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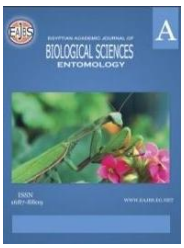

Morphology, Damage Symptoms and Biology of *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae) Strains in Egypt

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ABSTRACT

Fall armyworm (FAW) *Spodoptera frugiperda* (J.E. Smith) is a destructive pest of corn. FAW invaded Egypt in May 2019 and was recorded for the first time in corn plantations in Kom Ombo - Aswan Governorate. Recently *S. frugiperda* C-strain was Confirmed in Nile corn plantation in Meet AL - Deeba village, (Qillin-Kafr El-Sheikh); while R-strain was confirmed in summer corn plantations of Abo Al- Gadail, (Qillin-Kafr El-Sheikh) during season 2022. Moreover, The R strain of *S. frugiperda* infested corn plants beside uninfected neighbourhood rice fields. This indicated that, R- strain of *S.frugiperda* preferred summer corn plants, as a host, than rice.

Recent evidence of both strains, collected from corn fields of Kafer EL Sheikh, were identical morphologically and cause same damage symptoms. Biology of *S. frugiperda* C-strain and R-Strain was studied under natural conditions of field crop pests' laboratory at Plant Protection Research Institute Dokki, Giza, Egypt during Autumn and summer season (2022). C-strain larvae of *S. frugiperda* were fed on castor oil leaves. The average duration of life cycle and generation time at 23.0±2.0°c was 39.0±2.0 and 44.1±1.6 days, respectively. The corresponding averages for *S. frugiperda* R- strain corn - feed through, early 3 instars, at 28.0±2.0°c were 27.0±0. 5 and 29.0±0.6 days, respectively. The average of pre-oviposition periods {3.5±0.7 and 2.6±0.2 days} included the pre-mating period of 2.80 ±0.56 and 1±0.03 days for *S. frugiperda* C-strain and R-strain, respectively. The *S. frugiperda* C- strain virgin females deposited infertile eggs when the pre-mating period increased more than 4 days at 26.0±2.0°c and 75.0±5.0 RH. These results are important for planning an effective integrated control against fall armyworm to prevent it's rapid spread in Egypt.

INTRODUCTION

Fall armyworm (FAW), *Spodoptera Frugiperda* (J-E. Smith) is a lepidopteran polyphagous, migratory and invasive pest feed on leaves, stems and reproductive parts of economically important crops causing severe damage. This pest has two strains These strains are morphologically identical but differ genetically (Lu and Adang, 1996; Lewter *et al*., 2006 and Nagoshi*et al*., 2007). In Indonesia, Herlinda *et. al*. (2022) classified *S. frugiperda* genetically to *S. frugiperda* corn strain (C-strain) and rice strain (R- strain).

It invaded Egypt in May 2019 and was recorded for the first time in corn plantations in KomOmbo, Aswan Governorate according to the Agricultural pesticide committee, Ministry of Agriculture. After that, it spread in corn fields through Egypt. Also, Mohamed *et.al*. (2022) recorded *S. frugiperda* in Sorghum fields in Assuit Governorate, 2021 season. El-Lebody *et al*. (2024), revealed the presence of *S. frugiperda* C-strain in corn plantations, but they recorded *S.frugiperda*R-strain only in summer corn plantations of Meet AL-Deeba, Qillin, Kafer El-Sheikh, Egypt, in rice area during season 2022. Kaute *et. al* (2019) indicated that, both of the two strains may share the same host. Moreover, Herlinda *et al.* (2022) reported that, both strains prefer the corn plants as a host. Maharani *et al*. (2021) reared *S. frugiperda* on corn to observe the life cycle and life table parameters in comparison with those of rice-fed *S. frugiperda*. They found that, the corn was more suitable host for it's growth, development and reproductive capacity. They added, the corn-fed *S. frugiperda* with supportive environment reproduce and multiply faster than *the* rice-fed. Rajisha *et al*. (2022) reported that, the corn plants aged 2weeks were more suitable for growth and development of the first three instars compared to the plants aged 35- 60 days,while the duration of the late instars was not affected. Fatoretto *et al*. (2017), Herlinda*et al*.(2022) and Maharani *et al*. (2022) indicated that, the rapid resistance evolution of *S. frugiperda* was affected by it' s biology, ecology and genetic compositions. Also, the morphology and damage symptoms as well as life cycle and life table parameters of *S. frugiperda* are important for both farmers and specialists to discover the pest and increase of it' s population. The former items are essential for planning appropriate control strategies against *S. frugiperda.*

The present study recoded all life stages of the *S. frugiperda* in corn fields along with its attack appearance. Moreover, life cycle, some reproductive features and generation time (T) of both *S. frugiperda* C- strain and R –strain are recorded under the conditions of summer and autumn 2022, in Egypt.

MATERIALS AND METHODS

S. frugiperda Occurrence and Damage Symptoms:

Surveys were carried out sequentially in both summer and Nile corn plantations in 2 villages of Qillin city, Kafer EL Sheikh Governorate, Egypt, during season 2022. The collected samples of infested summer plants aged 33 days were from Abo Al –Gadial fields on 3rd August,while, infested Nile plants aged 40 days, were from Meet AL-Deeba on 17th October. The plants were dissected to exclude larvae to identify morphologically and record its damage symptoms according to Mohamed *et al.* (2022} and Herlinda *et al.* (2022). Excluded larvae of Abo AL- Gadail were subjected to genetic identification by El-Lebody *et al.* (2024) and confirmed twice as *S. frugiperda* R-strain (Accession Number, OP647509 & OP649580), while Meet AL-Deeba larvae were confirmed as *S. frugiperda* C-strain (Accession Number, OQ920981).

Separately, larvae of the strains (about 50 / each) were immediately kept in plastic containers (15x8x5cm) with punctured cover cemented with toilet paper and provided daily with youthful corn foliage to feed on until pre-pupation under natural conditions of the laboratory.The newly emerged moths were paired and kept in glass jars (1/2 kg) covered with muslin cloth with rubber bands. The pairs were replicated 5 times as one moth of each sex in each rep. The moths were fed on a 10% sucrose solution.Biological observations of the first generation for both *S. frugiperda* strains under 23±2°c and 28±2°c during Autumn and summer, respectively were recorded. All durations of the different stages in addition to life cycle and generation time (T) were registered. Moreover, the mortality percent of immature stages, adult malformation and sex ratio were recorded.

RESULTS AND DISCUSSION

Spodoptera frugiperda Occurrence, and Damage Symptoms:

The present results *confirmed* that*, S. frugiperda* infests corn fields of Abo-Al-Gadail and Meet AL-Deeba, Qellin, Kafer El Shiekh, Egypt, with incidence of 100 % in both summer corn aged 33 days on 3rd August and Nile corn aged 40 days on 17th October. In this respect, Herlinda *et. al*. (2022) found that, maximum incidence of *S. frugiperda* was after 21 to 43 days of sowing date. However, the first occurrence of this pest in Egypt was recorded in a village of Kom Ombo city in May 2019, then it was recorded in all corn and sorghum fields through Upper Egypt (Mohamed *et al*., 2022).

The results showed presence of creamiest or pale green (rarely, coppery) egg masses in one or more layer(s) covered with scales on both apixal and adixal of leaves as well as on the whorl of plants; Figs.1 & 2 (A, B & C). This result agrees with that recorded by Russianzi *et al*. (2021), who reported that, the scales were to protect its temperature. Also, Bhusal and Kamana (2019) recorded egg masses on both leaf surfaces, while, Julio *et al* (2017) found that, females laid their egg masses on the apixal of the newly leaves. On contrary, Rajisha *et al*. (2022) reported that, females prefer to lay their egg masses in 2 to more layers on the adixal of corn leaves.

In addition, the egg mass color turned to black just before hatching due to forming of the neonate head capsule; Fig.1 (D & E) The same result was illustrated by Poul and Sonali (2020) and Russianzi *et al*. (2021).

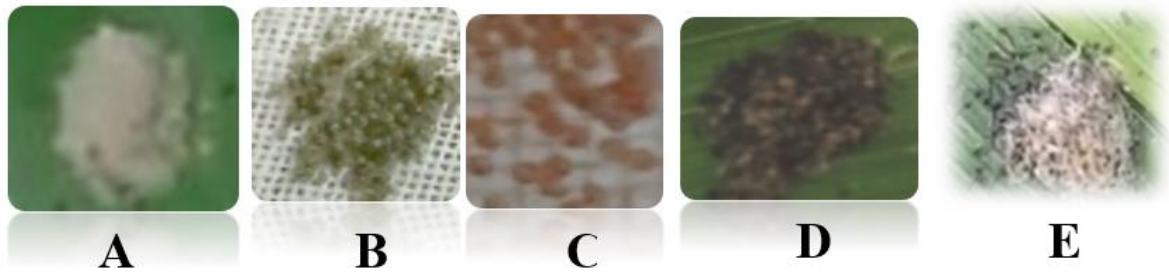

Fig. 1: *S. frugiperda* egg masses, different ages with their colors**.**

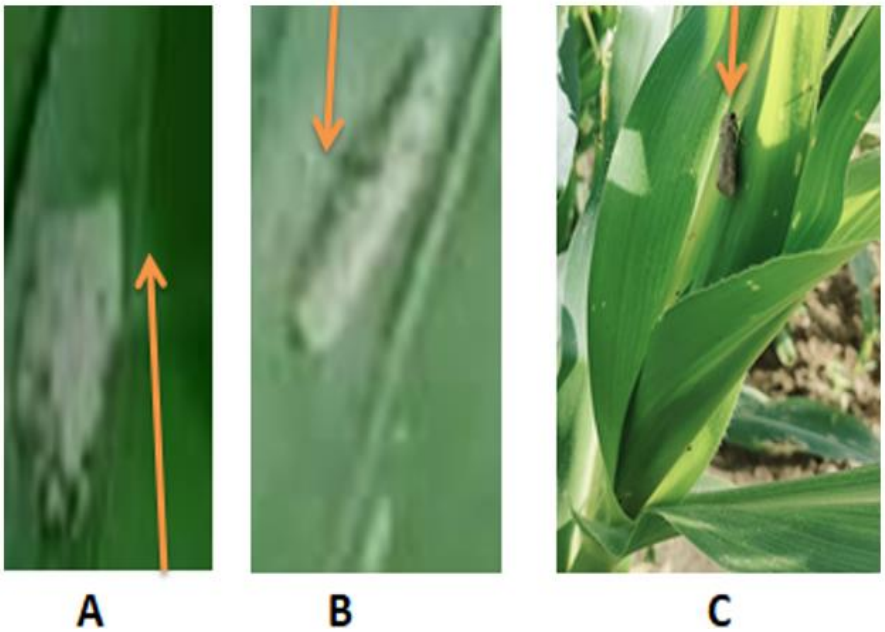

Fig. 2: *S. frugiperda* egg -position sites on corn leaf.

However, young larvae (1-3 instars) eating caused transparent patches on the adixalof corn leaves; (Fig. 3 A & B). The 4th instar larvae causes more rigorous scratches with small irregular holes on the leaves; Fig.3 (C). The later instars (5th& 6th) cause big holes and biting of leaf edge; Fig.4 (A) and may eat the whorl partially or totally; Fig.4 (B) causing death of plants and consequently replanting corn fields. Also, presence of larval faeces like sawdust on leaves is a good symptome of this pest; (Fig. 4 C). The body of the later instars have many colors; (Fig.5 A). The larvae from 3rd to 6th instars can be identified morphologically by presence of 4 black pincula forming a square shape on the 8th abdomen segment; (Fig.5 B). The full-grownlarvae shrunk its body to form the pre- pupae; Fig. 6 (A). The newly pupae are green in color; Fig. 6 (B), then turned to dark reddish brown color. The same characteristically features of *S. frugiperada* were recorded by Poul & Sonali (2020), Russianzi *et al*. (2021), Rajisha *et al*. (2022) and Sharma *et al*. (2022).

Fig. 3: Leaf – feeding of young larvae (1st- 3rdd instars) and more vigorous patches by 4thlarval instar.

Fig. 4: Leaf feeding of older larvae.

Fig. 5: A. *S.frugiperda*colors**, B.** black pincula

Fig. 6: Pre and new pupae

Host Preference:

Recently it was observed that the R-strain of *S. frugiperda* infested summer corn plantations neighbouring uninfected rice fields in Abo AL-Gadail village. Kuate *et al*. (2019) recorded that the same host may be share by *S frugiperda* R-strain and C- strain. Also, Maharani *et al*. (2021) proved that, *S. frugiperda* prefered the corn than rice plants

Biological results of *S. frugiperda* C- strain:

Data in Table (1) clarify that, the egg incubation period of *S. frugiperda* C-strain ranged 5- 6 days with an average of 5.5±0.2 days with hatching 100%. Also, the larval stage lasted 14-18 days with an average of 16 ±1.4 days, including 1 to 2 days for the pre-pupal stage. In this regard, Gamil (2020) studied the biological aspects of the 4th generation of *S. frugiperda* at 26±2°c and RH % 65±5, and found that, the egg incubation period was 2.9 days (hatchability 97.33%) and the larval stage lasted 21.4±0.59 days. Also, Sharma *et al*. (2022) recorded 3 days for incubation period and 2– 4 weeks for maize–fed larvae at 26°c and 75% RH. Dahi *et al*. (2020) reported that, the duration of *S. frugiperda* larval stage is affected by host type and rearing conditions.

Table 1: Durations (day) and mortality percent in C – Strain Immature Stages at 23 ± 2°C.

Immature stage	Egg	Larvae	♀ pupae	♂ pupae
Duration (days)	4 - 6 5.5 ± 0.2	14 - 18 16 ± 1.4	16 - 19 17.8 ± 1.2	16 - 21 19 ± 1.9
Mortality %	0.0	36.5	37.5	

Fig. 7: *S. frugiperda* cannibalism appearance

However, the mortality percentage of castor fed -*S. frugiperda* C-strain larvae was 36.5 Table (1). The most mortality occurred in 5th & 6th instars. Mortality by pathogens and cannibalism was few; (Fig. 7). Similarly, Poul and Sonali (2020) reported that, Cannibalism was significantly higher in late larval instars, 5th and 6th, than in early ones.

Also, Russianzi*et al*. (2021) Indicated that, there was a positive relation between

the mortality and the age of *S. frugiperda* population. In contrary, Sharma *et al*. (2022) recorded that, the larval mortality were 70 & 12 %, for the 1st and 2nd instars, respectively but no mortality was found in the later ones. However, Gamil (2020) cleared that, the larval mortality was 8.8% of castor-fed *S. frugiperda* in the fourth generation. The present study cleared that, the duration of female pupae of Castor-fed *S. frugiperda* C- strain was (17.87 ± 1.2 days) shorter than that of males (19.2±1.9) under 23 ±2°C. The pupal mortality recorded 37.5% (Table 1). Similarly, Poul and Sonali (2020) reported that, the duration of female pupae was slightly shorter than that of males. Moreover, these findings are almost in agreement with those recorded by Russianzi*et al*. (2021) who reported that, the *S.frugiperda* females emerged before males to ripen their eggs before mating. As for moth emergence and malformation, Table (2) show that, the adult emergence of *S. frugiperda* C- strain reached 100 % with 15.8 and 25% malformation for females and males, respectively. In this respect, Gamil (2020) reported that, the *S. frugiperda* adult emergence of the fourth laboratory generation was 96% with 1.08% malformation. However, in the present study adult emergence of *S. frugiperda* C- strain continued for 6 days and the females emerged before males by 1 to 2 days. This result is supported by that of Russianzi *et al*. (2021) who reported that the insect females have a pre-mating period to ripen their eggs.The present results confirmed that the life cycle ranged 33-35 and 34-38 days with an average of 34.2 ± 0.78 and 36.3 ± 1.3 days for females and males, respectively, Table (2). These results may agree with those of Rajisha *et al*. (2022), there was no significant difference between the total life cycle of *S. frugiperda* females and males. Also, Maharani *et al*. (2021) found that, the life cycle of corn fed-*S. frugiperda* was shorter than that fed on rice.

Sex ratio of *S. frugiperda* C-strain moths at 23 ±2°C was 1:1; Fig (8) & Table (2). (Gamil 2020) reported that there was balance in sex ratio (1.2♀: 1♂) of castor leaves- fed *S. frugiperda* at 26±2°c and 65±5 RH% . Also, Maharani e*t al*. (2021) found that, the sex ratio of both corn and rice-fed *S. frugiperda* was not affected by the host type. On other hand, they found that, mating process and fecundity of *S. frugiperda* were not affected by the balance of its sex ratio.

Table 2: Biological aspects of C – strain moths at 23 ±2°C.

Sex	%			Days		
	Emergence	Malformation	Sex ratio	Life cycle	* Pre-oviposition	T
♀	100	15.8	50	33-35 34.2 ± 0.78	3-5 3.5 ± 0.7	42.3 -47.3 44.0 ± 1.6
♂	100	25.0	50	34-38 36.3± 1.3	-	

It included pre - mating period for females ranged 2 – 4 days and averaged 2.8 ± 0.56 days, and 0.0 for males. T means generation time.

Fig.8: *S. frugiperda* imagos.

S. frugiperda C-Strain females have a pre-mating period of 3 - 4 days with an average of 2.8±0.56 days for ripen their eggs before mating (Table2). This result was supported by Russianzi *et al*. (2021). In contrast, the males of *S.frugiperda* C-strain can copulate on the same day of emergence. Observations (unpublished) referred that, the pre-mating period lasted more than 4 days at 26±2°c and 75±5 RH% of *S. frugiperda* C-strain caused increasing the pre- oviposition period and dramatically decreased eggs production (only 7: 10 eggs /3♀) with female sterility (zero hatchability). On the other hand, pairing of virgin females (5 days old) with virgin male (one day old) causes elongation in female life span (28 days) while the male lived only for 3:7 days. However, Rogers and Marti (1994) reported that, pairing of *S. frugiperda* females and males aged 2 days resulted in the highest reproduction capacity and fertility %, while delaying mating increased female life span. Also, our observations confirmed by Rupali *et al*. (2023) who concluded that, any technique delays mating such as disruption pheromones may successfully control this pest.

However, *S. frugiperda* C- strain started laying eggs immediately the next day of mating with pre - oviposition period of 3.5 ± 0.5 days (Table 2). The mated female laid several egg masses with grey scales for many times through the ovi-position period (9.0±0.9 days). The maximum daily deposited eggs was on the 2nd day of ovi- position period with a capacity of 187.5 egg / female with a total of 500 fertile eggs / female of the 1st generation. Gamil (2020) recorded 3.5 and 5.1 days for pre and oviposition period, respectively in the 4th generation at 26±2°c and 65±5 RH%. The reproductive capacity was 1787.5 ± 91 egg / female. On the other hand, Russianzi *et al*. (2021) found that, the corn –fed *S. frugiperda* produced females with a pre ovi- position period as 2-6 days and 10 days as the ovi- position period and fecundity 1012 eggs, the highest number was on the 2nd day of ovi- position. Maharani *et al*. (2021) studied the reproductive parameters of both corn and rice fed-*S. frugiperda* and found that, pre ovi- position periods lasted 2.3 and 2.7 days and ovi- position periods of 13 and 11 days with about1599 and 398.87 eggs / female, respectively.

However, in the present study, mean generation time (T) for castor-Fed *S. frugiperda* C strain at 23 +2°C averaged 44 ±1.6 days, (Table 2). Maharani *et al*. (2021)found that, the corresponding T for Corn-fed and rice-fed *S. frugiperda* reared at 25.7°c and 78 RH% was 35.27±0.6 and 39.36±0.41 days, respectively. They concluded that, this variation is due to the host type and other environment -tal factors, the population of *S. frugiperda* increased faster with the supportive factors and suitable host. In this context, using of *S. frugiperda* natural enemies will be ineffective against this pest.

Biological results of *S. frugiperda* R-strain:

As for R strain data, Table (3) shows some biological parameters of the first generation of *S. frugiperda* R-Strain under 28±2°c. The incubation period was 2±0.4 days. Early larval instars (1st, 2nd& 3rd) were fed on corn seedling while the older ones (4th, 5th& 6th) were fed on castor oil leaves. The duration of the larval stage was 18± 0.6 days (included 1-2 days for pre-pupae). The pupal stage lasted 7± 0.7 days. It is clear that *S. frugiperda* R-strain completed their life cycle in 27±0.4 days (Table 3). Similarly, Rajisha *et al*. (2021) found that the *S. frugiperda* egg incubation period, larval and pupal stages ranged 2-3, 13-20 and 7-11 days, respectively. and added, corn plants aged 15 days was more suitable for growth and development of early instars than those aged (35-60) days, Durations of late instars were not affected and the life cycle was 29 days. Russianzi *et al*. (2021) reported that, the life cycle of *S. frugiperda* lasted 32.26 days with no significant differences between females and males.

Table 3: Durations of R– Strain different Stages at 23 ± 2°C**.**

Stage	Egg	Larvae	Pupae	Life cycle	Pre-ovipostion	T
Days	2 – 3 2±0.48	18 –20 18±0.6	6 – 8 7±0.49	26 –29 27±0.4	2 – 3 2±0.3	28 –31 29±1.5

Egg hatching and adult emergence were 100%, sex ratio 1:1 and pre mating for females was a day.

The present results revealed that, the *S. frugiperda* R-strain females have only one day for pre-mating while the males can copulate on the same day of emergence. The pre - oviposition period was 2±0.2 days. So, the generation time (T) of corn- fed *S. frugiperda* R-strain lasted 29.0±0.6 days under 28±2°C.

However, the genetic composition, morphology, damage symptoms, life cycle, life table, ecology are important for identifying life stage, understanding the behaviour of the pest, determining the best time and technique for planning IPM strategies against *S. frugiperda* (Maharani *et al*. (2021), Rajisha *et al*. (2022) and Sharma *et al*. (2022).

Conclusions

The present study revealed that, *S. frugiperda* C- strain and R-strain injured both summer and Nile corn Plantations at Qillin Kafr – EL Sheikh. Both strains are typical morphologically and cause the same damage symptoms. Moreover, in the rice area; it was observed that *S.frugiperda* R-straine prefers corn plant as a host than rice during summer season where there was no infestation in neighbourhood rice fields in Qillin -Kafr – EL Sheikh, Egypt. Therefore, more effort should be directed to survey the host range, host preference and to specify the presence of both strains of *S. frugiperda* in Egypt. Additionally, the Egyptian conditions in summer are more suitable and supportive for *S. frugiperda* growth and development than that in autumn (T= 29±0.6 and 44±1.6 days, respectively). On the other hand, the host and /or environmental temperature didn't influence *S. frugiperda* sex ratio. Biologically, adult females didn't lay eggs without mating behaviour occurrence. Under optimum conditions, elongation of pre - mating period more than 4 days after females' emergence of C-strain decreased the reproductive capacity with laying unfertilized eggs. Recent results support the idea of recommending use of mating disruption pheromone in IPM strategies to prevent laying fertilized eggs and hinder the rapid spread of this dangerous pest.

Declarations:

Ethical Approval: Not applicable.

Authors Contributions: All authors contributed equally, and have read and agreed to the published version of the manuscript.

Conflicts Interests: No conflict of interest was reported by the authors.

Availability of Data and Materials: All datasets analysed and described during the present study are available from the corresponding author upon reasonable request.

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ARABIC SUMMARY

دراسة مورفولوجية وبيولوجية ومظاهر الضرر لسلالتى دودة الحشد الخريفية (*Spodopterafrugiperda* (J.E. Smith) (Lepidoptera: Noctuidae**) تحت الظروف الطبيعية في مصر**

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تعتبر دودة الحشد الخريفية (FAW*) Spodoptera frugiperda* (J.E. Smith) التابعة لرتبة حرشفية الاجنحة آفة مدمرة لمحصول الذرة. وقد غزت هذه الآفة مصر في مايو 2019 وتم تسجيلها لأول مرة في زراعات الذرة في إحدى قرى مدينة كوم امبو- محافظة أسوان. ومؤخرًا، تم تأكيد وجود سلالة الذرة *S.frugiperda* corn strain (C-strain)) في زراعات الذرة النيلية في قرية ميت الديبة (قلين-كفر الشيخ)، بينما تم تأكيد وجود سلالة ارز *S. frugiperda* rice strain (R-strain) في زراعات الذرة الصيفية في أبو الجدايل (قلين-كفر الشيخ) خلال موسم الذرة 2022. علاوة على ذلك فإن سلالة الأرز قد أصابت نباتات الذرة الموجودة بجوار حقول الأرز الغير المصابة بها. وأشارت هذه الدراسة إلى أن سلالة الأرز فضلت نباتات الذرة الصيفية كعائل في منطقة إنتاج الأرز وأظهرت النتائج أن أفراد السلالتين التي تم جمعها من حقول الذرة في محافظة كفر الشيخ، مصر متطابقتين على ما يبدو في الشكل الظاهرى وأعراض الضرر.

تمت دراسة بيولوجيا أول جيل معملي من سلالة الذرة C-strain وسلالة الأرز R-strain في ظل الظروف الطبيعية لمختبر آفات المحاصيل الحقلية بمعهد وقاية النبات بالدقي الجيزة، مصر أثناء الخريف (متوسط ±23 °2م) والصيف (متوسط 2 ± 28.0 °م) على التوالى. وكانت مدة دورة الحياة (من طور البيض حتى خروج الفراشات 39.0 (0.6± يوما و(0.5 ± 27) يوما على التوالى. وبلغ متوسط فترة ما قبل وضع البيض (0.7 ± 3.5 و 0.6.± 0.2 يومًا) شمل فترة ما قبل التزاوج (0.56± 2.8 يوما) و (0.3 ± 1.0 يومًا) و لسلالة الذرة والأرز على التوالي. وبذلك تكون مدة الجيل (441.6± يومًا) خريفا و (0.6 0±. 29 يوما) صيفا.

تبين أن زيادة فترة ما قبل وضع البيض لإناث سلالة الذرة إلى 5 أيام عند قد ينتج عنه وضع كميات قليلة جدا من البيض و كان غير مخصب عند 2.0 ± 26.0 درجة مئوية و 75.0 ± 5.0٪ رطوبة نسبية.

قد تسهم نتائج هذه الدراسة الحالية فى تعريف المزارعين بأطوار هذه الآفة وأعراض الضرر المصاحبة لها بشكل دقيق وكذلك أهميتها للتطبيقين والباحثين في التخطيط لبرامج مكافحة متكاملة فعالة ضد دودة الحشد الخريفية في مصر مع ضرورة استخدام فيرمون تشتيت التزاوج كأحد العوامل الصديقة للبيئة والمبتكرة لمكافحة الآفات لمنع الانتشار السريع لهذه الآفة الشرسة.