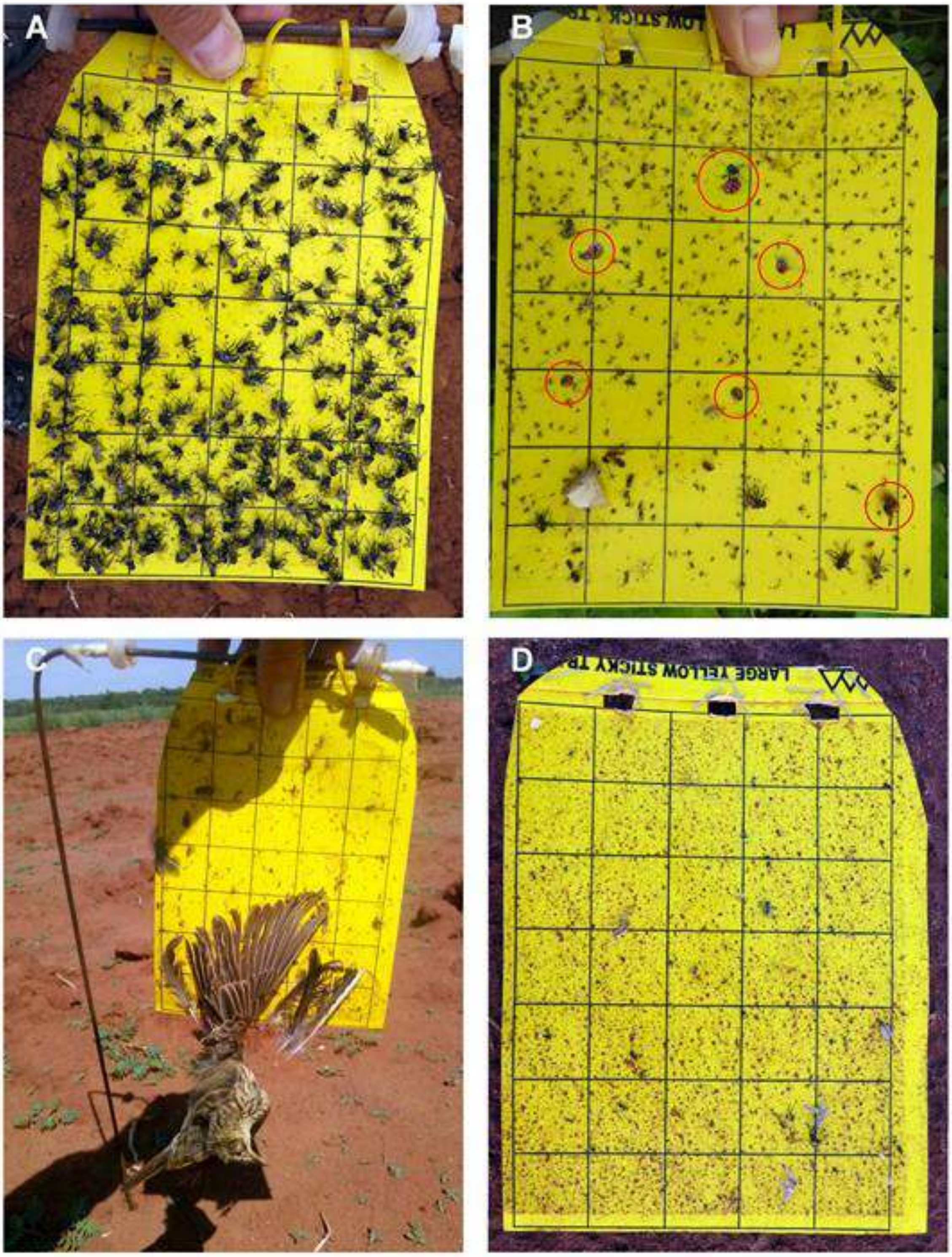

Fig. 21. The disadvantages of sticky traps: **A.** Other non-target flies may be attracted and caught in large numbers, **B.** Beneficial insects like ladybirds may be trapped (circled), **C.** small birds may be trapped, **D.** Wind-blown dust and mud slashes may clog traps.

Evaluation of registered insecticides

The results of the evaluations of potato leafminer pupae against insecticides can be found in Figs 22 and 23. Two important findings can be inferred from the results: Firstly, not one of the insecticides showed any significant effect on the pupae, and secondly, only about half of all pupae eclosed into adults, including those in the controls (Fig. 23, horizontal orange line).

The results of the evaluations of potato leafminer larvae in treated foliage can be found in Fig. 24. An average of approximately 30 pupae emerged per plant in the control treatment. The average number of pupae that emerged from the insecticidal treatments varied from three to six, which calculates to a control efficacy of approximately 70 % or higher for the 10 insecticides evaluated. The results, therefore, indicate that insecticides registered against the potato leafminer in South Africa are still effective and that poor control during some years may be attributed to other factors.

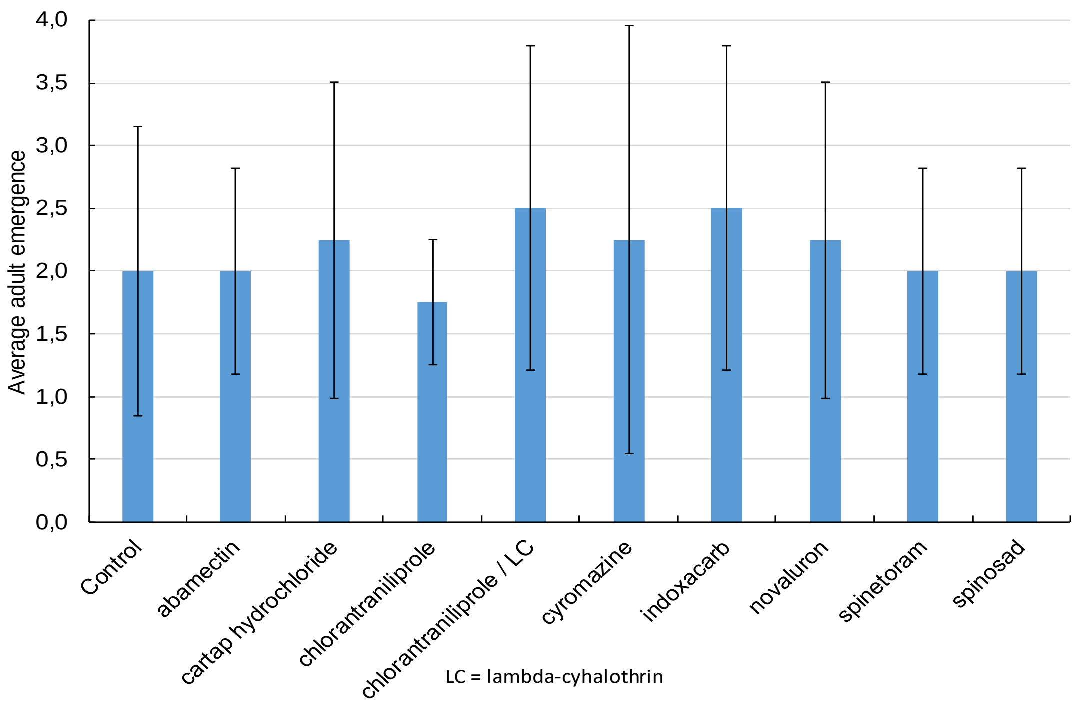

Fig. 22. The average number of adult leafminers emerging from treated pupae

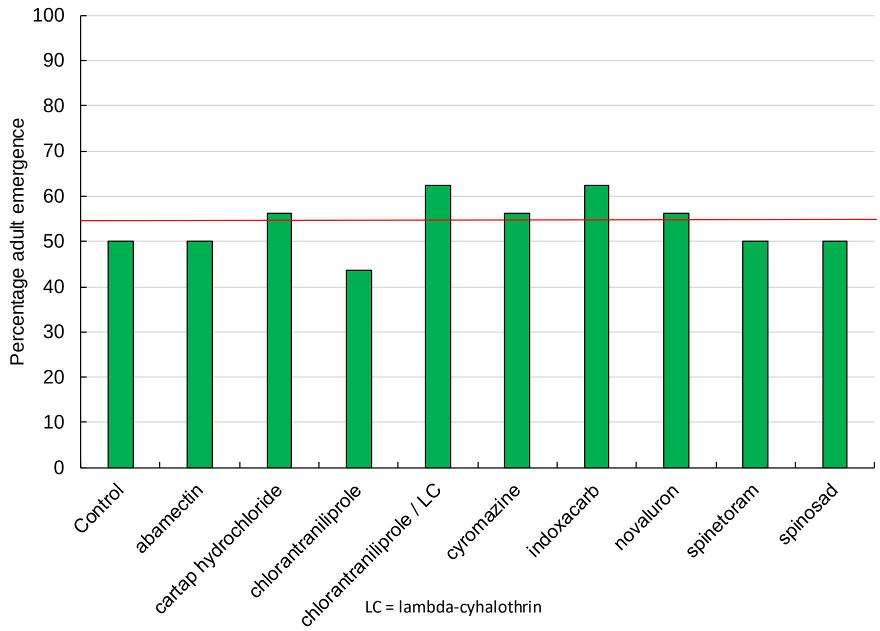

Fig. 23. The percentage of adult leafminers emerging from treated pupae (from Fig. 22). The average for all treatments is indicated with an orange horizontal bar.

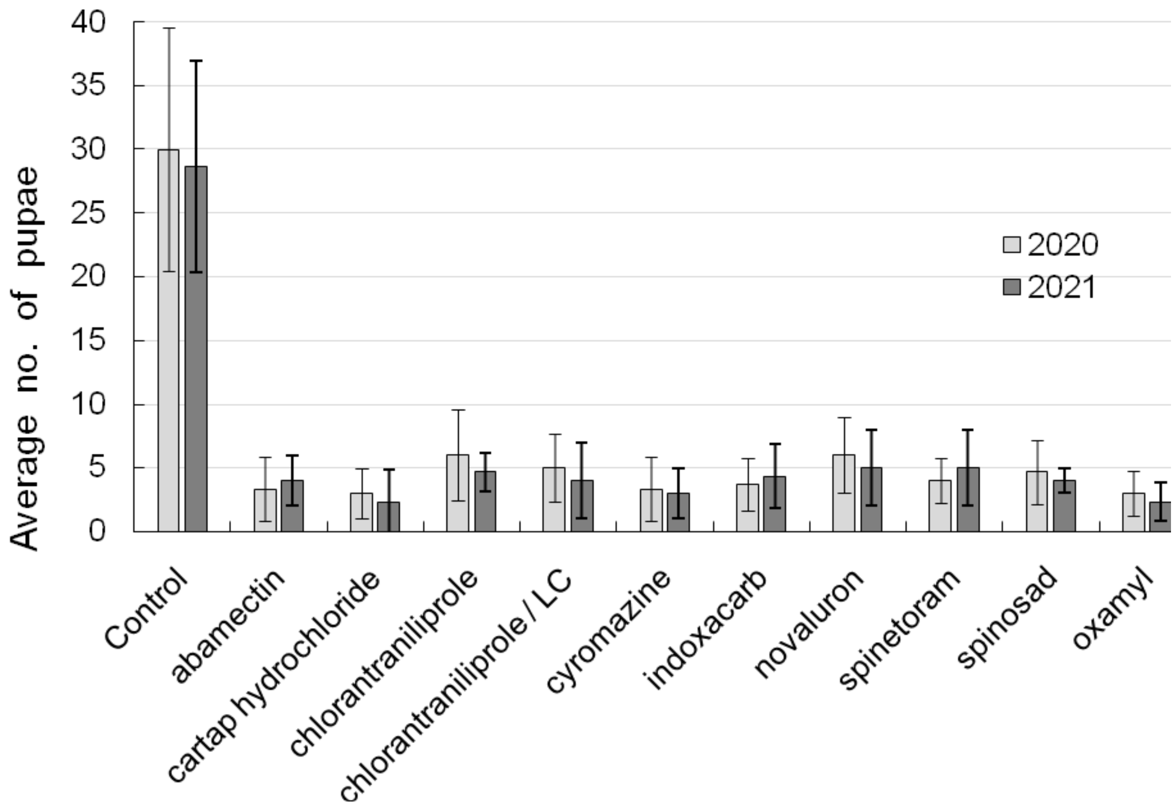

Fig. 24. The average number of pupae that emerged from tomato plants treated with the indicated insecticides. The control was a water treatment only. Abamectin was added with indoxacarb, as per label instruction.

Survival of buried pupae

The survival of pupae buried beneath a sandy or loamy-sandy soil, measured as the number of flies emerging above soil level, were as follows: 75 % of pupae buried at 1 cm survived, 40-55 % of pupae at depths of 2-4 cm survived, and 5-10 % survived when buried at depths of 8 cm (Figs 25 and 26). No flies emerged in experiments where pupae were buried at depths of 16 cm.

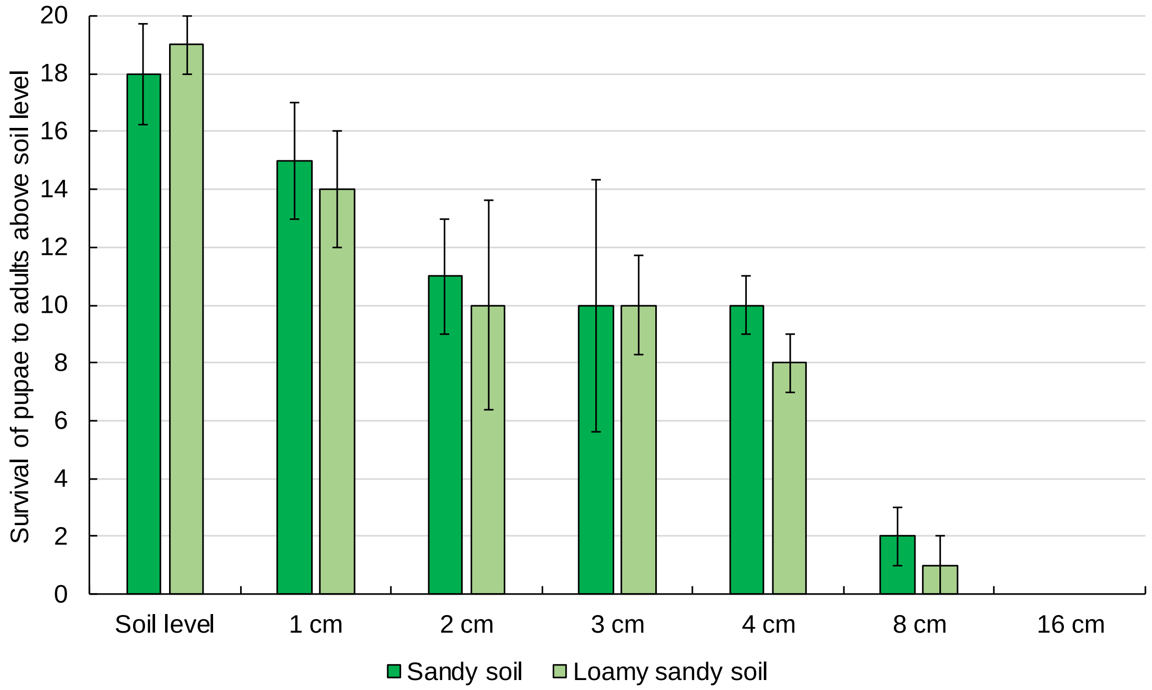

Fig. 25. The average number of flies emerged from two soil types, i.e., sandy soil and loamy-sandy soil after pupae were buried at depths up to 16 cm.

Sandy soil (95% sand)						
0 cm	1 cm	2 cm	3 cm	4 cm	8 cm	16 cm
90%	75%	55%	50%	50%	10%	0%

Loamy sandy soil (70% sand)						
0 cm	1 cm	2 cm	3 cm	4 cm	8 cm	16 cm
95%	70%	50%	50%	40%	5%	0%

Fig. 26. The percentage survival of flies, i.e. those that emerged from two soil types, i.e., sandy soil and loamy-sandy soil, after pupae were buried at depths up to 16 cm.

Chapter 4

DISCUSSION

After the introduction of the potato leafminer into South Africa in 2000, initial yield losses of up to 70 % were experienced by potato farmers, especially in the Sandveld production region. The problem subsided during the following years due to the registration of a few effective insecticides. However, leafminer infestations seemed to become serious again during some seasons, also in other regions of the country. It was clear that current and potential control strategies for leafminer control should be investigated.

Seventeen years after its introduction, the potato leafminer was ranked as “an always serious problem” in the majority of the potato production regions of South Africa (PSA, 2017). Although many new insecticides have been registered since 2000, farmers still experience challenges in suppressing fly numbers during most years. Because insecticidal resistance has not been proven against the potato leafminer in South Africa, a closer look into the ecology and alternative strategies were needed to assist potato farmers with control options against this important potato pest.

The major unsolved problem (or uncertainty) to date was that the origin of the first leafminer flies of the season was unknown. Infestation sources could not be identified and farmers just accepted that their fields would become infested at some point during the season. A phenomenon that worsened the scenario was that visible infestations (leaf mines) usually commenced unexpectedly during the second half of the season, although flies may have been present early in the season. These “delayed but sudden attacks” were studied by Videla and Valladares (2007), who found that the potato plant defends itself by “pushing” eggs and first instar larvae out of leaves, after which they die from exposure and desiccation. However, only the young and active-growing leaves that were able to accomplish this found links to our findings relating to the build-up of flies during the season.

The build-up of fly numbers during the four years of study always started after the middle of the season. The second half of the season usually correlates with tuber bulking and reduced growth of the above-ground parts. The foliage is not growing and expanding actively during this time, and the leafminer fly eggs and larvae start to overcome the plant’s defences – they are no longer pushed out by the leaves. More “stagnant” leaves are available for proper reproduction, and leafminer populations start to increase exponentially. Although flies were present in traps early in the season, they could only multiply at a slow rate on the relatively few, lower (older) leaves. Because leafminers multiply faster in potato fields during the second half of the season, short growers will comparatively show damage much earlier than medium to long growers. Care must therefore be taken when comparing cultivars, as long growers may seemingly be resistant to leafminers during the early season, but the damage to leaves is actually only “postponed” due to the long growth cycle. The question, however, was where the infestations came from that sustained the low numbers in new fields early in the season.

Our study showed that the source of leafminers in new fields early in the season was previously harvested fields. Similar to the findings of Mugala (2021), who found little to no leafminers in the natural vegetation surrounding potato fields, this study showed that leafminer flies were only recorded in previously harvested and new potato fields. These leafminers presumably originated from pupae that went into an “oversummer” or “overwinter” phase during the previous season. Potato leafminers pupate on leaf surfaces; the pupae are loosely attached to the leaf but nearly always fall to the ground soon afterwards. Therefore, most pupae are left in the field on the soil surface after harvest, and some may survive the winter until the next spring.

When volunteers are not controlled in the previously harvested fields, an added source may be the fly progeny that multiplies on these plants. Volunteer plants are often virus infested and stunted and may grow slowly due to the lack of water and fertilisers. Because such plants are usually not healthy and do not grow vigorously, they cannot defend themselves against leafminers. The conclusion of this study is, therefore, that pupae that emerge from soils in previously harvested fields, as well as flies that multiply on volunteer plants that may emerge in those fields, are the major source of infestations of newly planted potato fields.

Our study found that yellow sticky traps were effective in monitoring the adult leafminers in potato fields. Because flies occur randomly in fields (Muller & Kruger, 2008), it is not necessary to install many traps at different locations in large fields. Yellow water pan traps work well for pests like aphids, but the leafminer flies seemed to evade the water and landed on the edges of the water trap. Pheromones are not currently available for *Liriomyza* flies, and pheromone traps are therefore not an option to monitor their activity in potato fields. We evaluated several other attractants that are effective for fruit fly monitoring, but none were found to attract the potato leafminer adults.

Although yellow sticky traps are very effective in monitoring the potato leafminer in open fields, it must be kept in mind that they are not always environmentally friendly. Depending on the situation and non-target organisms present where the traps are used, many unintended captures may result (Chavez & Raman, 1987; Lu, Bei & Zhang, 2012). We often captured beneficial ladybirds and decomposer flies and, on two occasions, even small birds. Many more small non-target insects that are attracted to the yellow colour of the traps may be captured. However, because yellow sticky traps usually only attract insects in their vicinity and in line of sight, the impact on the entire ecosystem will be negligible.

Control of the potato leafminer in South Africa is mainly through insecticides. Three scenarios were found in a literature search for control options used against the leafminer in other countries. Some countries reported that the leafminer was not significantly important, while those that do experience serious damage mostly use insecticides. The third scenario was that the leafminer was only troublesome in crops in greenhouses, where they are usually controlled by augmentative releases of parasitoids (Weintraub *et al.,* 2017). Countries that reported that the leafminer was only a minor pest included Argentina (certain areas), Canada, some European countries, Israel, China, and Indonesia. The pest is currently absent from Australia and New Zealand (Weintraub *et al.,* 2017) and other control actions reported but with varying success include mass trapping with yellow sticky traps. This option is not practical on a large scale but may reduce adult fly numbers in smaller plots.

Twelve insecticides (active ingredients) are registered for use against leafminer flies on potatoes in South Africa. Although resistance is suspected most of the time when control is inadequate, our laboratory studies could not identify an active ingredient that shows significant lower control in relation to the untreated control treatment. Only translaminar and/or systemic larvicides can be used to manage the potato leafminer (Weintraub *et al.* 2017, and references therein). Registration trials are also based on larval mortalities in the field and not against the adults. Therefore, although some of the insecticides may have additional killing action against the adults, we evaluated them against the larvae (as per label). Therefore, our preliminary conclusions are that insecticides registered against the potato leafminer in South Africa are still effective, and variation in the control under field conditions, as experienced by farmers during some seasons, may be attributed to other factors. As shown in this study, one such factor may be the continuous augmentation of flies originating from pupae in previously harvested fields. We investigated the ecology and possible control options to eliminate the pupae in harvested fields to address this potential infestation source.

The pupae of the potato leafminer are nearly always found on the soil surface beneath plants. When the canopy of the potato plant closes naturally, insecticides mostly do not reach the soil surface due to the obstruction by the foliage. The pupae are therefore relatively well protected against field-applied insecticides. However, after haulm kill or after harvest, a huge number of exposed pupae may be found on bare ground or just below the soil surface.

To investigate if exposed pupae, e.g. those on the ground after haulm kill or in harvested fields, are vulnerable to insecticides, tests were conducted to evaluate the efficacy of "foliage"-registered insecticides in the laboratory. Our tests showed no effect against pupae in relation to the untreated controls. Fly pupae are formed inside a strong puparium that acts as an outer layer to protect the pupa (Fig. 2). Insecticides may therefore find it difficult to penetrate the puparium unless an affective penetrant is available. Most insecticides already contain wetters and penetrants as part of their formulations to penetrate foliage. Therefore, agro-chemical companies should investigate the potential of penetrants and formulations that may penetrate the leafminer puparium. If such a penetrant can be found, it may be used in formulations or as an additive to current insecticides. It may lead to new effective control agents against potato leafminer pupae in harvested fields.

Only 55 % of all the pupae used in the bioassays with insecticides eclosed into adult flies. In bioassays, when the control treatments also show low eclosal rates (as found in our experiment), it usually means poor natural survival and may make the bioassay results less reliable. However, there may be an important link between this "unexpected low survival" of the pupae in the controls and the rest of the larger study. Further investigations revealed that the pupae that failed to eclose in our study were not dead but dormant, which may indicate that the natural ecology of the potato leafminer fly is "regulated" in such a way that only a proportion of the pupae eclose at any given time. In our laboratory population, this was only 55 %. The rest of the population may eclose at a later stage, and according to our observations, also in harvested fields, even a few months later.

This preliminary finding of "delayed eclosion" of leafminer pupae may now conveniently link to the next study objective, i.e. can dormant pupae in harvested fields be killed by deep ploughing after harvest? Laboratory studies were conducted with sandy and sandy loam soil to investigate if pupae in harvested fields could be controlled by burying them under a layer of soil. Very few leafminer flies emerged from a depth of 8 cm, and no flies emerged from a depth of 16 cm. Therefore, it can safely be assumed that when pupae are buried at depths of 16 cm and deeper, no flies will survive. This can therefore be used as a cultural practice by farmers to eliminate dormant pupae in harvested fields.

The findings of various studies, e.g. Weintraub & Horowitz (1995), and references in Weintraub *et al.* (2017), suggest that the potato leafminer is a more serious problem in cooler areas. These studies showed that the potato leafminer is never a problem during warm summers in most countries, although they may be a serious pest during the autumn and spring months in the same countries. The only potato production region in South Africa that reported the potato leafminer as a minor pest, i.e. the Eastern Free State, is mainly a dryland region. It is, therefore, possible that there may be a link between continuous high temperatures in non-irrigated fields and lower leafminer survival and pest status. According to Weintraub *et al*. (2017), the potato leafminer reproduces the fastest at temperatures below 25°C. Most potato production regions in South Africa experience much higher temperatures but still report that the leafminer is a serious problem. Other environmental and production factors may also play critical roles in determining the pest status of the potato leafminer. One such factor that has not been investigated before is how overhead irrigation, especially the fine mist irrigation by a typical centre pivot, may play in cooling the micro-ecosystem down to favour the potato leafminer.

From this study, we identified three possible aspects for further research. Firstly, we recommend that the elimination of dormant pupae in harvested fields be investigated on a larger scale, e.g., using tillage equipment to bury and kill the pupae. Secondly, we recommend studying the ecology of the potato leafminer, especially the fly's possible ability to "postpone" eclosion, needing further infestation. If it can be established under what conditions the pupae are more likely to eclose and how long they may take to eclose under different conditions, new strategies may be implemented to avoid the main emergence cycle of the new season. And lastly, we recommend investigating the effect of different irrigation regimes on the ecology, and therefore pest status, of the potato leafminer.

Chapter 5

CONCLUSIONS AND RECOMMENDATIONS

After considering all the findings in this study, the following main conclusions can be reached regarding the occurrence, ecology, and origin of the first flies and control of the potato leafminer in potato fields in South Africa.

- Leafminer flies and the damage caused by the leafminers mostly occur in low numbers in new fields and exponentially increase in numbers during the second half of the season.
- Comparatively, short growers will therefore show damage much earlier than the medium and long growers, giving the impression that short growers are more susceptible to leafminer attacks.
- When large numbers of flies are present on foliage early in the season, a major infestation source may be present nearby.
- Previously harvested potato fields, even those harvested before winter, are a major source of new infestations of the new season.
- Yellow sticky traps, modified to withstand damage by animals and strong wind, may effectively be used to monitor leafminer adults in potato fields.
- Currently, registered insecticides are still effective, as evaluated under laboratory conditions.
- The insecticides that are registered for field conditions are not effective in killing leafminer pupae.
- When leafminer pupae are buried under a minimum of 16 cm soil, they will not succeed in reaching the soil surface and will therefore be killed.

The following recommendations are made regarding the findings and conclusions of this study. Some of these recommendations are preliminary and may need more detailed or on-farm studies to verify their feasibility.

- Potato producers must take cognisance of, and be aware that, infestation symptoms (leaf mines) appear on the upper leaves mostly during the second half of the season, even though the flies may be active, and leaf mines present on the lower leaves, early in the season.
- Regular scouting during the early season, focusing on the lower leaves in potato fields, and proactive control to prevent population explosions during the later season is recommended.
- When heavy infestations are experienced in a potato field, producers may deep-plough the harvested field in an attempt to bury and kill the overwintering or oversummering (dormant) pupae.
- Commercial yellow sticky traps can be used when monitoring is needed, modified to rotate in both the horizontal and vertical axes.
- Registered insecticides are still effective and must be used according to label instructions.

This research project focused on the control strategies of the potato leafminer on potatoes in South Africa. It was shown that the use of insecticides is still an effective option but that alternative strategies are also available to enhance control during years of high infestations. In particular, the origin of the first leafminers was identified. This information broadened and enhanced our knowledge about the ecology and epidemiology of one of the more serious pests of potato in South Africa. Other aspects that could be investigated in the future include up-scaling the laboratory experiments to test the hypothesis that pupae can be eliminated by deep ploughing after harvest; investigations into the development of formulations that may enhance insecticides to be able to kill pupae (with the chemical companies); further studies into more attractants and traps to monitor, or attract-and-kill, potato leafminers in fields; and the role that different irrigation regimes may play in the ecology, and therefore pest status, of the potato leafminer.

Scientific publications that emanated from this study were or linked to the author of this report through collaboration, include Weintraub *et al.* (2017) (D Visser, co-author); Mugala et al. (2021a) (D Visser co-author); Mugala et al. (2021b) (D Visser co-author); Mugala *et al.* (2021c) (D Visser, co-author). The latter three publications are "in press" and collaborated with Stellenbosch University relating to postgraduate studies in the Sandveld. Three oral presentations were presented at congresses and symposia; another two oral presentations were presented at lecture seminars at the University of Pretoria.

REFERENCES

Atakan, E. & Canhilal, R. 2004. Evaluation of yellow sticky traps at various heights for monitoring cotton insect pests. *Journal of Agricultural and Urban Entomology,* 21: 15-24.

CABI. 2019. Invasive species compendium: *Liriomyza trifolii*, American serpentine leafminer. Available online at; https://www.cabi.org/isc/datasheet/30965

Chabi-Olaye, A.; Mujica, N. Löhr, B. & Kroschel, J. 2008. Role of agroecosystems in the abundance and diversity of *Liriomyza* leaf mining flies and their natural enemies. *Abstracts of the XXIII International Congress of Entomology*, 6-12 July 2008, Durban, South Africa.

Capinera, J.L. 2001. *Handbook of vegetable pests*. New York, NY: Academic Press.

Chang, P.E.C. & Metz, M.A. 2021. Classification of *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae: Gelechiinae: Gnorimoschemini) Based on cladistic analysis of morphology. *Proceedings of the Entomological Society of Washington,* 123(1): 41-54.

Chavez, G.L. & Raman, K.V. 1987. Evaluation of trapping and trap types to reduce damage to potatoes by the leaf miner, *Liriomyza huidobrensis* (Diptera, Agromyzidae). *Insect Science and its Application,* 8(3): 369-372.

Chen, B., & Kang, L. 2004. Variation in cold hardiness of *Liriomyza huidobrensis* (Diptera: Agromyzidae) along latitudinal gradients. *Environmental Entomology,* 33: 155-164.

Croplife South Africa. 2016. *Agricultural remedies database – insecticides.* Available online at: www.croplifesa.co.za (Accessed on 1 October 2016).

Dara, S.K. 2019. The new integrated pest management paradigm for the modern age. *Journal of Integrated Pest Management,* 10(1): 1–9.

Deadman, M.; Khan, I.; Thacker, J. & Habsi, K.A. 2000. Interaction between leaf miner damage and leaf necrosis caused by *Alternaria alternate*, on potato in the Sultanate of Oman. *Plant Pathology Journal,* 18: 210-215.

Ewell, P.T.; Fuglie, K.O. & Raman, K.V. 1994. Farmers' perspectives on potato pest management in developing countries: Interdisciplinary research at the International Potato Center (CIP). In: G.W. Zehnder; M.L. Powelson; R.K. Jansson & K.V. Raman (Eds). *Advances in Potato Pest Biology and Management*. St. Paul, MN: APS Press, pp. 597-615.

Fenoglio, M.S. & Salvo, A. 2009. *Liriomyza commelinae* (Diptera: Agromyzidae): An alternative host for parasitoids of the leaf miner pest *Liriomyza huidobrensis*. *International Journal of Pest Management,* 55(4): 299-305.

Foba, C.N.; Salifu, Z.O.; Lagat, L.M.; Gitonga, K.S. & Fiaboe, K.K.M. 2015. Species composition, distribution and seasonal abundance of *Liriomyza* leafminers (Diptera: Agromyzidae) under different vegetable production systems and agroecological zones in Kenya. *Environmental Entomology,* 44: 223-232.

Lanzoni, A.; Bazzocchi, G.; Burgio, G. & Fiacconi, M.R. 2002. Comparative life history of *Liriomyza trifolii* and *Liriomyza huidobrensis* (Diptera: Agromyzidae) on beans: Effect of temperature on development. *Environmental Entomology,* 31: 797-803.

López, R.D.; Carmona, D.; Vincini, A.M.; Monterubbianesi, G. & Caldiz, D. 2010. Population dynamic and damage caused by the leafminer *Liriomyza huidobrensis* (Blanchard: Agromyzidae) on seven potato processing varieties grown in temperate environment. *Neotropical Entomology,* 39: 108-114.

Liu, T., Kang, I., Heinz, K. & Trumble, J. 2009. Biological control of *Liriomyza* leafminers: Progress and perspective. *Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources,* 4(4): 1-16.

Lu, Y.; Bei, Y. & Zhang, J. 2012. Are yellow sticky traps an effective method for control of sweetpotato whitefly, *Bemisia tabaci*, in the greenhouse or field? *Journal of Insect Science,* 12(1):1-13.

McCravy, K.W. 2018. A review of sampling and monitoring methods for beneficial arthropods in agroecosystems. *Insects,* 9(4): 1-27.

Mugala, T. 2021. *Ecology and biocontrol options of the leaf miner Liriomyza huidobrensis, on potatoes in the Western Cape Province*. MSc Thesis, Stellenbosch University, Stellenbosch.

Mugala, T.; Visser, D.; Malan, A.P. & Addison, P. 2021a. Occurrence of the potato leaf miner, *Liriomyza huidobrensis* (Diptera: Agromyzidae), and parasitoids in potato fields and natural vegetation of the Western Cape Province, South Africa. *African Entomology* (submitted).

Mugala, T.; Visser, D.; Malan, A.P. & Addison, P. 2021b. Isolation and identification of biologicals from South African potato soil and their pathogenicity against the potato leaf miner, Liriomyza huidobrensis (Diptera: Agromyzidae). *African Entomology* (submitted)

Mugala, T.; Visser, D.; Malan, A.P. & Addison, P. 2021c. Review of *Liriomyza huidobrensis* (Diptera: Agromyzidae) on potatoes in South Africa, with special reference to biological control using entomopathogens and parasitoids. *African Entomology* (submitted).

Mujica, N. & Kroschel, J. 2011. Leafminer fly (Diptera: Agromyzidae) occurrence, distribution, and parasitoid associations in field and vegetable crops along the Peruvian coast. *Environmental Entomology,* 40: 217-230.

Mujica, N. & Kroschel, J. 2013. Pest intensity-crop loss relationships for the leafminer fly *Liriomyza huidobrensis* (Blanchard) in different potato (*Solanum tuberosum* L.) varieties. *Crop Protection,* 47: 6-16.

Mujica, N. 2016a. Ecological approaches to manage the leafminer fly *Liriomyza huidobrensis* (Blanchard) in potato-based agroecosystems of Peru. In: J. Kroschel (Ed.) *Tropical Agriculture 21: Advances in Crop Research*. Weikersheim, Germany: Margraf Verlag, p. 11.

Mujica, N. 2016b. Population dynamics of the leafminer fly *Liriomyza huidobrensis* (Blanchard) and its associated parasitoids in two contrasting Peruvian potato agroecologies. In: J. Kroschel (Ed.). *Ecological approaches to manage the leafminer fly Liriomyza huidobrensis (Blanchard) in potato-based agroecosystems of Peru.* Weikersheim, Germany: Margraf Verlag, pp. 13-25.

Muller, L., & Kruger, K. 2008. Landing patterns of the serpentine leafminer *Liriomyza huidobrensis* (Blanchard) (Diptera: Agromyzidae) in potato fields. *Potatoes South Africa, Annual General Meeting of Potato Work Groups,* September 2008. Golden Gate, South Africa, p. 8.

Musundire, R.A.; Chabi-Olaye, B. & Kruger, K. 2011. Diversity of Agromyzidae and associated hymenopteran parasitoid species in the Afrotropical region: Implications for biological control. *BioControl,* 56: 1-9.

Myburgh, A.C. 1988. *Crop pests in Southern Africa – Potatoes and other vegetables*. Silverton: Promedia Publications.

Parrella, J. 1987. Biology of *Liriomyza*. *Annual Review of Entomology,* 32: 201-204.

Prinsloo, G.L. & Uys, V.M. 2015. *Insects of cultivated plants and natural pastures in Southern Africa*. Hatfield: Entomological Society of Southern Africa.

PSA (Potatoes South Africa). 2017. *Descriptions of 16 potato pests in South Africa*. Available online at: https://www.potatoes.co.za/research/research-introduction/ (Fact sheets – English fact sheets).

Rauf, A.; Shepard, B.M. & Johnson, M.W. 2000. Leaf miners in vegetables, ornamental plants and weeds in Indonesia: Surveys of host plants, species composition and parasitoids. *International Journal of Pest Management,* 46(4): 257-266.

Scheffer, S.; Wijeskera, A.; Visser, D. & Hallett, R.H. 2001. Polymerase chain reaction-restriction fragment-length polymorphism method to distinguish *Liriomyza huidobrensis* from *L. langei* (Diptera: Agromyzidae), applied to three recent leafminer invasion. *Journal of Economic Entomology,* 94(5): 1177-1182.

Van der Linden, A. 1993. Overwintering of *Liriomyza bryoniae* and *Liriomyza huidobrensis* (Diptera: Aromyzidae) in the Netherlands. *Proceedings of the Section Experimental and Applied Entomology of the Netherlands Entomological Society,* 4: 145-150.

Videla, M. & Valladares, G. 2007. Induced resistance against leafminer eggs by extrusion in young potato plants. *International Journal of Pest Management,* 53: 259-262.

Visser, D. 2001. A new devastating pest, *Liriomyza huidobrensis*, on potatoes in South Africa. *Proceedings of the 13th Entomological Congress of the Entomological Society of Southern Africa*, 2-5 July, Pietermaritzburg, South Africa.

Visser, D. 2002. Insecticides for controlling the potato leafminer. *Chips,* 16(6): 38-39.

Visser, D. 2005. *Guide to potato pests and their natural enemies in South Africa*. Pretoria: ARC-Roodeplaat, p. 105.

Visser, D. 2009. *A complete guide to vegetable pests in South Africa*. Pretoria: ARC-Roodeplaat, Vegetable and Ornamental Plant Institute, p. 316.

Visser, D. 2011. Potato leafminer – number one pest on potatoes. *Afgriland*, 55(1) Jan/Feb.

Visser, D. & Barnard, S. 1992. Is die Amerikaanse blaarmyner nou ook 'n plaag op aartappels? *Roodeplaat Bulletin,* 33: 6-7.

Visser, D. & Weintraub, P. 2001. Outbreak of the potato leafminer – comparisons between Israel and South Africa. *Chips,* March/April: 38-39.

Weintraub, P.G. & Horowitz, A.R. 1995. The newest leafminer pest in Israel, *Liriomyza huidobrensis*. *Phytoparasitica,* 23: 177-184.

Weintraub, P.G. & Horowitz, A.R. 1996. Spatial and diel activity of the pea leafminer (Diptera: Agromyzidae) in potatoes, *Solanum tuberosum*. *Environmental Entomology,* 25: 722-726.

Weintraub, P.G.; Scheffer, S.; Visser, D.; Valladares, G.; Correa, A.S.; Shepard, B.M.; Rauf, A.; Murphy, S.T.; Mujica, N.; MacVean, C.; Kroschel, J.; Kishinevsky, M.; Joshi, R.C.; Johansen, N.S.; Hallett, R.H.; Civelek, H.S.; Chen, B. & Metzler, H.B. 2017. The invasive *Liriomyza huidobrensis* (Diptera: Agromyzidae): Understanding its pest status and management globally. *Journal of Insect Science,* 17(1): 28: 1-27.