EFFECTIVENESS OF CYROMAZINE APPLICATION AND PARASITOIDS RELEASE FOR CONTROLLING AGAINST LEAFMINER FLY *Liriomyza huidobrensis* (BLANCHARD) (DIPTERA: AGROMYZIDAE)

UJI EFEKTIVITAS APLIKASI INSEKTISIDA DAN PELEPASAN PARASITOID UNTUK PENGENDALIAN LALAT PENGOROK DAUN *Liriomyza huidobrensis* (BLANCHARD) (DIPTERA: AGROMYZIDAE)

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ABSTRAK

Liriomyza huidobrensis (Diptera: Agromyzida) merupakan hama invasif yang menjadi penting pada berbagai tanaman. Selama ini pengendaliannya hanya mengandalkan aplikasi insektisida yang diduga justru memberikan efek negatif antara lain peningkatan resistensi dan penurunan populasi musuh alaminya. Penelitian ini bertujuan untuk menguji keefektifan insektisida berbahan aktif siromasin dan parasitoid dalam menurunkan serangan lalat pengorok daun L. huidobrensis. Penelitian dilakukan di Desa Sirukem, Kecamatan Kalibening, Kabupaten Banjarnegara, Provinsi Jawa Tengah dari bulan Juli sampai November 2009. Percobaan dilakukan dengan Rancangan Acak Kelompok dengan empat perlakuan, yaitu kontrol (tanpa perlakuan), aplikasi siromasin, pelepasan parasitoid, dan kombinasi antara aplikasi siromasin dengan pelepasan parasitoid. Hasil penelitian menunjukkan bahwa aplikasi siromasin yang dipadukan dengan pelepasan parasitoid secara nyata menurunkan serangan lalat pengorok daun L. huidobrensis lebih besar dibandingkan dengan perlakuan lain. Selama percobaan, parasitoid dominan yang ditemukan adalah Opius chromatomyiae dan Hemiptarsenus varicornis. Kedua parasitoid tersebut lebih banyak ditemukan di daun kacang babi (Vicia faba) dibandingkan pada daun kentang (Solanum tuberosum). Analisis menunjukkan bahwa penggunaan insektisida berbahan aktif siromasin justru meningkatkan intensitas parasitisasi.

Kata kunci: *Liriomyza huidobrensis, Siromasin, Parasitoid*

ABSTRACT

Liriomyza huidobrensis is an invasive pest for various crops. Till now, the management of this pest is by the only use of pesticides which hence increased its resistance and decrease of its natural enemies. The objective of the study was to assess the effectiveness of cyromazine and parasitoids for reducing the population of leafminer fly (L. huidobrensis) and to analyse the economic impact of the different treatments. The study was conducted in the Sirukem village, Kalibening sub district, Banjarnegara district, Central Java from July to November 2009. To address the objective, four treatment strategies were applied: no treatment, cyromazine, parasitoid release and cyromazine-parasitoid, and observation of damage intensity, sampling of infested leaves, assessment of yield and an economic analysis were undertaken.. The results found that cyromazine and parasitoids were effective to control against infestation of Liriomyza. Cyromazine was also harmless for parasitoids. The dominant parasitoid found *were Opius chromatomyiae and Hemiptarsenus varicorni which preferred to parasitize the larvae of L. huidobrensis on Vicia faba than on Solanum tuberosum.*

Keywords: *Liriomyza huidobrensis, Cyromazine, Parasitoid*

INTRODUCTION

Liriomyza is considered as an important pest because of its high levels of damage and difficulties to control. Damage on leaves is caused by adult and larval feeding activity. Parrella[1] reviewed that *Liriomyza* damages to the crops and argued that it happened in at least six ways: (1) becoming the disease vector, (2) destroying young seedlings, (3) reducing crop yield, (4) accelerating leaf drop, (5) reducing aesthetic value of ornamental crops, and (6) causing some plants to be quarantined. In Indoneasia, leafminers were reported first in 1995[2] and now they are found in all islands of Indonesia.[3,4] Rauf et al.[3] found that attack of leafminer on vegetables reduced yield by 60–70%. A similar percentage of damage was reported by Hidrayani et al.,[5] who found that, at the end of vegetative growth stage, damage reached 50–100%. In Indonesia, two dominant species of *Liriomyza* are *L. huidobrensis* (Blanchard) and *L. sativae* Blanchard. *Liriomyza huidobrensis* is found in high altitudes where potatoes are the main crop, while *L. sativae* dominates in low altitudes where vegetables like tomatoes, cucumber and beans are the main crops.

Up to recent, the only method used to control leafminer in Indonesia is heavy application of insecticides. Even though some insecticides are reported to be effective to control this pest, their indiscriminate use has resulted in the development of resistance of *L. huidobrensis* to the active compounds of the insecticides. Surveys have revealed that most farmers intensively applied insecticides twice a week, but this did not suppress the leafminer population.[3] Thus, alternative methods must be developed using natural enemies. As suggested by Murphy and LaSalle,[6] parasitoids might have more potential as a control agent, given the rich community of natural enemies that is found in association with the leafminer. As reported by Warsito,[7] twelve parasitoids were discovered associated with the leafminer in Java. The parasitoids fell into three families: *Asecodes delucchii, A. erxias, Chrysocharis* sp., *Cirrospilus ambiguus, Closterocerus* sp., *Hemiptarsenus varicornis, Neochrysocharis beasleyi, N. formosa, N. okazakii, Quadrastichus liriomyzae* are from the Eulophidae family, *Gronotoma micromorpha* is from the Eucoilidae family, and *Opius chromatomyiae* is from the Braconidae family.

Parasitoids diversity decreases with altitude, whereas *L. huidobrensis,* relatively increases.[6] This may be explained by the fact that insecticides were intensively applied at these altitudes. Laboratory research has shown that natural enemies, especially parasitoids, are more susceptible to insecticides. Besides causing mortality of beneficial insects, insecticides also reduce the searching capacity and number of eggs laid, which can result in a reduced level of parasitism.[3,5,8] However laboratory study of Prijono's shows that cyromacine is effective against larvae of *L. huidobrensis* and has harmless for parasitoids.[9]

That's different results rise a debate of insecticides use. Therefore this study was conducted to assess the effectiveness of cyromazine and parasitoids in reducing the population of leafminer fly (*L. huidobrensis*) in high altitudes conditions.

MATERIALS AND METHODS

The study was undertaken in the Sirukem village, Kalibening sub district, Banjarnegara district of Central Java from July to November 2009. The altitude of the site is around 1,300 meters above sea level (a.s.l.). Good quality potato seed was planted for this study on 0.5 ha, with 0.70 cm by 0.30 cm of planting distance. To assess the effectiveness of cyromazine application and parasitoids release against infestation of *Liriomyza*, a study with four treatments and three replications was undertaken. The treatments were 1) monocrop potato without insecticide application and parasitoids release (no treatment), (2) monocrop potato with cyromazine, which is applied based on recommended dosage (cyromazine), (3) habitat modification by intercropping potatoes with faba bean (*Vicia faba*) and releasing parasitoids collected from low and mid altitudes (parasitoids release), and 4) integrating cyromazine applications and parasitoid release on intercropping plot between potato (*Solanum tuberosum*) and faba bean (cyromazine-parasitoid). Each plot was 5 m by 3 m. Except for treatments, all cultivation practices, including fertilizing, fungicides, up hilling, weeding, irrigation, etc., were held similar among treatments. To control infestation of late blight and other diseases, Wendry 75WP (clorotalonil active compound) and Victory Mix 8/64 WP (mancozeb active compound) were applied ten times.

In the cyromazine plot, whether for single treatment of cyromazine or cyromazine integrated with parasitoids release, Cyrrotex 75 SP (75% of cyromazine active compound) was applied three times for 35 days after planting (DAP), 50 DAP and 62 DAP. The recommended dosage is 0.3–0.6 gr per litre. Moreover, rather than following the Zamzami[10] work in which parasitoid release was performed by hanging infested leaves over one meter height for one week, parasitoids were released after infested leaves were kept for 2 weeks in the laboratory letting all parasitoids emerge. The Zamzami work was tested, but the leaves hung dried rapidly before the parasitoids emerged.

In order to rear the parasitoids, the infested leaves of potatoes and faba beans were collected and then cleared from other insects by brushing the surfaces to ensure that the parasitoid that emerged was only from *Liriomyza* rather than other insects. Infested leaves were kept in plastic containers (width by depth by height: 27 by 18 by 15 cm) with tissue paper to keep humidity high and increase survival of parasitoids. Infested leaves were kept for 10–14 days until almost all parasitoids emerged. The parasitoids were then released in both treatments plots, with or without cyromazine application. Parasitoids release was conducted twice, 35 DAP and 50 DAP. For the first release, infested leaves were collected from the Kalibening and Jatilawang villages where their altitudes are 1,100 and 1,300 a.s.l. respectively. Only 25 parasitoids were released in which the dominant parasitoid *O. chromatomyiae* shared about 92% of total. For the second release, infested leaves were collected from the Pesantren and Karang Malang villages where altitudes are 1,100 and 1,500 m.s.l. respectively. In dominance of *O. chromatomyiae* (89%), 200 parasitoids were released.

Observation and Analysis

This study was designed in three observation approach, which were monitoring damage intensity, sampling infested leaves to assess population of *L. huidobrensis* and parasitoids, and measuring yield.

Damage intensity: For each plot, five crops were randomly sampled. To asses the intensity of damage, infested leaves were counted and compared to the total leaves. Observation was conducted three times when crops were at 40 DAP, 55 DAP, and 65 DAP. To analyse the impact of each treatment on damage intensity, ANOVA was undertaken. The factors for which significant effects were detected were further analysed with Tukey's posthoc test to assess which treatments differed significantly at the 5% level. The analyses were performed using SPSS version 15.

Infested leaves sampling: Number of individuals that emerged and percentage of parasitism were observed by collecting 10 damaged leaves a week after releasing parasitoids (42 and 57 DAP) wanting to allow parasitoids to work before observing. Collecting infested leaves was undertaken for potato in no treatment and cyromazine plots and potato and faba beans in parasitoids release plots. Potato and faba bean leaves were then kept singly in plastic vials (width by dept by height: 27 by 18 by 15 cm). To prevent rapid drying and the development of mould, tissue paper was placed into the vials. The infested leaves were kept for one month until all parasitoids and leafminers emerged. Parasitoids and leafminers were then identified and counted. Specimens from each sample were stored in a labelled vial containing 70% ethanol. Parasitoids were identified using Liriomyza Parasitoid Lucid3 Key.[11] As for damage intensity, the parasitoid level among the treatment was compared through ANOVA and continued by Tukey's B posthoc test with 5% level to assess whether treatments were significantly different. Besides differences among treatments, percentage of parasitism was also compared

Percentage of damage was counted as:

$$\text{Percentage of parasitism} = \frac{\text{number of parasitoids}}{\text{number of parasitoids} + \text{number of leaf miners}} x100\%$$

between its occurrence on potatoes and beans. Moreover, number of individuals that emerged and percentage of parasitism collected from parasitoids release and cyromazine-parasitoids release plots were compared between potato and faba bean through ANOVA and continued by Tukey's B posthoc test with 5% level.

Yield: Yield was measured for sampling crops and the data was further used to analyse a correlation between damage level and yield. The correlation was assessed by performing regressions between damage intensity and yield

RESULTS

Damage intensity: Infestation of *Liriomyza* occurred immediately after the crops appeared above soil. However intensity of damage was first observed on 40 DAP. The intensity increased with time of observation and ANOVA showed that treatments significantly influence the intensity of damage for all observation time (F=29.93, df=3,48, *P*=0.00). At first observation, intensity of damage was still low in all treatments, ranging less than 20%. Analysis showed that cyromazine and parasitoid release effectively reduced the infestation, indicated by significantly lower intensity of damage in cyromazine, parasitoid release and cyromazine-parasitoids plots compared to the no treatment one (Figure 1). The lowest intensity occurred on the cyromazine-parasitoids plot in which a difference reached about 10% compared to it on the no treatment plot.

Different conditions occurred on the second observation. Even though treatment signifi-

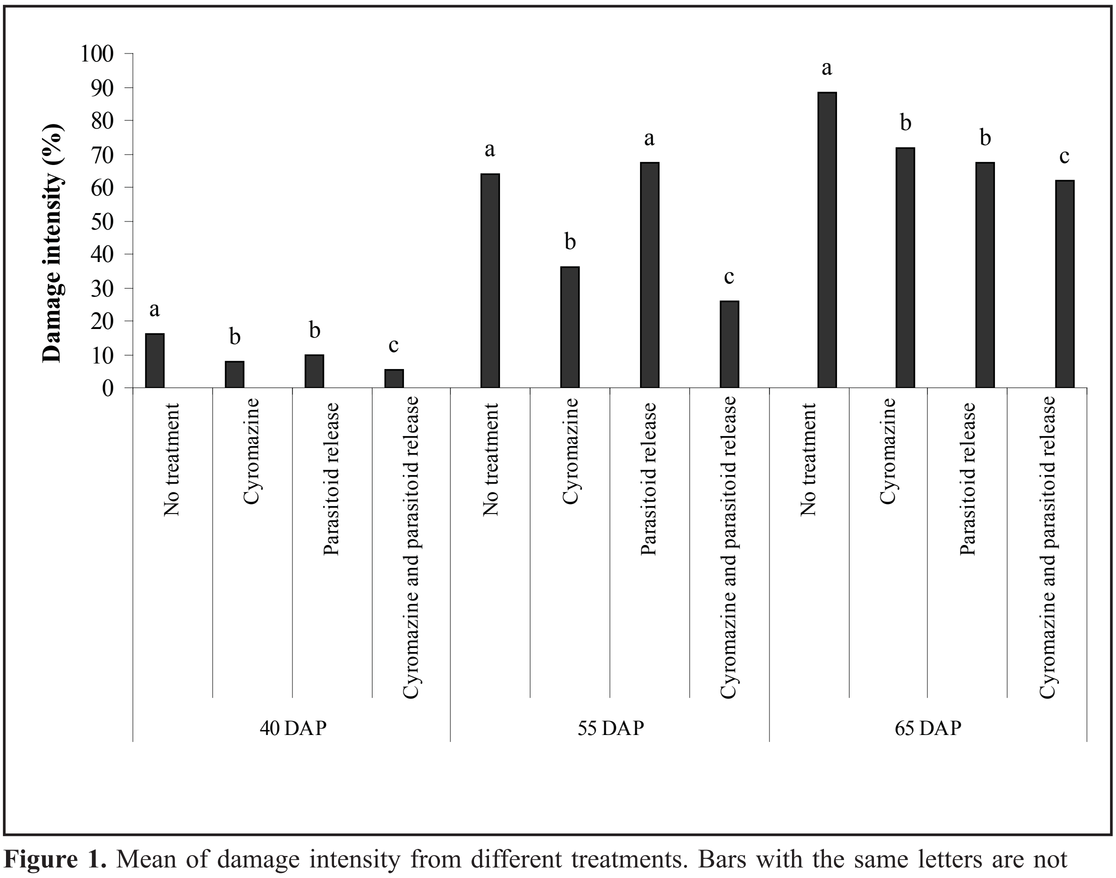

Figure 1. Mean of damage intensity from different treatments. Bars with the same letters are not significantly different based on Tukey's B posthoc analysis at the 0.05 level.

cantly reduced the intensity of damage (F=39.84, df=3,48, *P*=0.00), it only happened on the plots on which cyromazine was applied. The lowest damage intensity still occurred in cyromazine-parasitoids plot. In this plot, the intensity averaged only 25% compared to more than 60% on no treatment and parasitoids release plots.

Similar to the first observation pattern, the intensity of damage on the third sampling significantly differed among treatments (F=42.99, df=3,48, *P*=0.00) in which cyromazine and parasitoid was likely to successfully reduce the intensity. In all plots in which both components were tested, the intensity of damage was significantly lower than the control (no treatment) one. The lowest damage still occurred in cyromazine-parasitoids plot with almost 25% difference compared to the no treatment one. However, the intensity of damage in all plots was relatively high which it averaged more than 60%.

Infested leaf observation: At the first infested leaves sampling (42 DAP), the average number of *L. huidobrensis* and parasitoid ranged from 4.8 to 6.4 individuals per leaf and those numbers did not significantly differ among the treatments (F=1.75, df=3,108, *P*=0.16) (Figure 2). *Opius chromatomyiae* dominated the parasitoid colony and shared more than 90%. The rest of the parasitoids were only *H. varicornis.* Percentage of parasitism differed with treatments in which cyromazine-parasitoid release treatment shared about 57% and it was significantly higher compared to others (F=20.81, df=3,108, *P*=0.00).

The population of individuals that emerged (*L. huidobrensis* and parasitoid) per leaf declined, but the percentage of parasitism increased with time of observation. At the second sampling (57 DAP), treatment also did not influence the number of *L. huidobrensis* and parasitoid (F=7.38, df=3,108, *P*=0.07); only about 3.7-4.7 *L. huidobrensis* and parasitoids were found in each leaf (Figure 2). At this observation, percentage of parasitism significantly differed with treatments (F=11.70, df=3,108, *P*=0.00); parasitism rates in parasitoid release and cyromazine-parasitoid release were higher than the other two treatments. In both plots, the parasitism rate reached 50.7% and 63.0% compared to only 30.6% and 38.0% in no treatment and cyromazine plots, respectively.

Beside potatoes, infested leaves were also collected from faba beans in parasitoid release plots. The number of individuals that emerged and percentage of parasitism were compared between two parasitoid release plots or between potato and faba beans. Between plots, no significant data occurred in all sampling times (F=0.42, df=1,54, *P*=0.52; F=0.10, df=1,54, *P*=0.75; F=3.61, df=1,54, *P*=0.07 for number

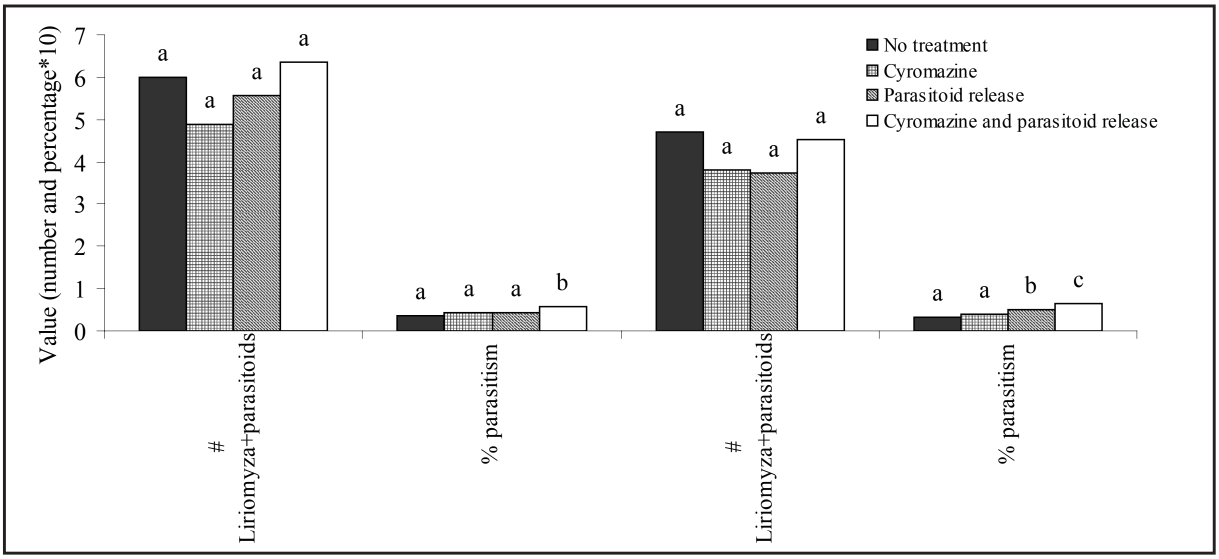

Figure 2. Mean of number individuals emerged (*L. huidobrensis* and parasitods) and percentage parasitism at the 1st and 2nd observations collected from potato infested leaves in all treatments. Bars with the same letters are not significantly different based on Tukey's B posthoc analysis at the 0.05 level.

of individuals emerged at the 1st observation, percentage parasitism at the 1st observation and number of individuals emerged at the 2nd observation respectively), with the exception of the number of individuals that emerged at the second sampling (F=10.99, df=1,54, *P*=0.00). In this case, the number of individuals that emerged from the parasitoid plot was significantly higher than from the cyromazine-parasitoid plot in which they averaged 9.4 and 8.8 individuals per leaf. All *Liriomyza* were *L. huidobrensis,* while parasitoid consisted 98% *O. chromatomyiae* and *H. varicornis* for the rest.

The number of individuals that emerged and the percentage of parasitism were significantly higher on faba beans than on potatoes (F=18.85, df=1,98, *P*=0.00; F=6.80, df=1,98, *P*=0.00; F=20.13, df=1,98, *P*=0.00 for the number of individuals emerged at the 1st observation, percentage of parasitism at the 1st observation and the number of individuals at the 2nd observation respectively) with the exception of the percentage of parasitism at the second sampling (F=0.27, df=1,98, *P*=0.60). At the first sampling, the number of individuals that emerged on beans reached 8.7 per leaf compared to 6.0 on potatoes and the percentage of parasitism was 60.1% compared to 50.7%. Moreover at the second sampling, only the number of individuals that emerged was different between those found on potatoes and beans. On beans, it reached 6.1 individuals per leaf compared to 4.1 individuals per leaf on potatoes. As with the other infested leaves samplings, *O. chromatomyiae* dominated the parasitoid colony.

DISCUSSION

Looking to the damage intensity among treatments, the study proved that cyromazine and parasitoid could successfully reduced infestation of *L. huidobrensis*. Moreover, they worked better when they were integrated. In the case of cyromazine, the result was consistent with the laboratory study conducted by Prijono et al.[9] suggesting that even though cyromazine impacted less toxicity to adults, it effectively killed larvae of *L. huidobrensis* and *L. sativae* because cyromazine is a translaminar insecticide with larvae as the target. In this study, cyromazine was only applied three times, but with only cyromazine applied, the damage intensity was reduced by 52.2%, 46.4%, and 20.9% compared to the no treatment plot at the first, second and third samplings, respectively. Higher reduction of intensity occurred when cyromazine was integrated with parasitoid release, where the integration treatment reduced the intensity by 67.5%, 61.8%, and 26.2% at the first, second and third sampling respectively.

The study also suggested that application of cyromazine was relatively safe to the parasitoids indicated by the fact that the highest parasitism rate always occurred in the plot of parasitoid-cyromazine. This evidence was also consistent with the laboratory study conducted by Prijono et al.[9] which examined that cyromazine were not harmful to the adults of any kind of parasitoids. In this study, only two parasitoids were found, *O. chromatomyiae* and *H. varicornis.* As mentioned by Warsito,[7] the two parasitoids were the most dominant ones at the same altitude and crop (potato); both shared 88.4% and 11.6% respectively. However compared to this study, it seems that the parasitoid proportion has been changing by time, since the percentage of *O. chromatomyiae* found in this study increased to more than 90%. The common pattern of parasitoid colonisation is that the population of the specialist parasitoid (*O. chromatomyiae*) replaces and dominates the generalist ones (*H. varicornis*). Indeed, for those parasitoids that successfully colonize hosts as new invaders, the expectation is that generalists, which can switch hosts more easily, will initially be more successful and dominate parasitism of introduced pests, but then specialists which may be more efficient will increase and replace generalists. *Hemiptarsenus varicornis,* which was reported to be the most dominant species in Indonesia at the time of early infestation,[3] is a generalist parasitoid attacking not only *Liriomyza* sp., but also several other agromyzid genera such as *Chromatomyia*, *Ophiomyia* and *Phytomyza.*[12] Later *H. varicornis* seems to have been replaced by *O. chromatomyiae* which are more specialists, and their hosts are limited to leafminers from the genus *Liriomyza.*

However, releasing parasitoids only in one season may not have any significant impact on yield. As it is known, parasitoid work is relatively slow in reducing the population of targets and in this study, percentages of parasitism in all plots

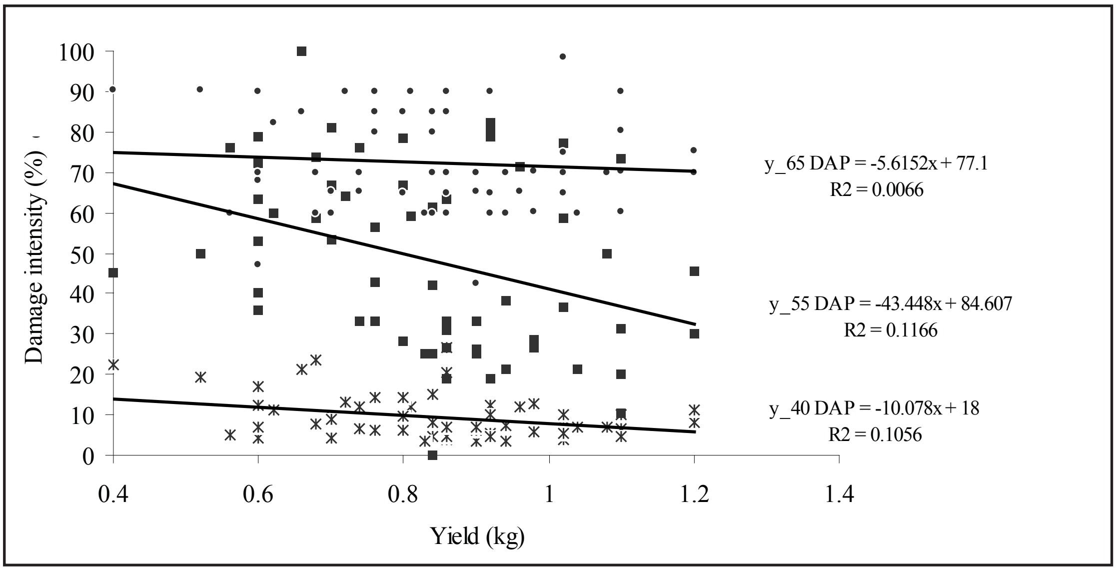

Figure 3. Linear regression of damage intensity and yield at different time observation.

were relatively low, especially in the week when the population caused damage which significantly reduced yield (early-mid vegetative growth). As stated by Warsito,[7] intensive survey in different altitudes showed that parasitoids did not significantly influence the distribution and abundance of leafminers. Thus there may be the need to assess other parasitoids of *O. chromatomyiae* and *H. varicornis* and their impact to the *L. huidobrensis* population.

Parasitoids prefer to parasitize *L. huidobrensis* attacking faba beans rather than potato. This may be caused by the fact that leaf features such as trichomes, which are found in potato, inhibit oviposition by certain parasitoids and reduce parasitism rate.[3] Based on this study, intercropping potato with faba bean was recommended to enhance parasitoids role in controlling for *L. huidobrensis.* Faba bean is a kind of reservoir for parasitoids which accelerates the population. Parasitoids parasitise *L. huidobrensis* attacking potato when the population of *L. huidobrensis* and parasitoids increase.

Impact of damage intensity to yield was shown on the regression analysis with every single data considered as a replication. Data suggests that damages to young plants (40 DAP and 55 DAP) significantly reduce the yield (F=6.85, df=1,58, *P*=0.01; 55 DAP : F=7.66, df=1,58, *P*=0.01; 65 DAP : F=0.39, df=1,58, *P*=0.54 for 40 DAP, 55 DAP and 65 DAP respectively). As mentioned by Parrella[1] that yield lost on some crops was greatest when mining occurred in young plants. Even though Parrella[1] also mentioned that it is difficult to accurately associate specific levels of mining activity with reductions in crop yield, from this study it clearly shows that the biggest index of yield reduction occurred at the 55 DAP observation (Figure 3).

CONCLUSION

In conclusion, cyromazine was effective to control the infestation of *L, huidobrensis* and it was unlikely harmful to parasitoid populations. Two dominate parasitoid found were *O. chromatomyiae* and *H. varicornis* in which both of them prefer to attack hosts on faba bean than potato. Moreover, infestations of *L. huidobrensis* in young plants reduced the potato yield.

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