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# Controlling *Spodoptera exigua* Using Parasitoid *Trichogramma japonicum* in Acacia Nurseries (*Acacia crassicarpa*) at Kerinci Central Nursery PT. Riau Andalan Pulp & Paper

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**ABSTRACT**

Armyworm (*Spodoptera exigua* Hubner.) is the main pest of acacia. Controlling such pests generally uses synthetic insecticides, which cause negative impacts such as they would resistance & resurgence, environmental pollution, residual effects, secondary pest explosions, and reduced biodiversity. We need an alternative in pest control, such as using parasitoid Trichogramma japonicum. This study aims to test the parasitism after the release of Trichogramma japonicum on Spodoptera exigua eggs in Acacia crassicarpa nurseries. The research was carried out at the Kerinci Central Nursery PT. RAPP on October 2019-March 2020. The research started with exploring, rearing and releasing parasitoid Trichogramma japonicum. The treatment was plots with and without releasing Trichogramma japonicum on Spodoptera exigua eggs for seven days of observation and analyzed using T-test at a 5% level. The significance of Trichogramma japonicum parasitized eggs in Spodoptera exigua eggs was 41.9% after releasing parasitoids in Acacia crassicarpa nurseries. The highest parasitization by Trichogramma japonicum was in the 3rd plot with an average parasitization of 53.8% and the lowest was in the 1st plot with an average 31.3% parasitization. Trichogramma japonicum that emerged from the total mass of eggs brought to the laboratory after parasitized eggs were 14.2%. Trichogramma japonicum showed significantly different results in reducing the egg population of Spodoptera exigua in the mother plant nursery Acacia crassicarpa.

**Keyword** ***:*** *Army worm (Spodoptera exigua Hubner), Spodoptera exigua, Trichogramma japonicum, Acacia crassicarpa, parasitoid*

## 1. INTRODUCTION

The well-known Industrial Plantation Forest Company (HTI) in Riau Province is PT. Riau Andalan Pulp and Paper (RAPP). PT. RAPP began operating in 1992 and started production in January 1995. The company has opened up job opportunities and new hopes for the people of Riau, especially areas within the factory and Industrial Plantation Forest (HTI), such as Pangkalan Kerinci, Pelalawan, Kuantan, Singingi and South Labuhan Batu Regency. PT. RAPP has 5 acacia nursery centers, including Kerinci Central Nursery 1 (KCN 1), Kerinci Central Nursery 2 (KCN 2), Baserah Central Nursery (BCN), Pelalawan Central Nursery (PCN), and Sei Kebaro Central Nursery (SKCN).

Pelalawan Regency, especially in the East Cross area of Pangkalan Kerinci, stands Kerinci Central Nursery 1 (KCN 1), one of the divisions of PT. RAPP, which operates in acacia nurseries, is tasked with providing seedlings for planting needs on the company's peatlands, where these acacia trees will become the raw material for paper production. KCN 1 can produce 50 million Acacia (Acacia crassicarpa) seeds yearly.

One of the woody plants used as raw material for pulp production and developed in PT RAPP's nursery is Acacia crassicarpa. Acacia crassicarpa wood belongs to broadleaf plants. Acacia crassicarpa is a tree that can reach a 30 m height, and its wood has can Acacia crassicarpa planted on peatlands has a potential of up to 110.2 m3/ha at the age of 4 years (Suhartati, et al., 2014).

Problems in monoculture forests can occur from various aspects, regarding silviculture, environment, and pests. Problems often reported as important issues in nurseries are economically detrimental pests. One of the main pests causing damage to Acacia crassicarpa plants through the results of pest monitoring in 2019 at Kerinci Central Nursery 1 PT Riau Andalan Pulp and Paper (RAPP) is the armyworm (Spodoptera exigua). The armyworm is a leaf pest that is important to note as the main pest of onion crops in Indonesia but is also found in acacia (Moekasan et al..,2012). Armyworm (Spodoptera exigua) attacks cause damage to acacia shoots and leaves. Damage to Acacia leaves is seen with symptoms of holes, bite marks until the leaves run out and the buds are damaged so that they are not suitable for harvesting. Based on the 2019 KCN 1 pest and disease monitoring report, the damage caused by the Spodoptera exigua pest ranges from 5-28% per month.

The use of synthetic pesticides is one of the commonly used control techniques so far. The synthetic pesticides application causes serious problems, such as emergence of cases of resistance, resurgence, death of natural enemies, the explosion of secondary pests, environmental pollution, residual effects, and reduced biodiversity (Georghious and Saito, 2012). Currently, alternative and effective pest control is needed, does not cause residues, and is friendly to the environment.

It is believed that pesticides in controlling pests will not immediately solve the problem. Integrated pest control is a concept with a multidisciplinary ecological approach utilizing a variety of control tactics with compatible natural enemies in pest control. It is necessary to investigate many forms of environmentally friendly control, such as biological control.

One form of biological control is egg parasitoids. Egg parasitoids are interesting biological control agents to look for, especially concerning developing alternative pest control technologies (Ahmad et al. 2012). The advantage of using egg parasitoids is that they can control pest populations early, eggs. Egg parasitoids such as Trichogramma sp. belonging to the Trichogrammatidae family have a very big opportunity to be utilized. Besides their ability to control pest populations early, they are also polyphagous, and easy to breed on alternative hosts.

Several countries use egg parasitoids of the Trichogrammatidae family as biological control agents for Lepidoptera pest populations. So far, approximately 15 species have been reported in Indonesia from the Trichogrammatidae family (Buchori et al. 2002). In Java, there are five reported species belonging to the genus Trichogramma family Trichogrammatoidea (Buchori et al. 2010). In Indonesia, the parasitoid family Trichogrammatidae has been used to control several pests, such as rice stem borer (Yunus, 2018) and some corn cob borers (Edi, 2018).

The results of Yunus' research (2018) show that Trichogramma japonicum in the field has successfully controlled the Scirpophaga incertulas population by releasing 250,000 parasitoids per ha. It has more effective results than releasing 125,000 parasitoids per ha. This increase in parasitoid release can increase the parasitization by 236% compared to no parasitoid administration.

This research studied a biological control: Trichogramma japonicum as a parasitoid from Spodoptera exigua eggs and their level of parasitism. This effort is expected to reduce pest populations and damage caused by Spodoptera exigua in acacia nurseries in KCN 1. This study aimed to obtain the parasitic ability of Trichogramma japonicum release against Spodoptera exigua eggs that were integrated into an integrated control strategy in Acacia crassicarpa nurseries.

## 2. MATERIALS AND METHOD

The research was carried out at Kerinci Central Nursery (KCN 1), one of the divisions of PT. RAPP is engaged in acacia plant nurseries, especially in the eastern cross area of Pangkalan Kerinci City, Pelalawan Regency. Research was continued at PT RAPP.'s Entomology Research and Development (R&D) Laboratory. The average temperature and humidity in the laboratory are 27-28°C and 75-80%, and 27-40ºC and 60-70% in the mother plant house (MPH).

The materials used in this study were Acacia crassicarpa seeds, Spodoptera exigua eggs found in nurseries and species identification confirmed by the

Molecular Laboratory of PT. RAPP, parasitoid Trichogramma japonicum, 10% distilled water Arabic gum solution, and Corcyra cephalonica egg.

The tools used in this study were paperboards or flat cardboard, label paper, sand beds, stereo microscope, cameras, markers, plastic containers, plastic sacks, masking tape, tweezers, scissors, killing bottles, lups, label paper, organza bags , tissue rolls, cloth, camera, stationery, data book, 20 mesh filter, PVC tube, glass tube, glass container, UV chamber, and stapler.

The implementation of the research consists of several stages, namely:

**a. *Trichogramma japonicum***
**Exploration**

The parasitoid Trichogramma japonicum was collected in Bunga Raya, Siak from corn, rice and sugar cane through a survey using a random sampling method. This method is carried out in three stages. The first is to collect various types of egg parasitoids that are likely to parasitize Spodoptera exigua eggs. Manual collection of parasitoids by collecting stem borer eggs and Spodoptera spp. found in maize, rice, and sugar cane.

The second stage of parasitoid collection was carried out manually in corn, rice and sugarcane plantations using Corcyra cephalonica and Spodoptera exigua egg traps. The initial step begins with exposition of cardboard containing Corcyra cephalonica eggs as host traps on maize, rice and sugarcane (3 paperboards egg traps per plant species) for 24 hours followed by the same method exposition of Spodoptera exigua eggs as host traps (3 paperboards traps eggs per plant species) in maize, rice, and sugarcane for 24 hours.

All eggs were collected and put into petridishes for incubation in the laboratory. Eggs parasitized in the field will appear gray to black in color. After 5-7 days of incubation, parasitoids will appear from the parasitized eggs, and then a compatibility test is carried out on Spodoptera exigua eggs we get from the nursery.

The next third stage is netting insects around corn, rice and sugarcane plants to collect possible parasitoids caught using entomological nets.

After collecting the parasitized eggs and trapped parasitoids, they were manually taken to the laboratory to identify the type of parasitoid and to carry out parasitization tests on the pest Spodoptera exigua.

**b. Rearing *Trichogramma japonicum***

After Trichogramma japonicum was found, rearing was carried out at the Entomology Laboratory. Rearing of Trichogramma japonicum in the laboratory is carried out at a temperature of 27-280C with 75-80% humidity using an alternative host Corcyra cephalonica then develops inside the host's egg and then leaves the body of the host Corcyra cephalonica.

The process of rearing Trichogramma japonicum starts with rearing Corcyra cephalonica as an alternative host in a plastic container (36 x 27.8 x 7.3 cm) filled with media mixed with damaged corn with purified chicken and rice bran (2:1:2). The eggs of Corcyra cephalonica are spread on the stored media. to become larvae and pupae. After reaching the imago stage,

they are collected manually using a glass tube and transferred to a mating/oviposition container (the PVC tube is covered with a net). After Corcyra cephalonica lays eggs, the eggs are collected in petridishes and labeled with the egg harvest time and the storage start date. Corcyra cephalonica eggs are then stored at 4-80C so that the eggs can last for about 6 months.

After the Corcyra cephalonica host egg is available, then provide a paperboard with an information label on the date of rearing Trichogramma japonicum and apply glue (10% distilled water Arabic gum solution) then slowly strain the Corcyra cephalonica host egg on the paperboard that has been smeared with Arabic gum glue then put it in a glass container ( glass containers). After that, incubate in UV chamber for one hour to sterilize Corcyra cephalonica embryos. After leaving the UV chamber, put the paperboard with the parasitic Corcyra cephalonica eggs and Trichogramma japonicum imago in the glass container for 3-4 days. Parasite eggs will be visible on the 5th to 7th day after parasitization begins. Trichogramma japonicum will appear after the eggs are black on the 8th or 9th day and are ready to be released to the nursery (Figure 1).

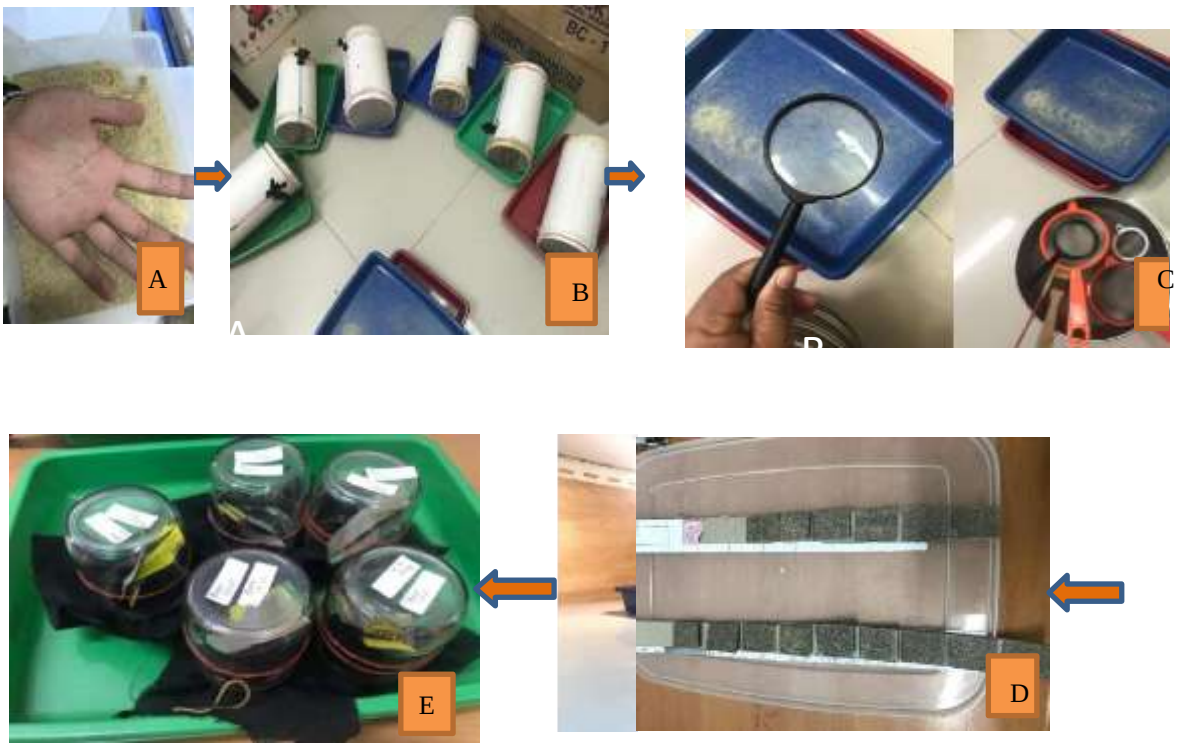


**Figure 1.** The rearing process of Trichogramma japonicum starts from rearing Corcyra cephalonica in a mixed medium of damaged corn with chicken pur and rice bran until they become larvae, pupae and imago (A), transferred in mating/oviposition tube (B), collecting Corcyra cephalonica host eggs ready to be filtered on paperboard Arabic glue smeared (C) paperboard with host eggs ready for sterilization (D) paperboard host eggs put in a glass container and sterilized in a UV chamber (E) paperboard with adult Trichogramma japonicum put in with new paperboard containing Corcyra cephalonica eggs in a glass container ( F) and parasitized Corcyra cephalonica host eggs by Trichogramma japonicum ready to be released to nurseries (G).

**c. Pelepasan *Trichogramma japonicum***

Before release, a survey was carried out on the location of the mother plant, where many eggs of the pest Spodoptera exigua were found. The Acacia crassicarpa plant, where the first natural egg cluster of Spodoptera exigua was found is labeled as number 3. The two beds to the left of the first bed are marked (Bed 1 & 2) and the two to the right (Bed 4 & 5) are also marked. All natural egg groups found in beds 1, 2, 3, 4, and 5 were searched and the plants found in egg groups were labeled. One cardboard containing 5000 Corcyra cephalonica eggs which have been parasitized by Trichogramma japonicum is clamped per area marked (one area = five beds) on the plants in the middle of the beds 3. Control treatment is five beds, at least one group of eggs is found without Trichogramma release japonicum.

Evaluation of the number of parasitized Spodoptera exigua egg groups was carried out every day for 7 days (3 days in the nursery followed by 4 days in the laboratory) using a magnifying glass after Trichogramma japonicum was released

Distinguishing parasitic and non-parasitic eggs can be seen from the color, each has a black color if it is parasitic and white if it is non-parasitic. If the egg is silver it means it was recently parasitized. The area of the release area for Trichogramma japonicum was 5 beds x 24 cm x 1 cm, i.e. 120 cm2 each in the observation plot where natural eggs of Spodoptera exigua were found (Figure 2)

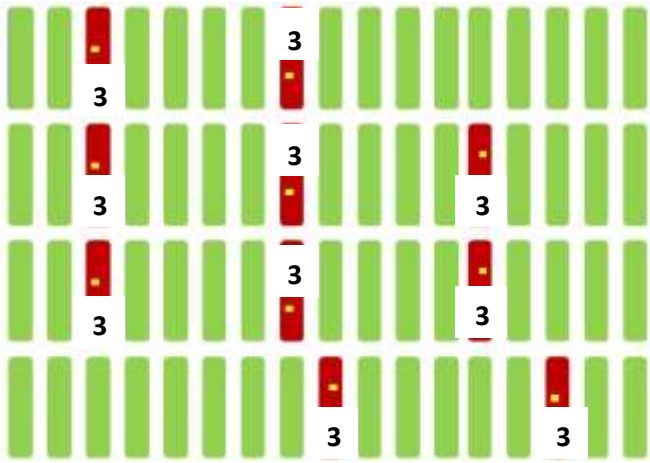


**Figure 2.** Lay out of Trichogramma japonicum release experiment

**d. Pengamatan**

Adapun parameter yang diamati:

1. Number of parasitic eggs of Spodoptera exigua

The eggs of Spodoptera exigua collected in the field were counted, and the number of pest eggs per observation plot and the number of Spodoptera exigua eggs parasitized by the parasitoid. Evaluation of the number of parasitized Spodoptera exigua egg groups was carried out after the cardboard was clamped at the study site for 7 days (observations were made 3 days in the nursery followed by the next 4 days in the laboratory) by differentiating eggs that were not and parasites. The way to distinguish them can be seen from the color, each has a black color if it is parasitic and white if it is not parasitic which is then counted in the entomology laboratory. If the egg is silver, it has recently been parasitized

2. Number of Trichogramma japonicum emerging from parasitic eggs

Spodoptera exigua eggs that have been collected are stored in petridishes separately, labeled, and brought to the laboratory and then maintained in a room with a temperature of 27 ± 2 °C and RH = 70–80%. On the fourth day, Trichogramma japonicum parasitoids quantity emerged from the parasitized eggs was observed to see the parasitic level until the 7th day of observation..

3. Parasitization level of Trichogramma japonicum

The percentage of egg mass parasitization per plot was calculated after the seventh day. Parasitism (%) of egg mass per plot by dividing the number of egg groups parasitized by the number of egg groups evaluated in the plot multiplied by 100 percent. Percentage of parasites and Trichogramma japonicum emerging from parasitized Spodoptera exigua by counting the number of parasitized eggs and Trichogramma japonicum emerging from parasitized host eggs by (divided by) the total number of eggs multiplied by 100 percent (Tang et al. 2017*).*

To see the difference in the release of Trichogramma japonicum to Spodoptera exigua eggs in the Acacia crassicarpa nursery, it was analyzed using the t-test (T-test) at the 5% level. To be more accurate it is done with the SPSS version 19 program.

## 3. RESULT AND DISCUSSION

### Types of parasitoids collected from agricultural land

The results of a survey conducted in Bunga Raya, Siak on corn, rice, and sugarcane plants found species capable of

parasitizing the egg masses of Spodoptera exigua and the host eggs of Corcyra cephalonica traps, namely Trichogramma japonicum.

Parasitoid Trichogramma japonicum collected from agricultural land according to reference from Yunus (2005), the morphological characters of Trichogramma japonicum are: brownish yellow imago, red eyes and black thorax. Imago length 0.4 - 0.5 mm with a head width of 0.17 - 0.21 mm. The female antennae are club-shaped and the male antennae are straight. The forewings have trichia Rs1 of 7-10 and the edges of the wings are covered with hairs. The duration of the parasitoid life cycle in the pre-imago phase was 8.2 + 0.3 days, the imago phase was 1.8 + 0.7 days and the total life cycle was 10.0 + 0.8 days. The parasitoid imago emerges from the host egg by breaking through the chorion. As soon as the parasitoid is outside the host's body, the imago immediately looks for a partner and marries (Figure 3).

**Figure 3.** Trichogramma japonicum parasitoid
(Source: Research documentation, 2020)

After the parasitoid Trichogramma japonicum was screened in the laboratory, a parasitation test was carried out on the eggs of Spodoptera exigua in the nursery. Natural eggs of Spodoptera exigua can be found in the main house of the MPH 2 plant block D KCN 1, so this location is the release site for Trichogramma japonicum for field parasitation tests.

**Egg mass number of parasitized Spodoptera exigua by Trichogramma japonicum**

Egg mass of Spodoptera exigua in acacia nurseries with the release of parasitoids, generally parasitic Trichogramma japonicum. This species was found to parasitize the egg mass of Spodoptera exigua starting three days after the first day of release of Trichogramma japonicum at the mother plant acacia house. The results of calculating the mass number of Spodoptera exigua parasitized eggs by the parasitoid Trichogramma japonicum (black eggs) can be seen in Table 1.

Table 1. Total mass of eggs of Spodoptera exigua parasitized by Trichogramma japonicum.

| Observation | D-4 | D-5 | D-6 | D-7 |
|---|---|---|---|---|
| Spodoptera exigua egg mass | 93.0 | 93.0 | 93.0 | 93.0 |
| Number of parasitic eggs | 23.0 | 36.0 | 39.0 | 39.0 |
| *Parasitism* (%) | 24.7 | 38.7 | 41.9 | 41.9 |

Table 1. shows the percentage of egg mass of Spodoptera exigua parasitized by Trichogramma japonicum. Spodoptera exigua pest eggs began to parasitize 3 days after the release until the 7th day of observation. The parasitized Spodoptera exigua eggs percentage by Trichogramma japonicum 3 days after release reached 24.7% (4th day). Then on the 5th day, it increased to 38.7% then the number of parasitized eggs also increased on the 6th day to 41.9%, but there was no increase in parasite egg mass on the 7th day. This is supported by Yunus' research (2005) which states that as soon as the parasitoid is outside the host's body, the imago immediately looks for a partner and mated. Oviposition activities are carried out by female imago to get a suitable host egg.

Oviposition/parasitization activities on the first day after mating can reach 15-30 times per female imago. On the second and following days, there were only a few oviposition activities left, namely between 1-5 times or not doing oviposition at all and then the imago died immediately. Parasite eggs 2-3 days later are marked by the color of the eggs turning black or silver. The time needed by the Trichogramma spp. to produce one generation generally ranges from 1–2 days after the eggs turn black as stated by Nurindah (2016) (Figure 4)

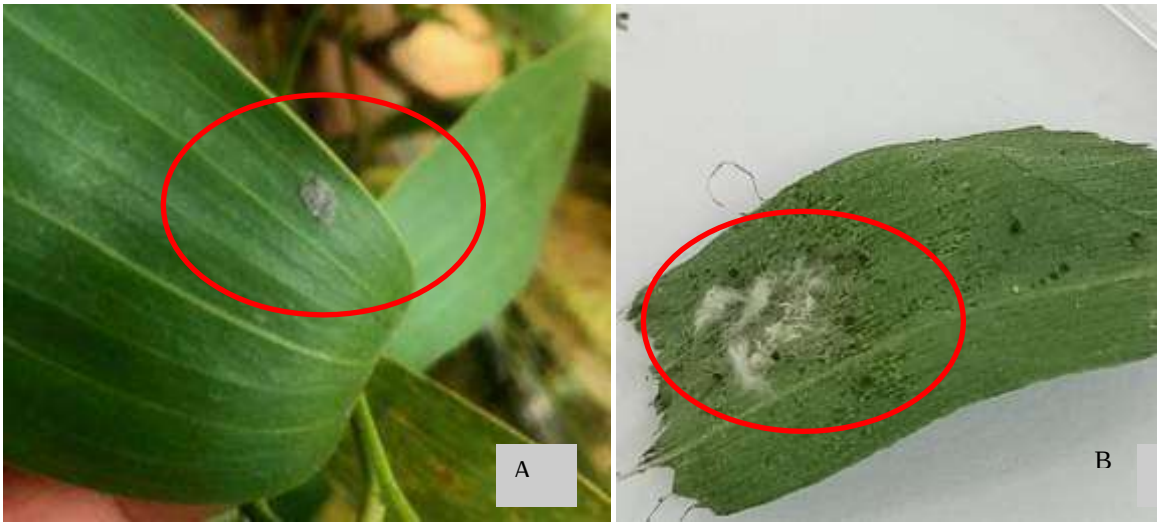


**Figure 4.** Blackish Spodoptera exigua egg masses parasitized by Trichogramma japonicum in the nursery on the 3rd day of observation (A), parasitized Spodoptera exigua eggs on the 5th day of observation in the Entomology Laboratory (B) and emergence of the parasitoid Trichogramma japonicum from Spodoptera exigua eggs (C).

Observational plots (5 plots = 120 cm2) found that the average parasitic mass of Spodoptera exigua eggs released by parasitoids was higher than without releasing parasitoid Trichogramma japonicum (Table 2).

Table 2. After seven days of observation, the egg mass of Spodoptera exigua parasitized by Trichogramma japonicum in plots (pias).

| Treatment | Control | 1st Pias | 2nd Pias | 3rd Pias | 4th Pias | 5th Pias |
|---|---|---|---|---|---|---|
| Average parasitism per plot (%) | 0,0 | 31,3 | 42,6 | 53,8 | 42,8 | 33,2 |

Table 2 shows that there is a significant effect on the level of pest parasitization after releasing Trichogramma japonicum (1 paper pias = ± 5000 Trichogramma japonicum) compared to without releasing parasitoids. The highest parasitization by Trichogramma japonicum was in the 3rd pias plot with an average parasitation of 53.8% and the lowest was in the 5th pias plot with an average parasitation of 31.3%..

Some of these differences can be caused by Spodoptera exigua which was selected by the parasitoid Trichogramma japonicum, the ability of the parasitoid to walk or fly to the location of the egg mass of Spodoptera exigua and the density of the host parasitoid and the female population in it (Pabbage and Tandiabang, 2011) .

Parasitization of Spodoptera exigua eggs by the parasitoid Trichogramma japonicum in this nursery is quite high, with an average of 31.3-53.8% per observation plot. The same study's results were also carried out by Khan et al (2020) on acacia, the average Spodoptera spp. the parasite Trichogramma yousufi can reach 24.79 - 40.82%. These results indicate that the parasitoid Trichogramma japonicum is a potential biological agent in controlling Spodoptera exigua in acacia nurseries.

**Number of Trichogramma japonicum that emerged from the mass of parasitized S.exigua eggs.**

The results of observations in the laboratory on day 4 to day 7 for the number of Trichogramma japonicum emerging from the parasitic Spodoptera exigua egg mass, can be seen in Table 3.

Table 3. Number of Trichogramma japonicum emerging from the parasitic Spodoptera exigua egg mass.

| Observation | Day-4 | Day-5 | Day-6 | Day-7 |
|---|---|---|---|---|
| *Egg mass Spodoptera exigua* | 3160 | 3160 | 3160 | 3160 |
| *Trichogramma japonicum* muncul | 253 | 421 | 451 | 451 |
| *Trichogramma japonicum* (%) | 8,0 | 13,3 | 14,2 | 14,2 |
| T-test test scores = 0.025 | | | | |
| (P<0.05) | | | | |

Table 3 shows that Trichogramma japonicum that emerged from the total mass of eggs at the study site after parasitized eggs was 8.0% on day 4, then increased to 13.3% on day 5 and then on day 6 and day 3 -7 observations increased to 14.2%. Some eggs were blackened, but Trichogramma japonicum did not appear during the 4th to 7th day of observation. Several Trichogramma japonicum may have already hatched from these eggs in the field, parasitizing natural eggs around the plot before being collected and brought to the laboratory and other environmental factors while in the field. Maharani (2009) stated that the high and low percentages egg parasitoids level are caused by environmental factors affecting the development of parasitoids. Environmental factors (external factors) play an important role in determining parasitoid species' high or low population.

Altitude is very closely related to the effect of air temperature, the higher a place above sea level, the lower the air temperature, making it more difficult for

insects to reach their hosts. The results of the calculation of the t test (T-test) from the release of Trichogramma japonicum in acacia nurseries significantly affected the number of parasitic egg masses. The t test (T-test) results showed that Trichogramma japonicum significantly different results in reducing the egg population of the pest Spodoptera exigua in the mother plant nursery compared to no release of Trichogramma japonicum. Ahmad et al (2012) stated that this parasitoid (Trichogramma spp.) is a proven biocontrol agent because it kills pests at the most critical stage (eggs) before damage occurs (Goebel et al., 2010).

## 4. CONCLUSION

The significance of the eggs of the parasitic Trichogramma japonicum on the eggs of Spodoptera exigua is 41.9% from the results seven days of observation after the release of parasitoids which is integrated into an integrated pest control strategy in the Acacia crassicarpa nursery.

The highest parasitization by Trichogramma japonicum was in the 3rd pias plot with an average of 53.8% parasitization and the lowest was in the 1st pias plot with an average of 31.3% parasitation. Trichogramma japonicum that emerged from the total mass of eggs brought to the Laboratory after the black parasitic eggs were 14.2%. Trichogramma japonicum showed significantly different results in reducing the egg population of the pest Spodoptera exigua at the mother plants of Acacia crassicarpa nurseries.

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