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Evaluation of the Predation Capacity of *Rhynocoris albopilosus* (Heteroptera: Reduviidae) on the Second Instar Larvae of *Spodoptera frugiperda* (Lepidoptera: Noctuidae)

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Abstract: The fall armyworm (CLA) *Spodoptera frugiperda* is currently the main pest of cereal crops, with a preference for maize. Chemical control of this pest has shown its limitations. Biological control based on the use of the predatory insect *Rhynocoris albopilosus* is considered in this study. To this end, the predatory capacity of *R. albopilosus* adults and stages 3, 4 and 5 larvae was evaluated on *Spodoptera frugiperda* stage 2 caterpillars at the laboratory. On the other hand, only adults and stage 5 larvae of the predator were tested in a semi-controlled environment. The results of tests carried out at the laboratory show that the predatory capacity of *R. albopilosus* increases progressively with the evolution of the stages. On average, 7.37 ± 1.67; 8.92 ± 1.78; 12.25 ± 2.71 and 11.78 ± 2.00 prey were respectively consumed by stage 3, 4, 5 and adult of *R. albopilosus*. In addition, high prey consumption was observed among female predators at all stages of development, with an average of 7.31, 9.38, 14.30 and 13.36 prey items consumed by stage 3, 4, 5 and adult females respectively. In a semi-controlled environment, high prey consumption was observed in stage 5 predators over the ten days of observation. The average numbers of prey consumed by the predator of stage 5 every 48 h over ten days were 8.83, 7.75, 6.75, 7.08 and 6.92 respectively.

Keywords: Fall Armyworm, *Spodoptera fugiperda*, *Rhynocoris albopilosus*, Maize, Biological Control, Predation, Côte d'Ivoire

1. Introduction

Cereal crops have a major role in the agricultural sector. Among these crops, maize (*Zea mays*) occupies a prominent position, both nationally and internationally [1]. Maize is an annual tropical plant of the Poacae family, native to central America and Mexico [2]. It is the most energetic cereal due to its nutrients (rich in starch minerals) and the most economical from a production point of view: a simple crop to produce, harvest and store [3, 4].

Maize is the world's leading cereal crop, followed by rice and wheat. Global maize production for the 2019-2020 season was 1,091 million tonnes [5]. The role of maize as a basic food in Africa is comparable to that of rice or wheat in Asia. Some 208 million people in Africa depend on maize as a source of food security and economic well-being [6]. In Côte d'Ivoire, it is the second most important cereal crop after rice, with an average production of 1,175,715 tonnes in 2020 [7]. Maize is cultivated in almost all zones and is the most widely used raw material for animal feed (poultry, pigs, cattle), human foodstuffs and the brewing, soap and oil industries [8].

The potential of the maize sector is very encouraging. But, unfortunately, it faces a number of difficulties, notably soil impoverishment and the pressure of pests and diseases, which are compromising its progress. Indeed, in addition to the effects of granivorous birds and other crop pests, the country has experienced the invasion of a new pest, the fall armyworm (FAW) *Spodoptera frugiperda* J.E Smith. First observed in West and Central Africa in [9], its presence was first reported in Côte d'Ivoire in October 2016, by growers benefiting from Agricultural Production and Marketing Support Project (PROPACOM). Between July and October 2017, similar entomological problems were also reported by other maize producers, both in the central and northern regions of the country.

According to the FAO [10], this pest threatens the food security of over 300 million people in Africa and can cause significant economic losses, up to $4.8 billion for maize production alone. Fall armyworm can cause famine in sub-Saharan Africa, where cereals are considered staple crops for some peoples [11]. Because of its polyphagous behavior and reproductive capacity, the fall armyworm is considered a cosmopolitan pest [12, 13].

To date, the main control methods have been chemical. According Togola *et al.* [14], chemical control with the use of synthetic chemical insecticides has disadvantages for human health, the environment as well as non-target organisms (crop beneficials). Moreover, the armyworm has developed a resistance to chemical insecticides.

In view of all the above, other alternatives for controlling this devastating caterpillar could be considered. Biological control using natural enemy insects would be judicious and highly efficacious against *Spodoptera frugiperda*. The aim of this study was to assess the biological control potential of the predator *Rhynocoris albopilosus* against the fall armyworm.

2. Material and Methods

2.1. Study Areas

This study was conducted at two different sites within the University Nangui ABROGOUA (UNA), namely in a controlled environment, in the Agricultural Entomology Laboratory, and in a semi-controlled environment, on an experimental plot. According to figure 1, the UNA (with geographical coordinates 5°17' and 5°31' North latitude and between 3°45' and 4°31' West longitude) is located in the Abidjan district, in the south of Côte d'Ivoire [15]. The climate of the study area is humid tropical, subdivided in four seasons: a long rainy season and a short rainy season from March to July and October to November respectively, with more rainfall in June [16]; a long dry season and a short dry season from December to February and August to September respectively. Recorded rainfall varies between 1,500 and 2,500 mm in the rainy season and 1,200 to 1,500 mm in the dry season. Average annual temperatures hover around 25 and 29°C (Figure 2).

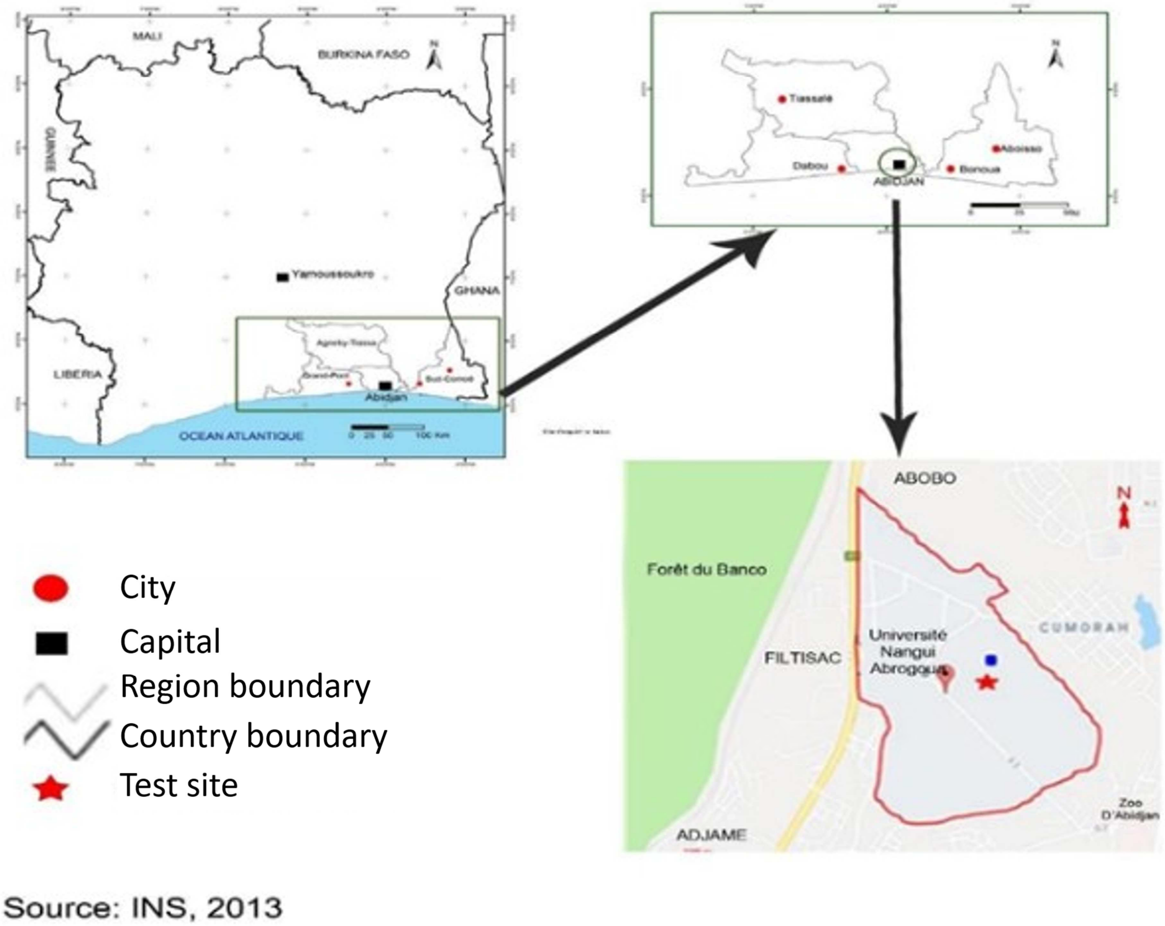

Figure 1. *Geographical location of University Nangui ABROGOUA [17].*

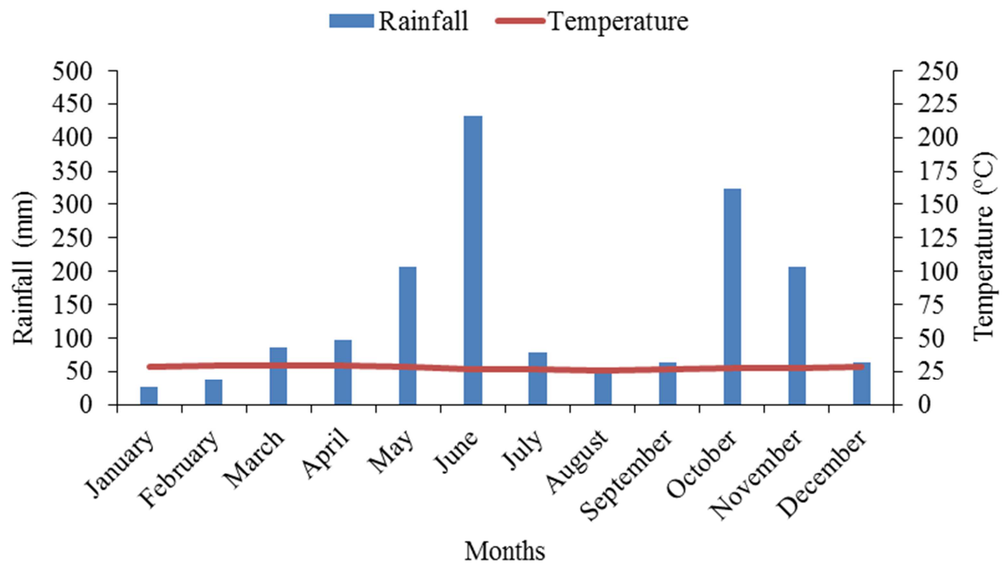

Figure 2. *Umbrothermal diagram for Abidjan district 2015-2021, SODEXAM.*

2.2. Choice of Insects Tested

The choice of *R. albopilosus* as predator is explained by the fact that this insect is naturally present in almost all maize ecosystems in Côte d'Ivoire and has been observed consuming FAW. Maize plantations where its presence has been strongly observed show low levels of infestation. In addition to this, its biology and ethology are well known, its rearing in the laboratory is easy [18]. *R. albopilosus* has been mentioned as a natural enemy of the fall armyworm by various authors [19, 20]. As for the prey, only stage 2 caterpillars of *Spodoptera frugiperda* were used. This choice was also justified by the fact that, at this stage, caterpillars are generally present on the aerial parts of plants, and therefore susceptible to capture by *R. albopilosus*.

2.3. Insect Rearing

2.3.1. The Prey Spodoptera frugiperda

Rearing of *S. frugiperda* was initiated using eggs and larvae collected in the Yamoussoukro department. The larvae were reared and fed on maize leaves and cobs. The number of caterpillars in the rearing bins was reduced each time the larvae progressed to a higher stage due to their cannibalistic behavior, resulting in 20 larvae during the second larval stage, ten larvae during larval stages 3 and 4, and five to three caterpillars during the last two developmental stages. As the larvae complete their development, they transform into chrysalises. These chrysalises were placed individually in pillboxes for pupation. Adult *S. frugiperda* that had emerged from the pupae were released into rearing cages for reproduction. Cotton soaked in honey water was placed in the cages as food for the butterflies. Organs of *Panicum maximum* were placed in the cage to support egg-laying.

2.3.2. The Predator Rhynocoris albopilosus

R. albopilosus rearing also began with eggs collected in the Yamoussoukro department. As soon as they had hatched, the larvae were reared individually in rearing tanks. *Tribolium castaneum* nymphs served as food for stage 1 *R. albopilosus* larvae. From the second stage to the adult, *R. albopilosus* individuals were nourished with stage 2 and 3 *S. frugiperda* caterpillars and *Tribolium castaneum* larvae, and on rare occasions with maggots and Orthoptera (crickets, locusts). In the adult stage, pairs were formed and the same process was repeated until a large population of the predator was obtained. Insects were reared under ambient conditions (temperature: 28.8 ± 2.82 °C; relative humidity: 74 ± 11.33 % and photoperiod: 12: 12).

2.4. Rhynocoris albopilosus Predation Tests on Spodoptera frugiperda

2.4.1. In Controlled Environment

Newly emerged *R. albopilosus* stage 3 larvae were placed individually in the rearing tanks (Figure 3). Twenty (20) stage 2 *S. frugiperda* larvae, renewed every 24 h, were given as prey to each predator. The number of prey items consumed by the predators was counted over 24 hours. In addition, uneaten prey was removed from the bins and new prey was supplied. The number of prey items consumed daily by each predator was recorded, as were molts and mortalities. Tanks containing CLA caterpillars only served as controls. A total, 30 predators were monitored from stage 3 to stages 4, 5 and adult. The parameters studied were total consumption (Tc), average daily consumption (Dc) and prey consumption index (Ci).

Total prey consumption (Tc) for a given predator stage is the average number of prey items consumed by the predator during that stage, according to the formula:

$$\mathrm{Tc} = \sum_{n}^{i=1} \mathrm{NPi}$$

with NPi number of preys consumed on day i of the cycle and n=last day of the cycle.

Daily consumption (Dc) is the ratio between total consumption and the duration of development of the given stage, according to the formula:

$$Dc = Tc / D$$

with Tc=total consumption and D=development time

The consumption index (Ci) is the ratio between total consumption and the number of effective days of prey consumption.

$$Ci = Tc / EDpc$$

where Tc=total consumption and EDpc=effective number of days of prey consumption.

2.4.2. In Semi-Controlled Environment

This test was carried out on a 38 m^2 experimental field at the University Nangui ABROGOUA (UNA). Sowing was carried out after weeding, using the CNRA [21] technique. Spacing was 75 cm between rows and 50 cm on rows. Two to four seedlings were planted per stake. One month (30 days) after sowing, four plants were framed by pipe bars (L: 1 m; W: 40 cm and H: 1 cm), forming lots of plants. These plants were infested with stage 2 caterpillars of previously reared in the laboratory, with a maximum of three caterpillars per plant. After infestation, the frame was covered with mosquito netting and one predator was introduced. Two days (48 h) after the infestations, the predator was removed from the environment; the plants were then carefully examined to count the exact number of preys consumed by the predator. Uneaten prey was removed and new prey was offered to the same predator. Plants infested only with caterpillars served as controls. A total, 30 predators comprising ten stage 5 individuals, ten males and ten females, were used to carry out the tests which lasted just 10 days (Figure 4).

Figure 3. *Consumption of S. frugiperda L2 caterpillars by R. albopilosus L3 larvae in the laboratory.*

Figure 4. *Infested plants covered with mosquito nets in a semi-controlled environment.*

2.5. Statistical Analysis

The data collected were recorded in Excel 2013. After transformation, they were subjected to various analyses using R Studio 4.2.2 software. Single-factor analysis of variance (ANOVA 1) or its not-parametric equivalent was used to compare average prey consumption by the various predators in controlled and semi-controlled environments. In the event of a significant difference, the Newman-Keuls test at the 5% threshold was used to classify the means. Two-way comparisons (by sex) of the number of prey items consumed between predator stages were made using Student's T-test.

3. Results and Discussion

3.1. Predation Capacity in a Controlled Environment

3.1.1. Total Prey Consumption by Different Predator Development Stages

The total number of prey items consumed by *R. albopilosus* larvae (Table 1) and adults (Table 2) increases in parallel with their lifespan and stage of development. The prey consumption indices for larvae correspond to the average total consumption, as they consumed prey every day at these different stages.

Table 1. *Average total and daily prey consumption by Rhynocoris albopilosus larvae.*

Stage of developpment	Total prey consumption			Daily prey consumption			Duration (Days)		
	Min	Avg	Max	Min	Avg	Max	Min	Avg	Max
Stage 3	28	44.33	78	7	9.93	18	5	6.03	7
Stage 4	50	70.50	117	6.25	9.12	14.60	7	7.73	9
Stage 5	74	120.33	166	8	12.35	18.22	9	9.73	11

Table 2. *Average total and daily prey consumption by Rhynocoris albopilosus adults and consumption indices.*

Total prey consumption			Daily prey consumption			Duration (Days)		
Min	Avg	Max	Min	Avg	Max	Min	Avg	Max
270	472.77	689	6.40	10	13.78	35	61.43	104

NDC			NDNC			Ci		
Min	Avg	Max	Min	Avg	Max	Min	Avg	Max
34	61	104	0	0.45	3	3	7.97	13

NDC: number of days of prey consumption by the predator during its development;
NDNC: number of days of not-consumption of prey; Ci: consumption index

Daily prey consumption by different predator development stages

Prey consumption according to R. albopilosus development stage

The study of daily consumption showed a highly significant difference in the number of prey items consumed by the different developmental stages of the predator, using the Newman Keuls test (p=0.000). These tests reveal that the number of prey items consumed by the different predators increases progressively with the passage from one stage to the next (Figure 5). Stage 3 and 4 predators consumed an average of 7.37 ± 1.67 and 9.17 ± 1.78 prey per day respectively. From the fifth larval instar onwards, a considerable increase in prey numbers was observed (12.25 ± 2.71 prey). This high prey consumption dropped slightly when the predators reached the adult stage (11.78 ± 2.00).

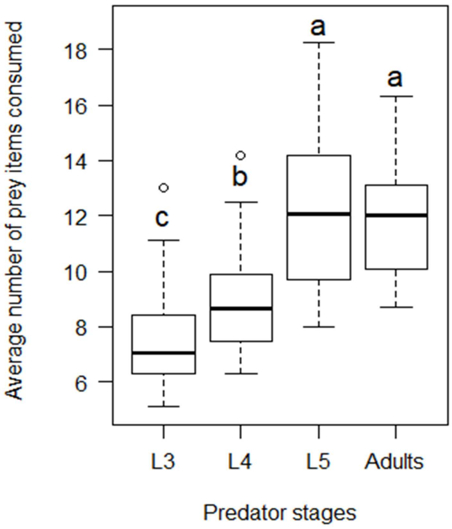

Figure 5. *Average consumption of Spodoptera frugiperda stage 2 larvae by the different development stages of Rhynocoris albopilosus.*

Histograms bearing the same letter are statistically identical according to ANOVA 1 and Newman-Keuls tests at the 5% threshold, p = 0.00.

3.1.2. Prey Consumption by Sex

Since the predators were monitored individually from stage 3 to the adult stage (sexual differentiation stage) and from there to their death, we were able to determine for each adult predator the number of prey items consumed at each developmental stage since its use. Figure 6 shows that regardless of the development stage of *R. albopilosus*, females consumed more prey (10.93 ± 2.88) than males (8.69 ± 2.30). The average numbers of prey consumed by females from stage 3 to the adult stage are respectively 7.89 ± 1.44; 9.70 ± 1.73; 14.21 ± 1.83; 11.90 ± 1.81 versus 6.91 ± 1.85; 8.71 ± 2.03; 10.75 ± 2.11 and 8.41 ± 1.46 in males during these stages with respective probabilities of p = 0.04412; p = 0.1605; p = 0.000; p = 0.000.

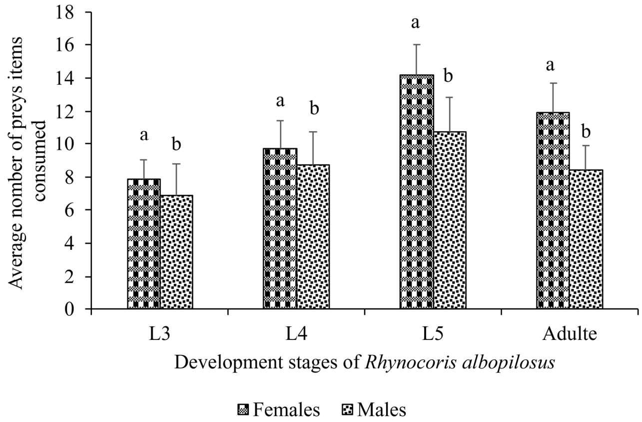

Figure 6. *Average consumption of stage 2 Spodoptera frugiperda larvae by males and females of the different development stages of Rhynocoris albopiosus*

For each stage, histograms bearing the same letter are statistically identical according to Student's t-test at the 5% threshold, p: probability associated with the tests.

3.2. Predation Capacity in a Semi-Controlled Environment

3.2.1. Overall Consumption of All Predator Stages in a Semi-Controlled Environment

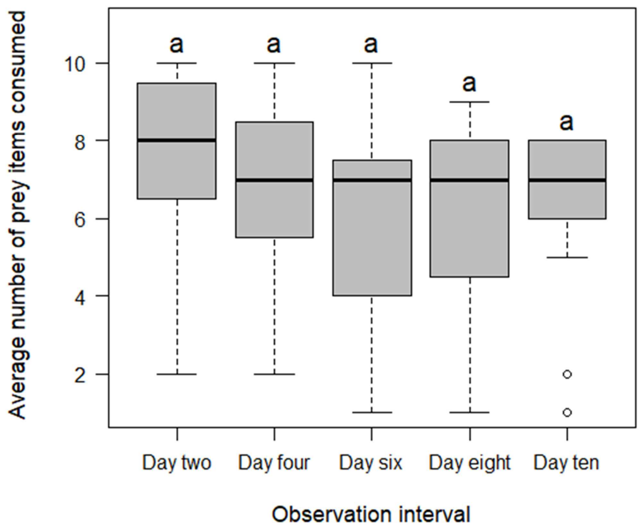

Figure 7. *Overall consumption of stage 2 Spodoptera frugiperda caterpillars by Rhynocoris albopilosus in a semi-controlled environment.*

Figure 7 shows that there was no significant difference in the average number of prey items consumed by all predators during the ten days of observation, which was confirmed by statistical analysis (p=0.398). Two days after the predators were provided with prey, the average number of preys consumed was 7.37 prey. From day four to day six, a slight drop in predation capacity was observed, with 6.53 and 5.95 prey consumed respectively. From the sixth day onwards, their predation capacity increased slightly to 6.32 prey and this quantity of prey consumed was kept constant until the tenth day.

Histograms bearing the same letter are statistically identical according to the multiple comparison of ranks associated with the Kruskal-Wallis ANOVA test at the 5% threshold, probability associated with the test (p = 0.398).

3.2.2. Average Daily Consumption of Prey by Stage 5 Larvae and Adult Males and Females of Rhynocoris albopilosus

The number of prey items Consumed evolved differently during the ten days of observation when the predators were considered individually. However, the predators of S5 were the most voracious, despite the trends observed, with a maximum of 8.3 prey items consumed on average on the first day, compared with 5 and 4.5 prey items respectively for females and males. From the sixth to the tenth day of observation, high prey consumption was observed in all predators, whatever their level of consumption (Figure 8).

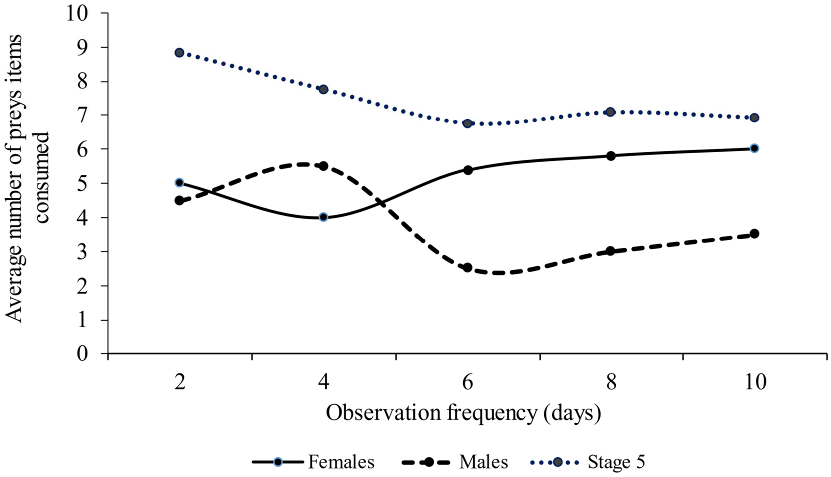

Figure 8. *Average consumption of stage 2 Spodoptera frugiperda caterpillars by stage 5 larvae and adult males and females of Rhynocoris albopilosus in a semi-controlled environment.*

The results concerning the predation capacity of *Rhynocoris albopilosus* on *Spodoptera frugiperda* larvae in a controlled environment revealed that the passage from one stage to another required a high level of prey consumption by *R. albopilosus* individuals. This high consumption of prey would be necessary for the predator's growth and development. About to the predator's growth, the size, shape and weight of the individual increased with each change of stage. These results corroborate those of Kwadjo *et al*. [18] who found on *R. albopilosus* and Hema [22] on *Phonoctonus lutescens* that the average number of prey consumed increased with developmental stage. Similar results were obtained by Biswas *et al*. [23] using the predator *Stethorus punctillum* Weise (Coleoptera: Coccinellidae) against the mite *Tetranychs urticae* Koch. In this study, the authors observed that the number of prey items consumed by *S. punctillum* increased in parallel with the different stages of development.

Since the development of individuals is qualitative, it requires the production of food energy in the organism to promote all these transformations. This accumulation of energy would require a high consumption of prey. The more prey an individual consumes, the more rapid is it development; the less it feeds, the less rapid is its development. Generally, in insects of the genus *Rhynocoris*, the number of prey consumed increases as the insect develops, as Moulet [24], Kwadjo *et al*. [18] and Soro *et al*. [25] have shown. The voracity of stage 5 predators may also be due to the fact that it is at is stage that the reproductive organs and developmental stages are completed. As a result, the larva needs a lot of energy to make the transition from this stage to the imago, which is why it consumes so much prey. The high prey consumption of stage 5 predators could also be explained by the fact that stage 5 larvae are naturally more voracious than other larval stages, as observed by Kwadjo *et al*. [18] in *R. albopilosus*. According to this author, in general, the daily and total consumption of prey by this insect depends on the developmental stages. Similar observations were reported by Namkosserena [26] in the species *Exochomus concavus* Fürsch (Coleoptera: Coccinellidae), this author affirmed that, whatever the developmental stage of the prey, the last larval stage of this predator is the most voracious. When gender is taken into account, the results show that females consumed more prey than males at all stages of development. These results can be explained by the fact that females are generally larger and fatter than males and grow more rapidly than males, so they consume more food to ensure this growth, as observed by Bouallam *et al.* [27] in a biological control context using larvivorous fish *Gambusia affinis* to control Anopheles larvae. On the other hand, females, being responsible for reproduction, would consume more food to produce energy for the development and growth of the eggs they carry. These results corroborate those of [28] in their work on turtle predation. The results of their research revealed that the number of prey items ingested was significantly higher in females than in males. Other similar studies investigating the predatory capacity of various insects, including *Ranatra parvipes* vicina against *Aedes aegypty* larvae [29], *Rhynocoris albopilosus* against *Podagrica decolorata* [30, 22] have shown that females consumed more prey than males. Arbogast [31] also made the same observations in a study on the control of stored food insects using an auxiliary, *Xylocoris flavipes* (Hymenoptera). The results of this study revealed that the females were more voracious than the males.

In a semi-controlled environment, the overall consumption of prey did not vary over the 10 days of observation for all the predators used. These results may be explained by the fact that, under these conditions, the prey does not constitute a danger for the predator, because as soon as the plants are infested by caterpillars, they migrate directly into them. This was not the case in a controlled environment, where the tests were carried out in the rearing tanks, where the predator was in direct contact with the prey. There was also the fact that the location of the prey in the plants or in the whorl made it difficult for the predators to spot them. As a result, the predator spent a lot of time looking for the prey over the 10 days of observation. As for the predator, under these conditions, it would hide and immobilized itself in the foliage to better apprehend the movements of the prey. Kwadjo *et al*. [18] mentioned that total immobility of *R. albopilosus* is a strategy adopted by this predator to detect and capture prey. These observations are confirmed by those of Villiers [32] in Esmesinae, which hide in grasses to capture their prey. The time involved in this whole process would justify these tendencies in a semi-controlled environment. In addition, this constant consumption of prey under these conditions could perhaps be the quantity of prey consumed by the latter per day under natural conditions.

When prey consumption was evaluated separately for the three types of predators (stage 5, males and females) in a semi-controlled environment, the number of preys consumed differed from one predator to another over the ten days of observation. The overall trends observed show that stage 5 predators consumed more prey than males and females. This high consumption of prey by stage 5 predators may be due to the fact that their voracious behaviour is genetic.

The trends observed in stage 5 predators can be explained by the fact that Reduviidae naturally consume less prey a few days after moulting, since the duration of this stage (stage 5) is 9.62 ± 0.27 [19]. However, in males and females, despite their low level of prey consumption, they had a progressive consumption of prey, this slight increase in the number of preys was observed from the third day of observation (day six) to the fifth day of observation (day ten). These results can be explained by the fact that at this period, after the imaginal moult and under the climatic conditions in which the tests were carried out (T = 28.8 ± 2.82 °C and H = 74 ± 11.33 %), *R. albopilosus* adults enter their pre-oviposition period, which would be the basis for this gradual increase in prey consumption. These results corroborate those of Kwadjo *et al.* [18] who observed in his work that the pre-oviposition period of *R. albopilosus* adults is between eight and ten days at 28°C.

4. Conclusion

This study consisted of identifying the natural insect enemies of *Spodoptera frugiperda* and assessing the predation capacity of *Rhynocoris albopilosus* on this insect, which is the main maize pest in Côte d'Ivoire. As a result of this work, the predator *R. albopilosus* was identified as a potential biological control agent against this pest. Predation tests carried out in a controlled environment showed that stage 5 and adult predators consumed more prey than stage 3 and 4 predators, but females were the most voracious, at all stages of the predator. On the other hand, in a semi-controlled environment, stage 5 individuals consumed more prey than males and females.

These data on the antagonistic actions of these two insects suggest that they could be used as biological control agents against *Spodoptera frugiperda* to offset the excessive use of chemical pesticides in the strategy for sustainable control of this pest.

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Conflicts of Interest

The authors declare no conflicts of interest below this section.

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