Cesar Australia

Monitoring to support integrated pest management of *Liriomyza* spp. pests in Australia

A mini-review of global monitoring plans

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Background

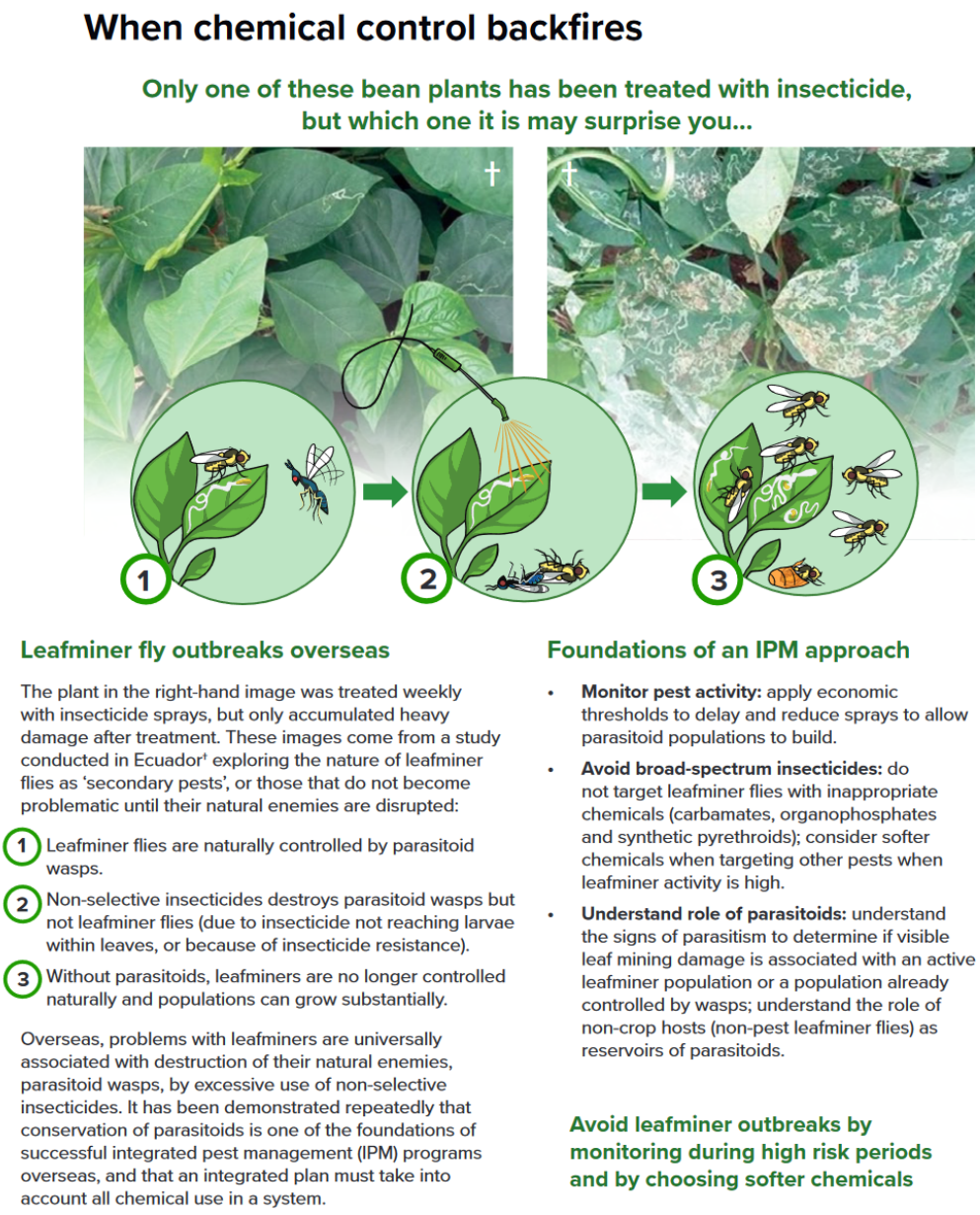
Three species of "leafminer flies" which have long been on the Australian federal government's 40 'high risk' biosecurity species, finally established in Australia between 2015 and 2020. They include the vegetable leafminer (VLM, *Liriomyza sativae*), the American serpentine leafminer (ASLM, *Liriomyza trifolii*) and the serpentine leafminer (SLM, *Liriomyza huidobrensis*). In 2008, VLM was detected for the first time throughout the north Australian islands of Torres Strait, and then on the Australian mainland at Seisia in 2015 (IPCC 2017). The pest has not yet been detected in any other regions of Australia despite ongoing surveillance efforts. Then in late 2020, SLM was detected in the Sydney region and eradication was subsequently deemed unfeasible (IPCC 2021a). Early the next year, ASLM was detected in northern Western Australia and within the Torres Strait, and final considerations on technical feasibility of eradication are still underway (IPCC 2021b), but eradication is unlikely.

> Read more about the recent SLM incursion here: https://cesaraustralia.com/pestfacts/serpentine-leafminer-detected-in-australia/

Referred to generally as the polyphagous *Liriomyza* leafminer, these flies are part of a well-known group (family Agromyzidae) of small, morphologically similar flies whose larvae feed internally on plants, often as leaf and stem miners. The majority of damage caused by polyphagous *Liriomyza* leafminer occurs during larval feeding between the upper and lower leaf surface, which curtails photosynthetic ability and reduces marketability of some crops.

Managing polyphagous leafminer

Global experiences support the notion that polyphagous Liriomyza leafminer are secondary pests, only reaching damaging levels after severe reductions in parasitoid populations. Polyphagous Liriomyza leafminer are also prone to evolving insecticide resistance, making control and eradication difficult. The most effective natural control of these pests comes from parasitoid wasps, but insecticide-based control disrupts beneficial predators and parasitoids, leading to secondary outbreaks.



The foundations of integrated pest management for exotic polyphagous *Liriomyza*. Image source: Chirinos, DT., Castro, R., and Garces, A. (2017). Read more about leafminer management here: https://ausveg.com.au/app/uploads/2020/12/Management-Plan-Exotic-leafminers.pdf

Monitoring is a cornerstone of a successful IPM approach to managing the polyphagous *Liriomyza* leafminer. As reviewed in Ridland et al (2020): "Successful field programs to manage a spectrum of insect pests including *L. sativae* and *L. trifolii* have been implemented for tomato and celery in California (Johnson et al. 1980a, 1980b, 1980c; Trumble 1985; Reitz et al. 1999), watermelon in Hawaii (Johnson 1987, 2005; Johnson et al. 1989) and melon and lettuce in Arizona (Palumbo & Kerns 1998;

Palumbo & Castle 2009).The foundations of these programs are to (1) reduce initial leafminer pressure by using uninfested transplants, destroying weeds and deep ploughing of senescent crops and avoiding planting new crops adjacent to old crops (Capinera 2017) and (2) conserve parasitoid wasps by avoiding broad-spectrum insecticides (Johnson et al. 1980b; Trumble & Toscano 1983) and using economic thresholds to delay and reduce sprays to allow colonising parasitoid populations to build up".

Monitoring goals as part of IPM programs may include:

1. Detecting early infestations, particularly in young crops or high value, zero-tolerance crops for which leaf mine damage reduces marketability, such as ornamentals, lettuce and celery;
2. Estimating population density in larger infestations in fruiting field crops, such as tomato and potato, in order to apply economic thresholds to chemical applications and to monitor the success of interventions (we focus here for the rest of this article). Sampling techniques aimed at estimating population density to support the use of ETs include:
    a. Counts of infested leaves
    b. Counts of live *Liriomyza* larvae within leaf mines (aided by a hand lens)
    c. Counts of *Liriomyza* pupae (caught in 'pupal trays' or rearing bags)
    d. Counts of *Liriomyza* adults on yellow sticky traps

Each technique has benefits/drawbacks for each of the monitoring goals discussed listed above and can be used in combination to effectively monitor populations of *Liriomyza* spp. pests in Australia.

Counts of infested leaves

Searching for leaf mines present on leaves is the simplest way to gauge the presence and activity of leafminer flies and some sampling plans have been developed that rely on count of leaf mines, without further confirmation of the presence of living larvae (which often requires a hand lens) (Burgio et al., 2005). These plans are usually based on counting the number of leaves bearing leaf mines (see Figure 1) in a subset of leaves on a subset of randomly selected plants.

However, a confounding factor for these plans is that the detection of mines does not always indicate active populations of flies (particularly in longer lifespan fruiting crops), as the mines persist on the leaf long after the emergence of the fly larva. Visual damage alone can be difficult to relate to active population size, as a result of the accumulation of older damage through time and the difficulty of detecting live larvae inside mines (Heinz & Chaney, 1995). In a worst-case scenario, inflated estimates of active population sizes may influence growers to spray unnecessary chemicals onto crops where leafminer populations have already collapsed, due to environmental factors or the influence of beneficial insects. In these cases, more harm

is done than good if beneficial parasitoids are destroyed, allowing the pest population to flourish once again (Ridland et al., 2020).

Pro: easy to see leaf mines and stippling damage without a hand lens

Con: can overestimate population activity and encourage inappropriate interventions

In Summary: Preferred when the goal is to detect early infestations or to monitor infestations in short lifespan crops, but may be inappropriate for monitoring infestation in long lifespan crops, or for monitoring the success of an intervention

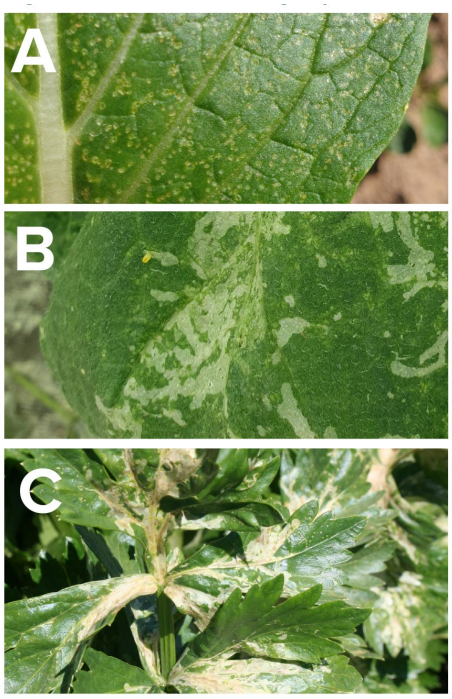


Figure 1: Damage caused by adult leafminer. A) SLM stippling damage to choy sum (Shannon Mulholland, NSW DPI); B) SLM damage to cucumber (Shannon Mulholland, NSW DPI); C) SLM damage to celery (John Duff, DAFF)

Counts of live larvae:

Counting larvae within leaf mines is more difficult than simply observing (or counting) leaf mines, as it generally requires the use of a hand lens to carefully check the wider ends of mines for a small whitish-yellow larva (see Figure 2), and for best results requires that living larvae can be distinguished from dead larvae. However, this method can produce significantly more accurate results for estimating population sizes, especially in longer lifespan crops like tomato, which can accumulate more damage before the plants are adversely affected. This method is most suited to

supporting the use of economic thresholds, and monitoring the success of interventions. Sampling plans based on larval counts are usually based on counting the number of 'active' mines in a subset of leaves on a subset of randomly selected plants, by checking mines for live larvae using a hand lens.

Counting larvae within leaf mines is the most labour intensive method, but also the most accurate method, having two major advantages over the use of traps such as pupal trays and yellow sticky traps: 1) it is most directly related to damage potential assessment as it focuses on the life stage responsible for the majority of damage; and 2) the resulting data is easier to incorporate directly into a decision making program that is based on population presence and allows for pesticide efficacy to be evaluated post sprays (Namvar et al., 2012).

Pros: accurate measure of population density, accounts for idiobiont ectoparasitoid activity (see Figure 5 and the "Monitoring for beneficial wasps" breakout box)

Cons: requires a hand lens and close inspection of leaves, underestimates koinobiont endoparasitoid activity (see Figure 5 and the "Monitoring for beneficial wasps" breakout box)

In Summary: preferred when monitoring infestation in long lifespan crops, or for monitoring the success of an intervention as it gives the most accurate population size estimates and is therefore a key component of global sampling plans aimed at using economic thresholds (ETs)

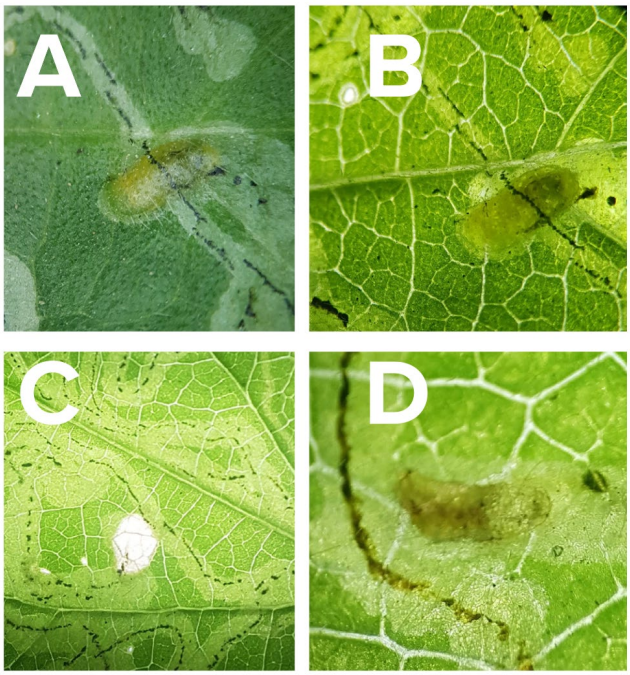


Figure 2. A) Live larvae (VLM pictured) can be seen feeding via a hand lens; B) Holding the leaf up to the sun can increase visibility of larvae inside mines; C) Inactive mines may be empty; or D) may contain a dead larva. (Elia Pirtle, Cesar Australia)

Counts of pupae:

PUPAL TRAYS

Johnson, Oatman, & Wyman (1980) described a method for monitoring leafminer in fresh market tomatoes based on counts of pupae collected within pupal trays (see Figure 3). The study showed that the number of pupae collected in pupal trays correlated significantly with the number of live larvae within leaflets. Thus, pupal tray sampling was efficient, inexpensive and more sensitive to population size changes than leaflet sampling (focusing on counting larvae within leaflets), and the trays became an integral part of an IPM program implemented for fresh market tomatoes in California. According to their method, Leafminer activity can be measured by collecting mature larvae which have fallen into polystyrene or plastic trays (pupal trays, between 8 x 11 to 12 x 15 inches in size) and pupated over a period of 3-4 days. These styrofoam trays are placed on the ground underneath plants and left in place for three days, at which point pupae trapped within the trays are counted, then removed and the traps replaced for further counts.

Pro: accounts for idiobiont ectoparasitoid activity (Figure 5), preferred as an alternative to counting live larvae in long lifespan fruiting crops such as tomato, as it does not require a hand lens as pupae are easier to observe and count after emergence

Cons: underestimates koinobiont endoparasitoid activity unless samples are retained for several weeks for rearing (Figure 5), can be poorly suited to short, leafy, or densely clumped crops such as lettuces and celery, can be poorly suited to wet areas

In Summary: Pupal trays are a popular method in long lifespan fruiting crops overseas due to being an easily visual indicator of whether leaf mine damage is caused by an active infestation, or whether the damage is old and thus intervention may be unwarranted; gives accurate population size estimates and can be used with Economic Thresholds.)

LEAF COLLECTION AND REARING

Pupae may also be counted by collecting a subset of leaves from a subset of randomly selected plants into plastic bags and observing the number of pupae that emerge and collect into the bottom of the bag (see Figure 3). This method has been incorporated into sampling plans such as in Foster (1986) to reduce reliance upon hand lens inspection of mines. Moreover, the pupae collected via pupal trays or via leaf collections may be retained in order to assess the level of parasitism by koinobiont endoparasitoids. Pupae may be kept in a plastic bag with a damp paper towel, out of direct sunlight, until adult flies or wasps emerge and adult flies may be counted. This improves accuracy of leafminer population size estimates because it accounts for

accounts for idiobiont ectoparasitoid and koinobiont endoparasitoid activity. However, it can take multiple weeks for all adult flies to emerge and wasps even longer, and is thus not suitable for quick decisions.

Pro: accounts for idiobiont ectoparasitoid activity (Figure 5), does not require a hand lens as pupae are easier to observe and count after emergence

Cons: underestimates koinobiont endoparasitoid activity unless samples are retained for several weeks for rearing (Figure 5),

In Summary: Colleting leaf samples for rearing provides clear visual indicators of whether leaf mine damage is caused by an active infestation, or whether the damage is old and thus intervention may be unwarranted; gives accurate population size estimates and can be used with Economic Thresholds.

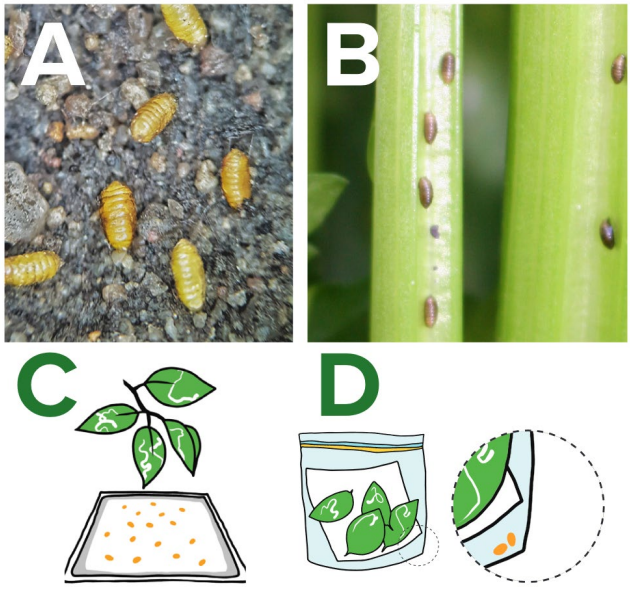


Figure 3. Small orange pupae (~2mm; VLM pictured) accumulate in the soil beneath infested plants (Elia Pirtle, Cesar Australia). B) SLM pupae collecting on plant surfaces in celery (John Duff, DAFF); Pupae can be collected into C) pupal trays placed underneath plants; or D) into the bottom of plastic bags on leaf collections.

Counts of adults:

Agromyzid flies are attracted to the colour yellow, and can therefore be captured on yellow sticky traps (see Figure 4), which are used to monitor a variety of invertebrate pests. Yellow sticky traps have been shown to be more effective for *Liriomyza* adults than other types of traps, such as funnel traps and yellow water pans, and vacuum sampling (Chavez & Raman, 1987; Weintraub, 2001).

A great deal of effort overseas has been dedicated to improving the effectiveness of yellow sticky traps for *Liriomyza* adults including the modifications of size, shape, adhesives, lures, height and orientation. For example, several studies report a strong effect of trap height on the number and species trapped, however these results do not always appear consistent and may be difficult to extrapolate across different crop types. Moreover, optimal height may vary considerable between *Liriomyza* species (Zehnder & Trumble, 1984). Sticky traps make for good indicators of leafminer presence and can be used to monitor movements of populations throughout or between paddocks, or indicate times of migration into a crop (Palumbo & Kerns, 1998). Sticky traps do have a few additional shortcomings, including (1) sticky traps require visual searches and rough morphological identifications must be made, (2) sticky traps appear to be are poor indicators of leafminer population sizes (sources) and are thus difficult to relate to damage and (3) sticky traps are poor indicators of parasitoid activity (Weintraub, 2001).

Experimental lures developed from the extracted volatiles of known plant hosts have been shown to be attractive to *Liriomyza*. For example, lures made from spruce, basil, juniper or clove oil have been shown to attract serpentine leafminer (Gorski, 2005). However, there are no products commercially available for use on *Liriomyza*.

Pros: does not require a hand lens as pupae are easier to observe and count after emergence

Cons: difficult to relate to population sizes and damage levels,

In Summary: Popular method overseas due to being an easily visual indicator of whether leaf mine damage is caused by an active infestation, or whether the damage is old and thus intervention may be unwarranted; gives accurate population size estimates and can be used with Economic Thresholds.

Figure 4. A yellow sticky trap hung above a tomato plant (left) and an adult VLM captured on the trap (right). (Elia Pirtle, Cesar Australia)

Monitoring for beneficial wasps

Idiobiont parasitoids:

SLM larvae which have been attacked by idiobiont parasitoid wasps (Fig. 5a) are immediately paralysed and never emerge from the leaf mine. Thus, counting *living* larvae (i.e. those actively feeding inside leaf mines) or pupae that have emerged from leaves avoids counting any larvae that were already attacked by idiobiont wasps, which would inflate the SLM population size estimate. Idiobiont ectoparasitoids* (which develop outside the body of the fly) can be observed inside leaf mines through a hand lens as either a larva, often found in close proximity to a dead leafminer larva (Fig. 5b), or as a pupa, flanked by black dots called meconial pillars (Fig. 5c).

In summary: Signs of idiobionts can be observed inside leaf mines via a hand lens (Fig. 5b/c)

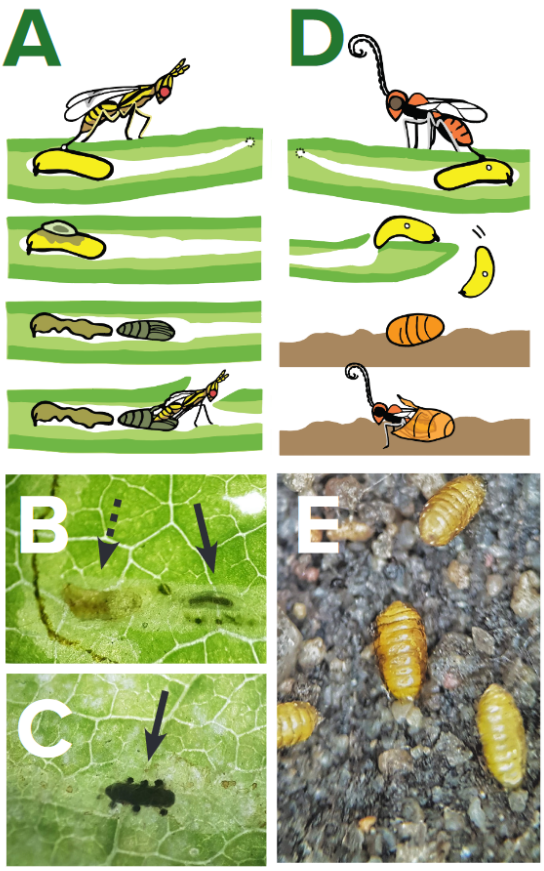
Koinobiont parasitoids:

SLM larvae which have been attacked by koinobiont wasps are NOT paralysed. They continue feeding and successfully pupate. Thus, counting living larvae or pupae that have emerged from leaves may still inflate the SLM population size estimates, because these counts may include individuals that will ultimately perish during the pupal stage. Koinobiont endoparasitoids (which develop inside the body of the fly) emerge from otherwise healthy looking leafminer pupae. Thus, pupae must be retained in rearing vessels for several weeks to confirm activity of these wasps.

In summary: There are no signs of koinobiont parasitism until emergence of adults from otherwise healthy looking fly pupae.

**some idiobionts are endoparasitoids, which pupate inside the dead fly larva, and thus may be less visible within the mine, however this is a minority of species*

Fig 5. Parasitoid lifecycles



A) An idiobiont ectoparasitoid lifecycle and signs of idiobiont parasitism under a microscope or hand lens, including B) a wasp larva (solid arrow) near a leafminer larva carcass (dotted arrow); and C) a wasp pupa inside a leaf mine (solid arrow). D) A koinobiont endoparasitoid lifecycle. E) There are no signs of koinobiont parasitism before emergence of adult flies and wasps from the fly puparium.

Getting an accurate population estimate

Making accurate estimates of leafminer populations is a prerequisite to using economic thresholds, aimed at reducing unnecessary chemical costs and unwanted toxicity effects on beneficials. However, leafminer distributions in a paddock are often clumped (as is true for many pests) which means that your population estimate may vary widely based on what part of the paddock you searched. However, you can use mathematical rules to tell you exactly how many plants you must search before you can be reasonably confident that your measured pop density captures enough variation to accurately reflect the whole paddock. In the case of these patchy distributions, Taylor's power law becomes an appropriate method for determining sample sizes (Ruesink, 1980).

Thus, several types of sampling plans, based on these mathematical rules for non random aggregations, have been developed and applied overseas to estimating leafminer populations (Burgio et al., 2005; Heinz & Chaney, 1995; Jones & Parrella, 1986; Namvar et al., 2012) for the purposes of making informed management decisions (Table 1). These can generally be split into 'conventional' and 'sequential' sampling plans.

- Conventional sampling plans operate on a fixed number of samples that are taken per unit of area, and the resulting precision of the population size estimate will vary with the population density (Lopes et al., 2019).
- Sequential sampling plans on the other hand have a pre-determined level of precision which must be reached, and samples are taken until that fixed level of precision is reached. The ultimate number of samples that must be taken relates to the population density, and surveyors know when sufficient samples have been collected by referring to a pre-calculated 'stop line' (see Figure 6 for an example).

Conventional sampling plans tend to be the starting points for developing decision making systems for pest control interventions (Lopes et al., 2019), while sequential sampling plans can provide increased efficiency (Namvar et al., 2012).

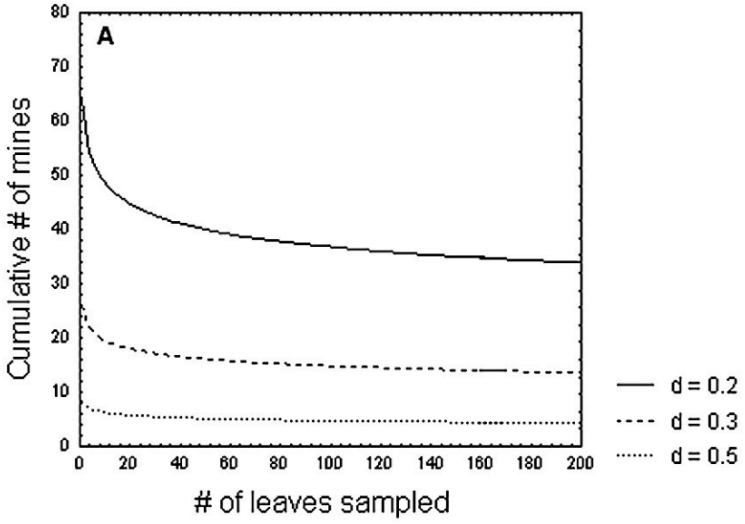


Figure 6. An example graph showing stop lines for leaf mine counts, reproduced from Burgio et al. (2005). Each line on the graph shows the "stop numbers" for three levels of precision, where you can stop counting once you reach the desired number of mines per the number of leaves you have checked. To use the stop line, keep a cumulative tally of how many mines you have counted alongside how many leave you have checked, and stop counting once you reach the number of mines per leaf corresponding to a point on the line of your chosen accuracy level. For example, if after checking about 40 leaves you if you count more than 40 mines, you know you have done enough sampling to estimate population size with only a 20% margin of error.

Table 1 provides a summary of several conventional and sequential sampling plans for leafminer in a variety of crops, and provides key rule of thumbs from these plans. These plans may provide some rough rules of thumb that can serve as starting point in Australia, however, they cannot be relied upon as accurate sampling plans in Australia until they are formally validated. Australian specific sampling plans and economic thresholds will need to be created to support successful IPM programs to manage exotic *Liriomyza* spp. leafminer.

Table 1: Population density sampling plans implemented for *Liriomyza* species globally in commercial crops.

Leafminer species and crop	Reference	Reference Title	Type	Sample unit	Summary of plan	Other notes
SLM in tomato	(Lopes et al., 2019)	Practical sampling plan for *Liriomyza huidobrensis* (Diptera-Agromyzidae) in tomato crops	Conventional	Active mines (e.g. live larvae)	Count active mines in 73 leaf samples per field (irrespective of field size up to 10 ha), taking random leaves from the basal leaf of the middle section of the plant canopy	Average time requirement was 30 min of leaf evaluation time (plus walking time which was up to one hour for 10 ha fields)
SLM in potato	(Alves et al., 2014)	A Sampling Plan for *Liriomyza huidobrensis* (Diptera: Agromyzidae) on a Potato (*Solanum tuberosum*) Plantation	Conventional	Active mines (e.g. live larvae)	Count active mines in one random leaf sample from the middle canopy section from 15 random plants (at least 50m apart) per 24.5 ha	Average 30 minutes total sampling time per 24.5 ha Cost was significantly lower than insecticides
VLM in glasshouse cucumber	(Namvar et al., 2012)	Estimation of larval density of *Liriomyza sativae* Blanchard (Diptera Agromyzidae) in cucumber greenhouses using fixed precision sequential sampling plans	Sequential	Active mines (e.g. live larvae)	Count active mines per leaf in random leaf samples until a larvae count stop line (based on desired level of accuracy) is reached (See Supp Fig 1).	With the precision of 0.28, samples required varied between 2 to 157 leaves, when mean larval density per leaf declined from 29.1 to 0.07. For precision of 0.25, densities > 4 larvae per leaf required < 11 samples, but densities of < 1 larvae required > 32 samples

VLM in glasshouse cucumber	(Namvar et al., 2011)	Fixed precision sequential sampling plans for leaf mines of Liriomyza sativae Blanchard (Diptera: Agromyzidae) in cucumber greenhouses	Sequential	Active mines (e.g. live larvae)	Count active mines per leaf in random leaf samples until a larvae count stop line (based on desired level of accuracy) is reached.	Sample sizes ranged from 3 to 197 and 15 to 1229 leaves at the precision levels of 0.25 and 0.1 respectively. This is an earlier analysis of the data used within Namvar er al. (2012)
LM in tomato	(Schuster & Beck, 1992)	Presence-absence sampling for assessing densities of larval leafminers in field-grown tomatoes	Presence-Absence	Proportion of leaflets that contain active mines	Record the proportion of leaflets that have any live larvae present by checking the upper surface of the terminal three leaflets of the 7th leaf from the top of either a main stem, lateral or sub-lateral stem from randomly selected plants.	Proportion infested leaves can be used to predict number of larvae present per sample, to reduce counting time per leaflet This study did not address how many samples needed to create an accurate paddock wide density estimate
SLM in lettuce	(Burgio et al., 2005)	Spatial Patterns and Sampling Plan for *Liriomyza huidobrensis* (Diptera: Agromyzidae) and Related Parasitoids on Lettuce	Sequential	Mined leaves (not distinguishing active from inactive mines)	Count leaves with mines from random leaf samples until the number of mined leaves collected exceed stop line values for the number of overall leaves collected (See Supp Fig 2).	This paper advises that damage thresholds cannot be predetermined as they may vary by environment/agroeconomic conditions
SLM in celery	(Heinz & Chaney, 1995)	Sampling for *Liriomyza*	Sequential	Active mines (e.g. live larvae)	Count all active mines per randomly selected plants until a larvae count stop	Sequential sampling plan accurately estimates mean densities > 17.5 live larvae per

		huidobrensis (Diptera: Agromyzidae) larvae and damage in celery			line (based on desired level of accuracy) is reached (See Supp Fig 3), with a possible maximum sample size of 100 petioles	100 petioles with a 0.25 level of precision Lower densities of larvae or mines required sample sizes > 100 petioles at a level of precision $\geq$ 0.25 to accurately estimate cumulative or mean leafminer densities. Validation tests showed that using frequencies of infested petioles as a proxy for counting active mines overestimated population density
LM in watermelon	(Lynch & Johnson, 1987)	Stratified Sampling of *Liriomyza* spp. (Dipetra: Agromyzidae) and Associated Hymenopterous Parasites on Watermelon	Stratified	Active mines (e.g. live larvae) per leaf	Count larvae within medium sized leaves, randomly selected within the area greater than 0.5 meters from either end of the plant vine (because of higher variation in insect densities in the extreme basal and distal portions of a vine)	Standard errors were reduced by >46 and 35%, respectively, when leaf sizes were stratified (by dividing vines into 50 cm intervals, or strata, starting at the plant base and ending in the distal end of the vine, and taking random leaf samples within each strata) This study did not address how many samples needed to create an accurate paddock wide density estimate
ASLM in chrysanthemum	(Jones & Parrella, 1986)	Development of Sampling Strategies for Larvae of *Liriomyza trifolii* (Dipetra:	Conventional	Active mines (e.g. live larvae)	Count active mines from three leaves per each randomly selected plant until 100 leaves have been samples.	After about 3 weeks, sampling should focus on the bottom strata of the plant, and after 6 weeks, sampling should focus on the middle strata of the plant (where larval numbers tend to be highest)

		Agromyzidae) in Chrysanthemums				
ASLM in celery	(Foster, 1986)	Monitoring Populations of *Liriomyza trifolii* (Diptera: Agromyzidae) in Celery with Pupal Counts	Conventional	Pupae emerging from picked leaves	Pick ten terminal leaflets from each of 10 randomly selected plants, at each of ten systematically placed sites within the paddock. Place leaflets into a plastic bag and maintain them for no more than ten days, and then count all emerged pupae.	As a rule of thumb, assume 5 pupa or less per 10 leaflet samples poses no economic threat Number of samples necessary depends on leafminer densities, where, if average density is >5 pupa per 10 leaflet samples, 10 sample sites (of 10 leaflets each) yields 25% level of precision The sampling plan required 30 to 45 minutes total time to sample an 11 hectare field
LM in tomato	(Zehnder & Trumble, 1985)	Sequential Sampling Plans with Fixed Levels of Precision for *Liriomyza* species (Diptera: Agromyzidae) in Fresh Market Tomatoes	Sequential	Adults on yellow sticky traps	Adults are counted on sticky traps until the cumulative number of adults exceeds the stop line value for the number of sticky traps checked (See Supp Fig 4).	Approximate number of sticky traps that must be place in a field to yield enough samples to reach the desired precision level can be estimated based on how many adults are caught on 'pilot' yellow sticky traps (see Supp Fig 5).
LM in tomato	(Zehnder & Trumble, 1985)	Sequential Sampling Plans with Fixed Levels of Precision for *Liriomyza* species (Diptera:	Sequential	Pupae within pupal trays	Pupae counted within pupal trays until the cumulative number of pupae exceeds the stop line value for the number of	

		Agromyzidae) in Fresh Market Tomatoes			pupal trays checked (See Supp Fig 4).	
ASLM in greenhouse chrysanthemum	(Parrella & Jones, 1985)	Yellow Traps as Monitoring Tools for *Liriomyza trifolii* (Diptera: Agromyzidae) in Chrysanthemum Greenhouses	Sequential	Adults on yellow sticky traps	Adults are counted on sticky traps until the cumulative number of adults exceeds the stop line value for the number of sticky traps checked (See Supp Fig 6).	Traps must be placed over 'homogenous' blocks of plants (planted less than 30 days apart) A validation trail showed only 18% of 792 traps that had been placed needed to be counted to provide sufficient accuracy for population size estimates.

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Supplementary Figures

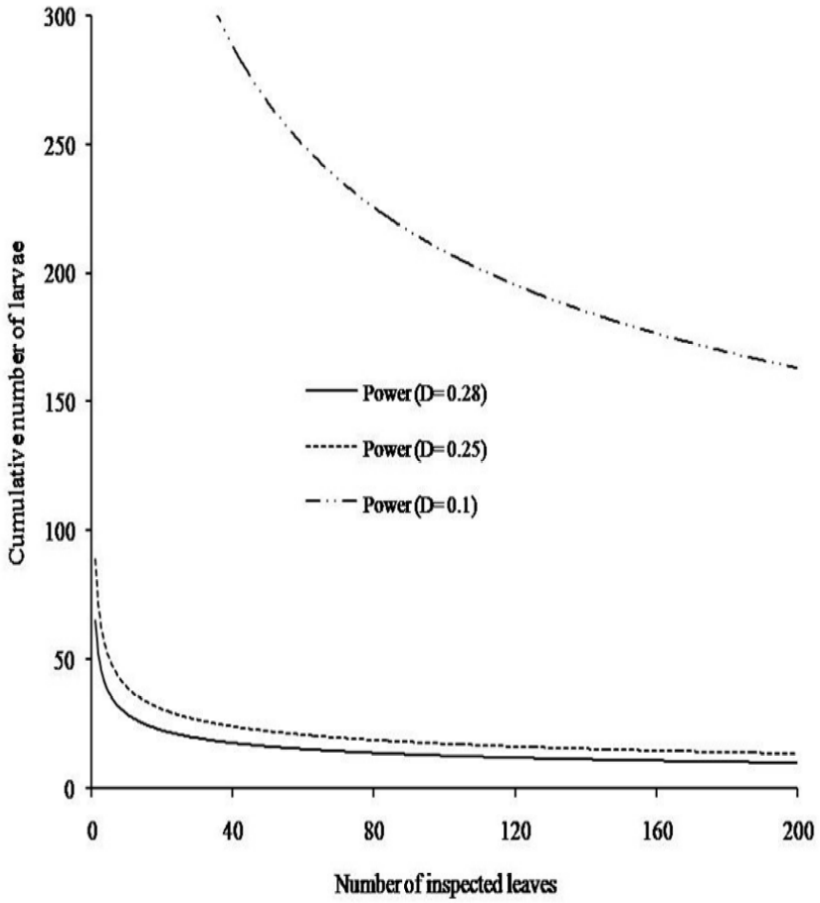

Figure 2. Sequential sampling stop lines for fixed- precision level (D) of 0.1, 0.25 and 0.28 for various *Liriomyza sativae* larval densities.

Supplementary Figure 1. Stop lines for live larva counts, reproduced from Namvar et al. (2012).

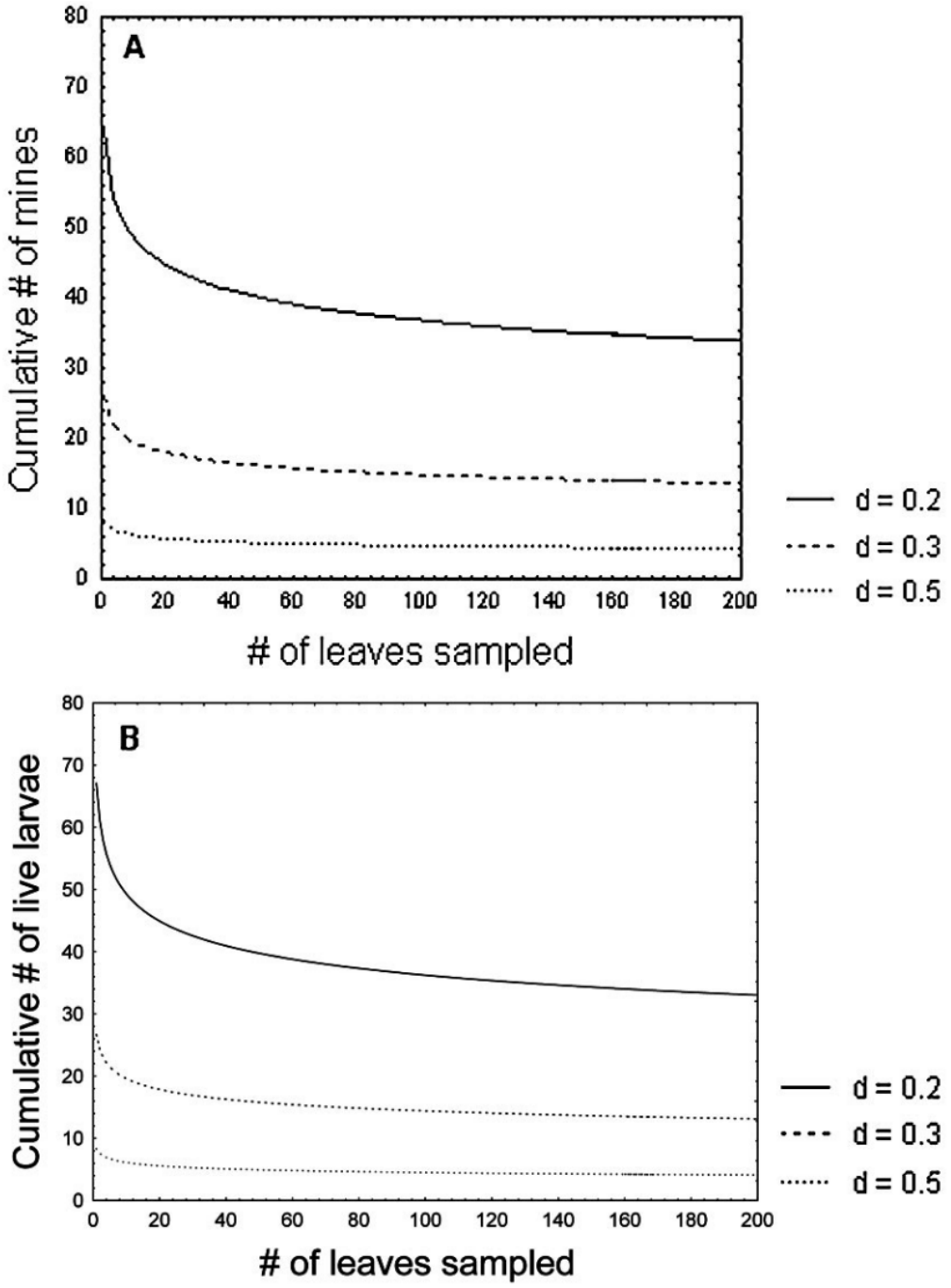

Fig. 1. Stop lines calculated for *L. huidobrensis* mines (A) and live larvae (B) at three different precision levels.

Supplementary Figure 2. Stop lines for live larva counts compared to mine counts, reproduced from Burgio et al. (2005)

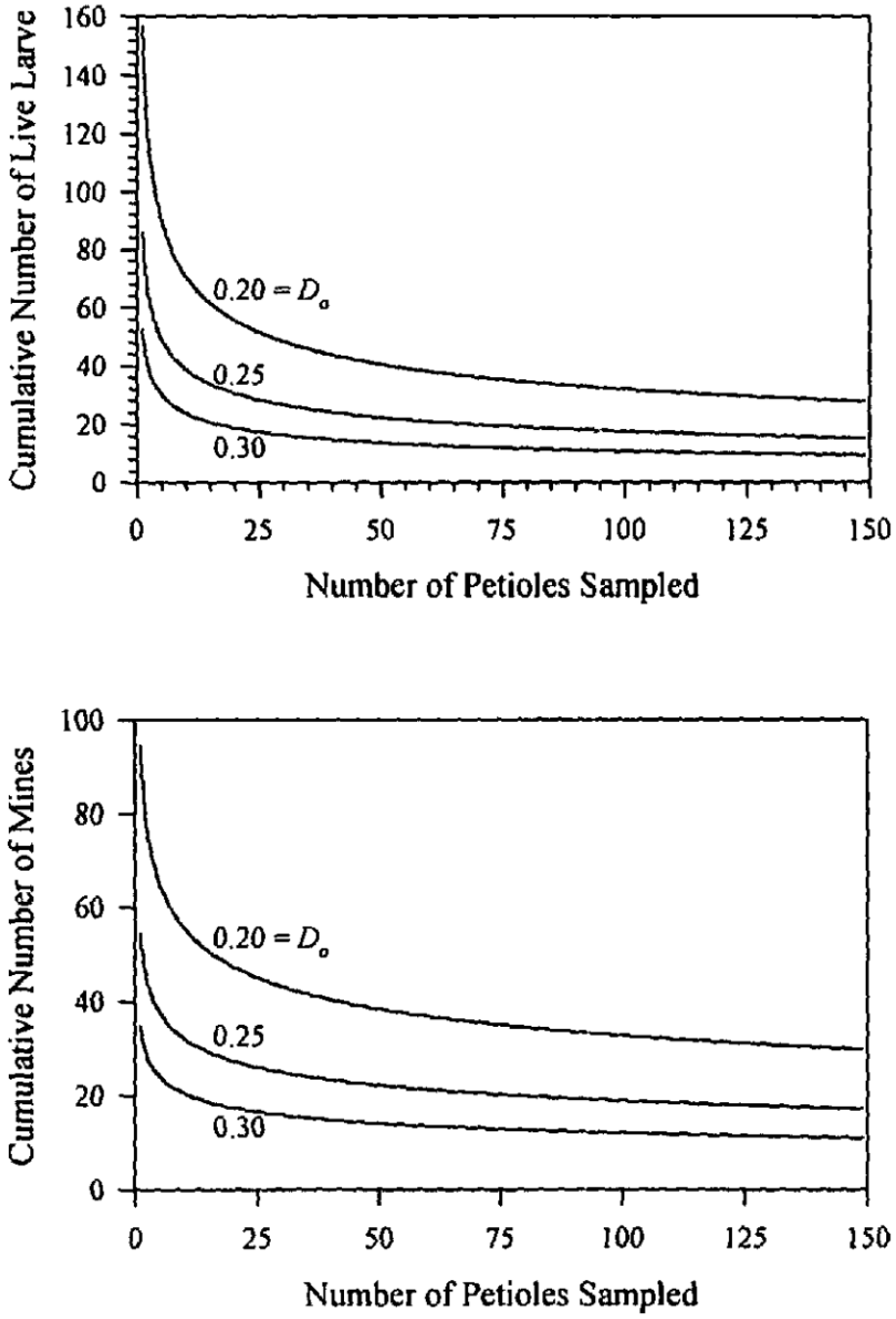

Fig. 3. Stoplines for constant-precision sequential samples for L. huidobrensis larvae (top) and mines (bottom) at three levels of precision (D_0) equal to 0.20, 0.25, and 0.30.

Supplementary Figure 3. Stop lines for live larva counts compared to mine counts, reproduced from Heinz & Chaney (1995)

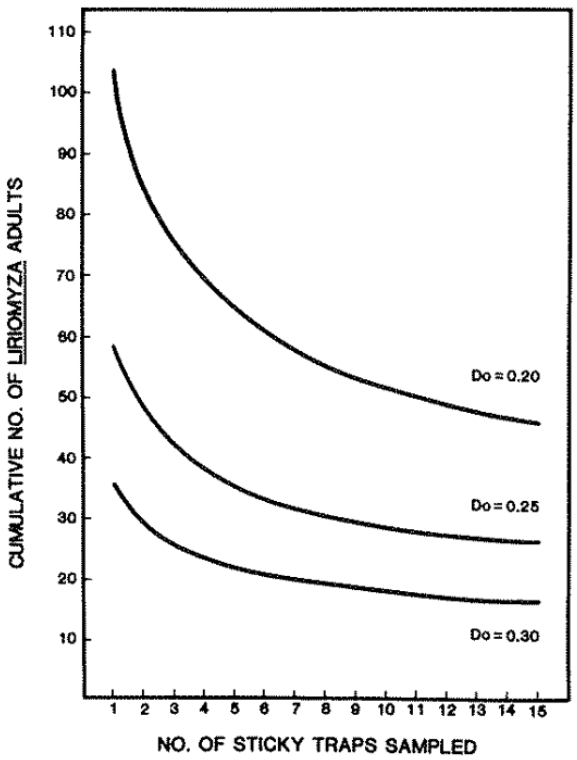

Fig. 2. Stoplines for constant-precision sequential samples for *Liriomyza* adults at three levels of precision (D_0) of 0.20, 0.25, and 0.30.

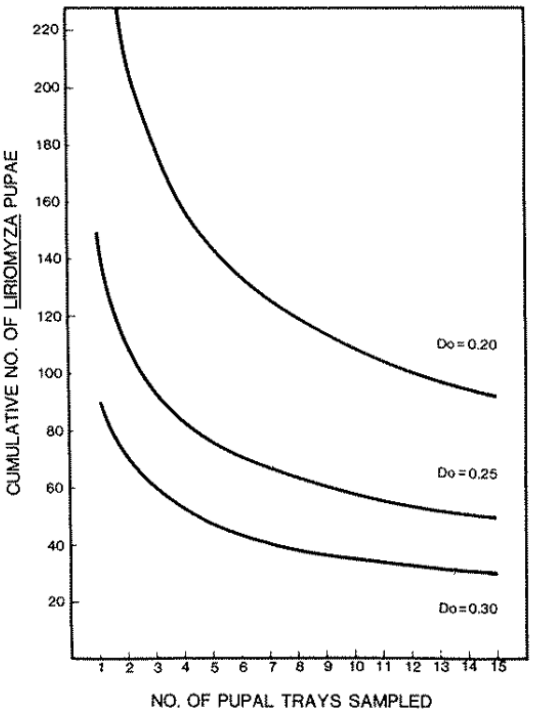

Fig. 3. Stoplines for constant-precision sequential samples for *Liriomyza* pupae at three levels of precision (D_0) of 0.20, 0.25, and 0.30.

Supplementary Figure 4. Stop lines for sticky trap and pupal tray samples, reproduced from Zehnder & Trumble (1985)

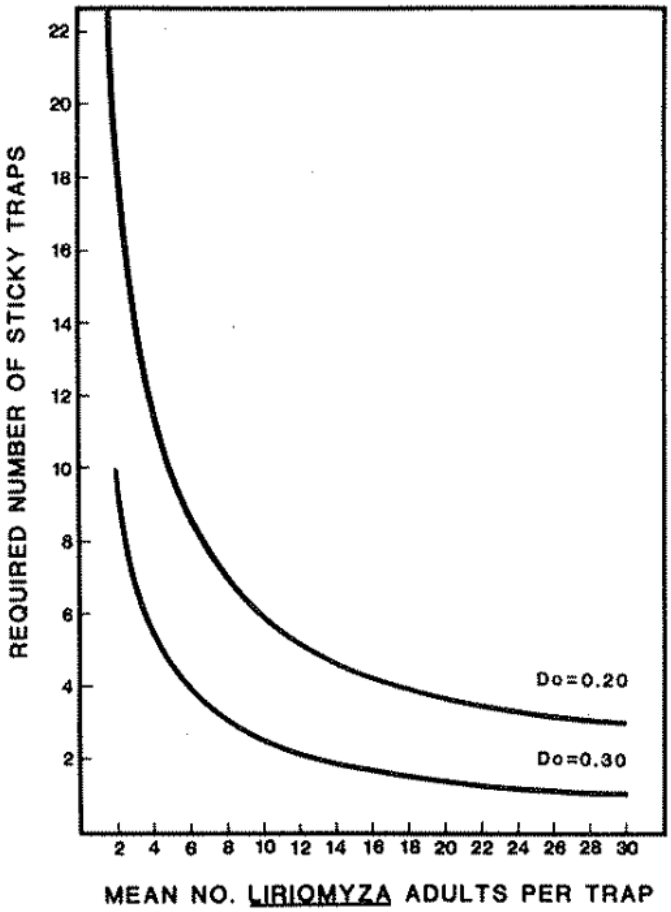

Fig. 4. Required number of sticky traps for various *Liriomyza* adult densities at two levels of precision (D_0) of 0.20, 0.30.

Supplementary Figure 5. A guideline for determining the approximate number of sticky traps that must be place in a field to yield enough samples to reach the desired precision level, based on how many adults are caught on 'pilot' yellow sticky traps, reproduced from Zehnder & Trumble (1985)

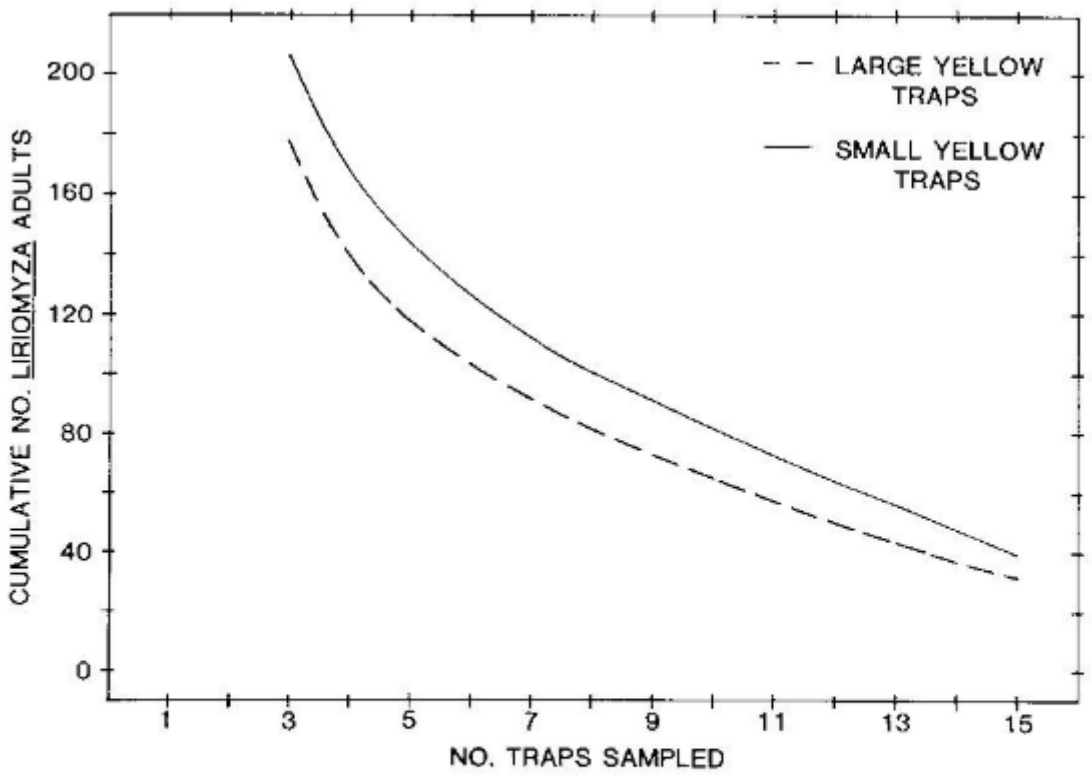

Fig. 3. Stoplines for constant precision sequential samples for large and small sticky cards used to trap *L. trifolii*, generated using Iwao's patchiness regression. The fixed level of precision is 0.25.

Supplementary Figure 6. Stop lines for sticky trap and pupal tray samples, reproduced from Parrella & Jones (1985)

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