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Laboratory Evaluation of Predation Potency *Euborellia Annulata* (Dermaptera: Anisolabididae) on Larvae of *Spodoptera Frugiperda* (Lepidoptera: Noctuidae), a Pest of Maize Crops

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ABSTRACT

Spodoptera frugiperda is an invasive pest found attacking corn plants in Indonesia in 2019. The pest attacks during the vegetative and generative periods and can cause heavy crop damage. Biological control by utilizing predators is an environmentally friendly control method. *Euborellia annulata* is a predator found in maize crops. This study was conducted to determine the predation potency of *E. annulata* on *S. frugiperda* larvae. This study arranged as a completely randomized design (CRD) with 5 treatments, 1 male *E. annulata* adult, 1 female *E*. *annulata* adult, 2 male *E. annulata* adult, 2 female *E. annulata* adult and 1 pair of male and female *E. annulata* adult. Ten *S. frugiperda* instar 3 larvae were placed in the treatment containers, then predator *E. annulata* was placed into the containers according to the treatment. Variables observed were the number of *S. frugiperda* larvae preyed and the predation behavior of *E. annulata*. The results showed that at 4 Days After Treatment (DAT) there was a significant difference in the predation ability of *E. annulata* on *S. frugiperda* larvae. At 5 DAT, 2 female *E. annulata* showed greater predation ability at 66% compared to treatment 2 male *E. annulata* at 52%. However, 2 female *E. annulata* showed the similar prey ability as 1 pair of male and female adult at 7 and 8 DAT with a mortality range of 96-100% of *S. frugiperda*. Predators with the female have a large prey ability and are more aggressive than the male. Morphologically, it is known that the forceps of female predators are larger than males, so that female predators are faster to prey on *S. fungiperda*. Female predators and have better prey potency as biological control agents for *S. fungiperda* pests in maize crops.

Keywords: *Predation potency, Euborellia annulata, Spodoptera frugiperda, biological control, Zea mays*

1. INTRODUCTION

Maize (*Zea mays* L.) is one of the second important food crop commodities after rice and is found in almost all parts of Indonesia. Maize is a strategic role in agricultural and economic development. This commodity contributes to provide of various foodstuffs and industrial raw materials (Balai Proteksi Tanaman Pangan dan Hortikultura. 2019).

The most commonly problem found in the field is the attack of *Spodoptera frugiperda* or Fall Armyworm (FAW). This pest is an invasive pest in Indonesia, first detected in 2019 in West Sumatra and now the pest has spread to various regions in Indonesia. *Spodoptera frugiperda* larvae cause damage maize plants to a severe attack level. It can destroy maize plants if the larvae damage the shoots, young leaves or growing points of the plant. In some locations, yield loss has reached 94% (Assefa et al., 2019; Nonci et al., 2019). *Spodoptera frugiperda* larvae also attack newly formed cobs and continue to the growing point of the plant. Usually, only 1-2 larvae are found in one plant because the larvae are cannibals. Damage to the plant is usually characterized by larval burrow marks, where there is a coarse powder resembling sawdust on the upper surface of the leaves, or around the top of the corn plant which is larval excrement (Trisyono et al., 2019).

Controlling of *S. frugiperda* can be done mechanical method by finding and killing the larvae and eggs mechanically (crushing by hand). But farmers generally practice chemical control using insecticides with the active ingredients deltamethrin, emmamectin benzoate, siantranilliprol and spinetoram (BBOPT, 2020). The use of insecticides in pest control is a short-term control that can rapidly reduce pest populations, and it can overcome the spread of pests immediately. However, the use of insecticides has several negative impacts such as killing non-target organisms, environmental pollution, residual effects on agricultural products, and increased production costs. Long-term use of insecticides can lead to pest resistance (Swastika et al. 2022). For this reason, it is necessary to promote biological control with the concept of sustainable agriculture through the utilization of natural enemies. The use of natural enemies to control pests does not have a detrimental impact on both humans and the environment. Natural enemies can reduce the population of insect pest by utilizing biological agents such as predator (Deole and Paul, 2018, Firake and Behere, 2020). In addition, it is not requiring considerable costs, the use of natural enemies is also an environmentally friendly action and does not cause adverse effects on the environment compared to the use of pesticides (Untung, 2006).

Euborellia annulata is one of the predators that appear a role in controlling pests in plants. Yasin et al. (1999) reported that *E. annulata* has a high prey ability against corn stalk borer *Ostrinia furnacalis*. Hadad et al. (1999) also reported that *E. annulata* has the ability to prey on coconut bud borer *Brontispa longissima* and found that female predators have a higher prey ability than males. Research by Asmanizar et al. (2023) reported finding the predator *E. annulata* preying on *S. frugiperda* larvae in corn plantations attacked by *S. frugiperda* in Deli Serdang, Langkat, Karo and Dairi districts. This predator has the potential to be used as a biological control agent to control *S. frugiperda* infestation because the flat body of the predator can reach the larvae in the burrows of infested maize plants. Therefore, it is necessary to know the prey ability of *E. annulata* on *S. frugiperda* larvae under laboratory conditions to determine its potential as a biological control agent for *S. frugiperda* pests.

2. MATERIALS AND METHODS

This research was conducted at the Plant Protection Laboratory (29±2^{o}C and 80±10% R.H.) Faculty of Agriculture, Universitas Islam Sumatera Utara, Medan from September 2022 to March 2023. This study arranged as a completely randomized design (CRD) with 5 treatments, 1 male *E. annulata*

adult, 1 female *E*. *annulata* adult, 2 male *E. annulata* adult, 2 female *E. annulata* adult and 1 pair of male and female *E. annulata* adult their ability in predating *S. frugiperda* larvae. There were five replications for each treatment. The data were analyzed by one way ANOVA. Means were separated using DMR Test at P=0.05 when the ANOVA was significant.

Preparation of *Euborellia annulata* predators

Euborellia annulata adults were collected from corn plants infested with *S. frugiperda* in Batang Kuis District, Deli Serdang Regency, North Sumatra (Latitude: 3.58956; Longitude: 98.81044). Rearing the *S. frugiperda* was done in the Laboratory. *Euborellia annulata* adult were put into plastic jar containers measuring 27 cm long, 19 cm wide and 8 cm high. Each container was filled with several male and female adult. Then the container was given soil and sand in a ratio of 3:1 to a height of 2 cm and sprayed with water using a hand sprayer to maintain media moisture. *Euborellia annulata* predators were fed with kibble pellets every day.

Preparation of *Spodoptera frugiperda* larvae

Spodoptera frugiperda larvae were collected from corn plants infested with *S. frugiperda* in Batang Kuis District, Deli Serdang Regency, North Sumatra (Latitude: 3.58956; Longitude: 98.81044). Rearing the *S. frugiperda* was done in the Laboratory. The larvae were put into plastic containers (6 cm in diameter and 5 cm in height). Each container contained one *S. frugiperda* larva and was fed with young corn leaves. The new emerged adults were put together in a glass container (60x40x40 cm) and fed with 10% honey dripped on cotton and hung in the container. Eggs laid by female insects hatched after 4-6 days, and first instar larvae are separated in another container and fed with young corn to become third instar larvae as test larvae.

Figure 1. *S. frugiperda* before exposing against *E. annulata*

Predatory ability study on *Spodoptera frugiperda*

Prior to the predation study, *E. annulata* predators were no fed for ± 24 hours. Then, ten *S. frugiperda* instar 3 larvae were placed in a plastic jar (length of 27 cm, width of 19 cm and height of 8 cm) then the predator *E. annulata* was placed into the container and covered with a piece a muslin cloth.

Variables observed

a) Predation ability by counting the number of larvae that were preyed upon and died each day starting with the joining of *E. annulata* predators with *S. frugiperda* larvae and then on subsequent days until all larvae were preyed upon. Predation ability by calculating the percentage of larval mortality with the formula:

% mortality of *S. frugiperda* larvae = $\frac{number\ of\ dead\ larvae}{number\ of\ tested\ larvae} \times 100\%$

b) The predation behavior of predator *E. annulata*. Observation of *E. annulata* behavior was done before preying, during preying and after preying *S. frugiperda* larvae.

c) Physiological characteristics of *S. frugiperda* that are preyed. The symptoms of *S. frugiperda* larvae dying due to predation were observed on their body parts.

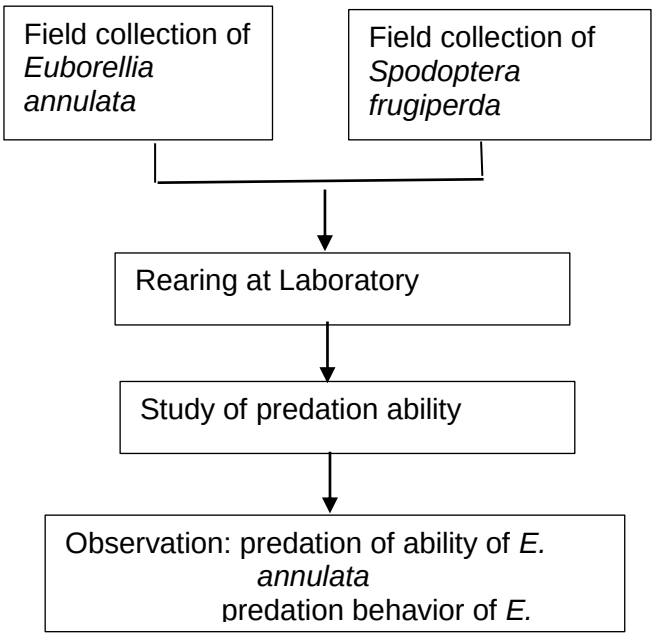

Figure 2. Research flow diagram

3. RESULTS AND DISCUSSION

a) Predation ability

The predation ability of *E. annulata* on *S. frugiperda* larvae can be seen in Table 1.

Table 1. Predation ability *E. annulata* on *S. frugiperda* larvae

Perlakuan	Percentage Mortality of *S. frugiperda* (DAT) ± SEM								
	1	2	3	4	5	6	7	8	9
P1 (♂)	2±2.00	16±3.99	26±3.99	34±5.09 b	44±5.09 c	58±5.82b	68±3.74 b	76±3.99 b	92±2.00 b
P2 (♀)	4±2.45	14±2.45	26±3.99	40±5.47 ab	48±5.82 c	64±7.47 b	70±5.47 b	78±3.74 b	94±2.45 b
P3 (♂♂)	6±3.99	16±3.99	28±4.89	38±4.89 ab	52±3.74 bc	58±3.74 b	68±3.74 b	78±3.74 b	100±0.00 a
P4 (♀♀)	10±0.00	22±2.00	38±2.00	52±3.74 a	66±2.45 a	76±3.99 a	86±3.99 a	96±3.99 a	100±0.00 a
P5 (♂♀)	2±2.00	14±2.45	26±2.45	50±3.16 a	62±2.00 ab	72±2.00 a	90±0.00 a	100±0.00 a	100±0.00 a

Letters followed by the similar letter in column indicates no significant difference at the 5% level according to the DMRT test.
DAT = Day After Treatment
SEM = Standart Error of Mean

Observations of 1-3 DAT treatment has not affected the predation ability of *S. frugiperda* larvae. However, at 4 - 9 DAT, there were significant differences in the predation ability of *E. annulata* predators against *S. frugiperda*. At 1 DAT predation by predators was not very aggressive, because predators were adapting and recognizing prey, so it took time for predators to prey on pests. Research by Nurmaisah (2022) found that the process of predation on pests begins with the discovery of prey carried out by male or female *E. annulata* predators by moving irregularly. This behavior is classified as pre-adaptation to the new environment. When prey is found, the predator pauses to look for prey suitability. If it is not suitable, the predator will leave the prey. After finding suitable prey, the predator touches and moves the prey with its mouth. Prey that did not defend immediately attacked by the predator.

Based on the results of the research conducted, it is known that the greater percentage of *S. frugiperda* mortality due to *E. annulata* predation in the P2 treatment (1 female adult *E. annluata* predator), P4 (2 female adult *E. annulata* predators), and P5 (1 pair of male and female *E. annulata* predators). This indicates that female *E. annulata* predators have a more active prey drive. The predatory behavior of *E. annulata* can be seen in its movement speed, greater strength, and a strong forceps.

Female adult predators have larger forceps than males, making it easier to clamp their prey. According to Adnan and Handayani (2010) that *E. annulata* is a predator that has straight and strong forceps that are used to defend itself and to hold its prey. This is in accordance with the results of Sirait's research (2021) which states that the prey ability of female *E. annulata* to *S. frugiperda* larvae is greater (15; 45; 75 and 100%) compared to male adult (0; 15; 35 and 55%) in observations of 1, 2, 3, and 4 DAT.

Generally, during the observation, female predators showed higher prey potential compared to male predators, although they did not show significant differences. Two female predators and two female and male predators showed higher predation ability (50-52%) and significantly different from the predation ability of one male predator (34%) at 4 DAT. A consistent increase in predation ability occurred until 9 DAT and the number of 2 predators had reached 100% predation ability against *E. annulata*.

b) **Predation behavior of *E. annulata***

The predation behavior process of male and female *E. annulata* adults before preying starts from searching by moving irregularly around the prey for about 2-5 minutes. This behavior shows the adapting to a new environment. When *E. annulata* has found its prey, predators spend a moment recognizing the prey, then some leave the prey to look for other prey**.**

The predation behavior process of *E. annulata* during prey is characterized by the position of *E. annulata* around the prey, then predating with several movements, namely the position of the caput which is lowered and then raises the abdomen higher than the body while moving the antenna actively. It then feeds on the liquid contents inside the body (abdomen) of *S. frugiperda* larvae from one direction. After feeding, *E. annulata* cleans its mouth by moving its forelimbs over its mouth.

The predation behavior process of *E. annulata* after preying on *S. frugiperda* is characterized by releasing *S. frugiperda* from the pincers and continuing to eat the body of its prey. While feeding on the body of its prey, *E. annulata* can also use its pincers to catch other pests that touch its body. *Spodoptera frugiperda* that has been preyed upon by *E. annulata* is characterized by blackening of the skin and shrinking of the larval parts of *S. frugiperda*. This is because the body fluids of *S. frugiperda* are destroyed by *E. annulata* using its mouthparts.

Predation behavior in each treatment condition can be explained as follows. The of P1 (1 male adult) *E. annulata* seemed to take longer to find prey because males do not need much food for energy in view of the data P1 is very lacking in prey ability and here it is clearly seen that P1 predated on 1 *S. frugiperda* larva, characterized by symptoms of blackening and wrinkling of the larval parts of *S. frugiperda*. while the other 9 were not predated.

The P2 treatment (1 female adult) of *E. annulata* seemed better than the P1 treatment (1 male predator) because of the predatory ability of P2 was more active due to the body size of the female adult which was longer than the male adult. Whereas female adult is more active when compared to male adult and female adult has longer pincers than male adult so that female adult is stronger than male adult. The P3 treatment (2 male predators) of *E. annulata* clearly shows that its predatory ability is slow to find prey, and when preying, *E. annulata* does not finish biting *S. frugiperda* larvae so that the liquid in the larval body is not finished being bitten and is characterized by shrinking or blackening of the body of *S. frugiperda* larvae and is very clear in the P2 and P4 treatments**.**

The P4 treatment (2 adult females) of *E. annulata* seemed more active as indicated by the data at 4 DAT reaching 50% and at 8 DAT reaching 100%, and there were no larvae that were not consumed and the larvae were

characterized by shrinking, blackening and drying of the body of *S. frugiperda* larvae. Treatment P5 (1 pair of adult males and females) of *E. annulata*, clearly showed the difference between males and females was stronger prey power of females because the females were earlier to prey on *S. frugiperda* larvae and when the larvae were dead, male predators joint and participated in preying on larvae that had been paralyzed by female predators. Female predators were able to prey on more larvae than male predators. This is thought to be because female predators are more active than male predators, which tend to remain motionless and only move when hungry, resulting in fewer larvae being preyed upon. During application, male and female mating was observed and occurred several times. Field surveys show that the predator *E. annulata* was found on corn plants infested with *S. frugiperda* (Asmanizar et al., 2023). Its presence can reach *S. frugiperda* larvae that are already in burrows. The results of this study indicate that this predator has the potential to prey on *S. frugiperda* larvae and can be used as one of the methods in the Integrated Pest Management method to control *S. frugiperda* pest attacks. Predation conditions are shown in Figures 1 and 2.

Figure 3. *E. annulata* behavior during the preying of *S. frugiperda*
(a). *E. annulata* predator pinching *S. frugiperda* larvae, (b). Predator *E. annulata* feeding *S frugiperda* larvae

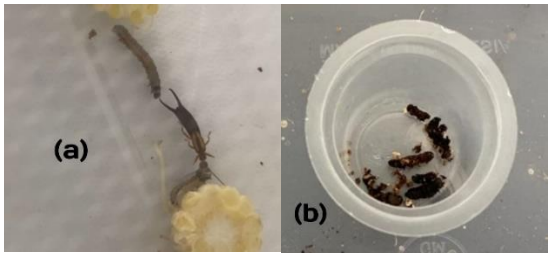

Figure 4. *E. annulata* behavior after preying of *S. frugiperda.*
(a). *E. annulata* Predator approaching *S. frugiperda* larvae,
(b). *S. frugiperda* larvae that have been preyed upon by *E. annulata*

c. **Physiological characteristics of *Spodoptera frugiperda***

Physiological characteristics of *S. frugiperda* larvae that die after being bitten by *E. annulata* predators are characterized by blackening and wrinkling of the larvae and 2-3 days of drying. Symptoms of dead prey (larvae) attacked by predators are generally brownish at first then blackened. The death of *Setothosea asigna* larvae attacked by the predator *Sycanus annulicornis* with symptoms that are slightly brownish and slightly wrinkled and within one day the body of *S. asigna* larvae that have been predated are blackish brown. This is because the body fluids (haemolymph) of *S. asigna* are sucked by *S. annulicornis* using its stylets (Pratama, 2021). Similarly, the predator *E. furcellata* attacks *Setora nitens* larvae by puncturing and sucking their prey until the liquid is lost and the

larvae of *S. nitens* appear shriveled and dry (Gani et al. 2019).

4. CONCLUSION

a) Female *E. annulata* predators have greater prey ability than male *E. annulata*.
b) Predation potential of two female *E. annulata* predators was greater than two male predators on *S. frugiperda* larvae.
c) Female predators are more active in the predation process and can prey on large numbers of *S. frugiperda* larvae.
d) The predator *E. annulata* has the potency to be developed as a natural enemy to control *S. frugiperda* in maize plants.

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