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Biology of *Spodoptera frugiperda* (Lepidoptera: Noctuidae) fed on different food sources

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Abstract
In order to measure the impact of a number of plants on the development of *Spodoptera frugiperda*, we carried out a comparative study on the biology of the fall armyworm using various food sources (millet, maize, sorghum, peanut, cowpea and artificial diet). When fed with cowpea and peanuts leaves, larval development was longer (21.46 ± 0.42 and 18.93 ± 0.18 days respectively). In contrast, adult stage duration became shorter when the larvae were fed with maize leaves and peanut (2.86 ± 0.21 and 3.33 ± 0.21, respectively). Similarly, the pupal development was longer when the larvae fed on peanut leaves and artificial diet (9.26 ± 0.11 and 9.40 ± 0.16 days, respectively), compared to the pupae whose larvae fed on maize, millet, sorghum and cowpea (8.46 ± 0.13; 8.40 ± 0.13; 8.13 ± 0.9; 8.93 ± 0.20 days, respectively). As for the adult stage, the duration is shorter when the larvae are fed with cowpea leaves (2.86 ± 0.21 days) and Peanut (3.33 ± 0.21days).

Keywords: *Spodoptera frugiperda*, maize, millet, sorghum, peanut, cowpea

Introduction
The fall armyworm (FAW) is an insect native to the Americas, which first appeared in Africa in January 2016 in Nigeria [1]. It then spread rapidly throughout sub-Saharan Africa, where it causes extensive damage [2-4]. In Senegal, *Spodoptera frugiperda* was reported for the first time in 2017 by [5]. This pest threatens the food security of more than 300 million people in Africa and can cause significant economic losses, up to $ 4.8 billion in maize production alone [6]. *Spodoptera frugiperda* is a highly polyphagous species, which can attack more than 80 plant species [7, 8]. *Spodoptera frugiperda* consists of two strains (maize strain and rice strain) that are morphologically indistinguishable. The identification of these two strains requires the use of genetic markers [1, 9]. The rice strain mainly attacks rice and certain forage grasses, while the maize strain predominates in agricultural settings where it mainly attacks crops such as maize, sorghum, and cotton [10]. In Senegal, there is a great diversity of crops which coexist in the various agricultural ecosystems of the country. However, FAW is a very polyphagous species and therefore well equipped to adapt to new plant species. This facilitates movement or migration of the pest through these different crops, which increases the risk of selecting new populations (*Spodoptera frugiperda* strains) of insects with new nutritional preferences.
This is the raison it seemed to be interesting to us to study the biology of *Spodoptera frugiperda* using as a food source the main plant species cultivated in Senegal as food sources. We evaluated the parameters such as the larval, pre-pupal, pupal and adult stages duration, the average weight of a pupae, the mortality rate of larvae and the sex ratio, depending on the food source.
The aim of the present study is therefore to make a comparative study of the biology of *Spodoptera frugiperda* on five plant species cultivated in Senegal.

Materials and Methods
Material
The animal material consisted of the eggs, larvae, pupae and adults of *Spodoptera frugiperda*.
The plant material consisted of all the plant species used to feed the larvae and which served as a substrate for laying eggs for adults of *Spodoptera frugiperda*. These are the leaves and seedlings of: maize, millet, sorghum, peanut and cowpea.

Methods

To study the biology of *Spodoptera frugiperda*, we collected larval samples at our experimental site located at the Direction de la Protection des Végétaux (DPV) in Dakar. These samples were brought back to the laboratory, or they were farmed to get imaginal stages and then eggs that we had incubated to get new generation larvae. And from the incubated eggs obtained from the generation of collected larvae, we had studied a number of biological parameters of *Spodoptera frugiperda* on various food sources (millet, corn, sorghum, peanut, and cowpea).

The experimental device consisted of 15 larvae with 6 repetitions, making a total of 90 larvae used from the third larval stage

Breeding of *Spodoptera frugiperda*

Egg incubation and larval stage rearing

The part of the leaf on which the eggs were laid was cut and placed at the bottom of a Petri dish. Leaves of the diffèrent plant species that were used in our study were cut into small pieces and previously placed in petri dishes so that at hatching, future larvae could feed. To get moisture, a few drops of water were poured on the leaves and then the Petri dishes were closed. The leaves were renewed daily to avoid yellowing or putrefaction until the eggs hatched.

Larvae nutrition

The larvae of *Spodoptera frugiperda* can be fed by an artificial nutrient medium specially made for them, or by a natural diet based on leaves (millet or maize for example).

Fig 1: Incubation of *Spodoptera frugiperda* eggs

Synthetic/artificial food support

As part of our study we had made an artificial diet inspired by the artificial food supports used by the International Center of Insect Physiology and Ecology (ICIPE), the Agricultural Research Council - Republic of South Africa (ARC-RSAE) and the International Maize and Wheat Improvement Center (CIMMYT). The ingredients and amounts of this regimen used as a control in our study are listed in the table below:

Table 1: The ingredients used for the preparation of the artificial diet

Fractions	Ingredients	Quantity (g or ml) for 3L of food support
A	Corn leaft powder	75,6g
	Cowpea powder	250g
	Milk powder	60g
	Ascorbicque acid	9g
	Sucrose	100g
	Distilled water	1,500ml
B	Agar-Agar	37,8g
	Ditilled water	1200ml
C	Formaldehyde 40%	6ml

Breeding management

The rearing of larvae was carried out as follows:

From the first to the third larval instar

After the eggs hatched, the contents of the Petri dishes (stage 1 larvae and feed) were transferred to the type A rearing dishes. The bottom of the type A boxes was covered with leaves used as a food source (e.g. maize) cut into small pieces. The leaves were renewed daily to avoid excessive water loss. Each box could accommodate a hundred newborn larvae up to the third larval stage. For the artificial diet, after hatching, the neonate larvae were transferred to the individual breeding boxes (type B boxes) containing feed beforehand (artificial diet). For the artificial diet, after hatching, the neonate larvae are transferred to individual breeding boxes (type B boxes) previously containing feed (artificial diet). All larval stages take place in these individual breeding boxes.

Fig 2: Box for breeding *Spodoptera frugiperda* type a larvae

From the third to the last larval stage

The cannibalism being more marked from the third to the last larval stage, the caterpillars are transferred individually using a flexible clamp from the third stage, into type B breeding boxes previously covered with leaves serving as a food source, cut into small pieces for individual rearing of these larvae. For each of the 6 food sources (millet, maize, sorghum, cowpea, peanuts and artificial diet), 15 larvae were transferred to individual breeding boxes. Since cannibalism was more marked from the third to the last larval stage, the caterpillars were transferred individually using a flexible clamp from the third stage, in type B breeding boxes previously covered with leaves serving as a food source, cut into small pieces for individual rearing of these larvae. For each of the 6 food sources (millet, maize, sorghum, cowpea, peanut and artificial diet), 15 larvae were transferred to the individual breeding boxes.

Fig 3: Individual breeding boxes (Type B boxes)

From the last larval stage to pupation

The droppings of the last stage larvae were only removed so that they could be used with the rest of the uneaten food as a substrate for pupation.

Adult rearing and egg laying

After pupation, the pupae were transferred to breeding cages. After the emergence of adults, a young seedling was placed in the breeding cage to serve as an egg-laying substrate. Males and females of *Spodoptera frugiperda* mated and females laid eggs on the young plant that served as a laying substrate. The part of the leaf on which the eggs were laid was then cut and placed in a type A rearing box after having previously counted the number of eggs using a stereo microscope.

Biology of *S frugiperda* on different host plants

The study of the biology of *Spodoptera frugiperda* based on the use of different food sources was carried out in the laboratory under controlled conditions with a temperature of 25°C ± 2°C, and a relative humidity of 70 ± 10%.

Colony of *Spodoptera frugiperda*

The colony of larvae used for our study came from eggs from larvae collected and identified on corn plants, on two of our study sites that are the DPV (Direction de la Protection des Végétaux) of Dakar and a field located in Bambilor.

Parameters evaluated

The parameters evaluated were:

- The duration of each stage (larval, pre-pupal, pupal and adult) depending on the food source
- The weight (in g) of 10 pupae, depending on the host
- The sex ratio: it is obtained by dividing the total number of females obtained by the number of males

Results

Prior to ANOVA analysis of variance, the results were subjected to exploratory analysis to test the normality and independence of the data (Shapiro and Wilk, 1965). To check the equality or homogeneity of the variances, we performed a Bartlett test. The mean treatment values obtained by ANOVA were compared using Tukey's test ($p \leq 0.05$).

Evaluation of the duration of the life stages of *Spodoptera frugiperda* on different host plants

The mean incubation time of *Spodoptera frugiperda* eggs is 2.45 ± 0.03 days. That of the pre-pupal stage is 1.07 ± 0.03 days. It does not vary significantly between different food sources (P-value = 0.393). In contrast, the duration of the pupal stage was longer when the larvae were fed on peanut leaves (9.21 days) and on the basis of the artificial diet (9.41 days) compared to the pupae whose larvae were fed on based on maize, millet, sorghum and cowpea (8.46; 8.40; 8.13; 8.93, respectively) (P-value = 5.2e-09). The duration of the larval stage was longer when the larvae were fed cowpeas (21.46 days) and peanuts (18.93 days).

Egg incubation time

Statistical analysis of the results showed that there is no significant difference between the average incubation time of eggs on the different food sources (F = 0.276; P-value = 0.92) (Table 2).

Table 2: Incubation time (Average, standard error, min, max) in days of *Spodoptera frugiperda* eggs

Regimes	Average	Std. Error	Min.	Max.
Peanut	2.50	0.50	2	4
Maize	2.25	0.25	2	3
Millet	2.75	0.47	2	4
Cowpea	2.50	0.28	2	3
Artificial regime	2.25	0.25	2	3
Sorghum	2.50	0.28	2	3

P = 0.92; Df = 4; Df Error = 18

Larval stage

Statistical analysis of the results by ANOVA shows that there is a significant difference (P-value = 2.334e-13) in the duration of the larval stage of *Spodoptera frugiperda* between the different food sources on which the study is based (Table 3). The duration of the larval stage is longer for larvae fed with cowpea leaves (21.46 ± 0.42) and peanuts (18.93 ± 0.18) The mean structuring test made it possible to obtain 4 statistically different groups (figure 1).

Table 3: Duration (Average, standard error, min, max) in days of the larval stage of *Spodoptera frugiperda* fed on different food sources

Régimes	Average	Std. Error	Min.	Max.
Peanut	18.93	0.18	18	20
Maize	12.40	0.29	11	14
Millet	13.46	0.16	13	15
Cowpea	21.46	0.42	19	24
Artificial regime	13.06	0.25	12	15
Sorghum	12.93	0.34	11	16

Df = 5; P = 2.334e-13

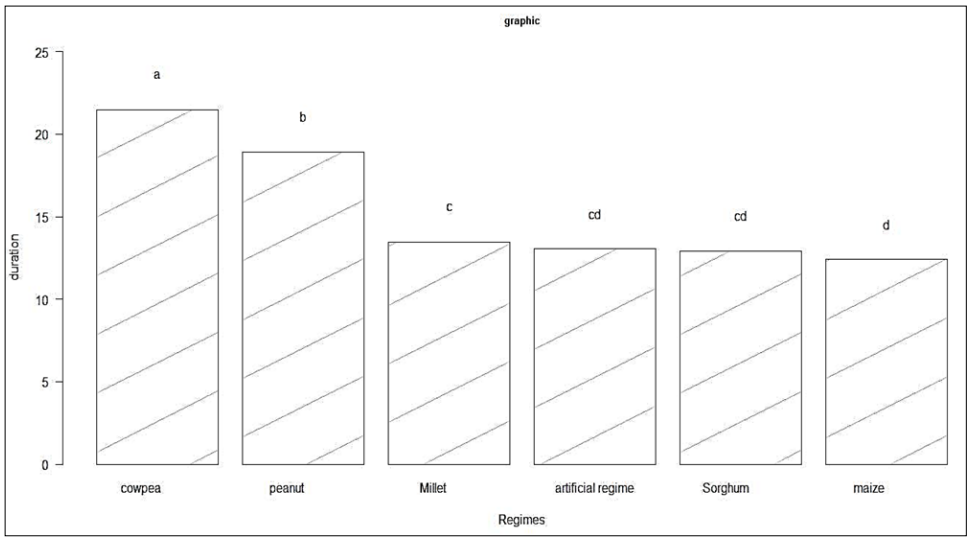

Fig 4: Bar diagram of the structure of the mean duration of the larval stage of *Spodoptera frugiperda* on different food sources

Pre-pupal stage

Statistical analysis of the results shows that there is no significant difference between the mean duration of the pre-pupal stage of *Spodoptera frugiperda* on the different food sources (P = 0.393) (Table 4).

Table 4: Duration (Average, standard error, min, max) in days of the pre-pupal stage of *Spodoptera frugiperda* on different food sources

Regimes	Average	Std. Error	Min.	Max.
Peanut	1.0	0.00	1	1
Maize	1.1	0.10	1	2
Millet	1.1	0.10	1	2
Cowpea	1.2	0.13	1	2
Artificial regime	1.0	0.00	1	1
Sorghum	1.0	0.00	1	1

F-value = 1,059; P-value = 0,393; Df Error = 54

Pupal stage

Statistical analysis shows that there is a significant difference between the duration of the pupal stage of *Spodoptera frugiperda* on different food sources (P = 5.2e-09). Table 5 and Figure 2 show that the duration is longer for pupae whose larvae are fed with peanut leaves (9.26 ± 0.11) and based on the artificial diet (9.40 ± 0, 16). The test of structuring of means made it possible to obtain 4 statistically different groups (Figure 2)

Table 5: Duration (Average, standard error, min, max) in day of the pupal stage of *Spodoptera frugiperda* on different food sources

Regimes	Average	Std. Error	Min.	Max.
Peanut	9.26	0.11	9	10
Maize	8.46	0.13	8	9
Millet	8.40	0.13	8	9
Cowpea	8.93	0.20	8	10
Artificial regime	9.40	0.16	8	10
Sorghum	8.13	0.09	8	9

Df = 5; P = 5.2e-09

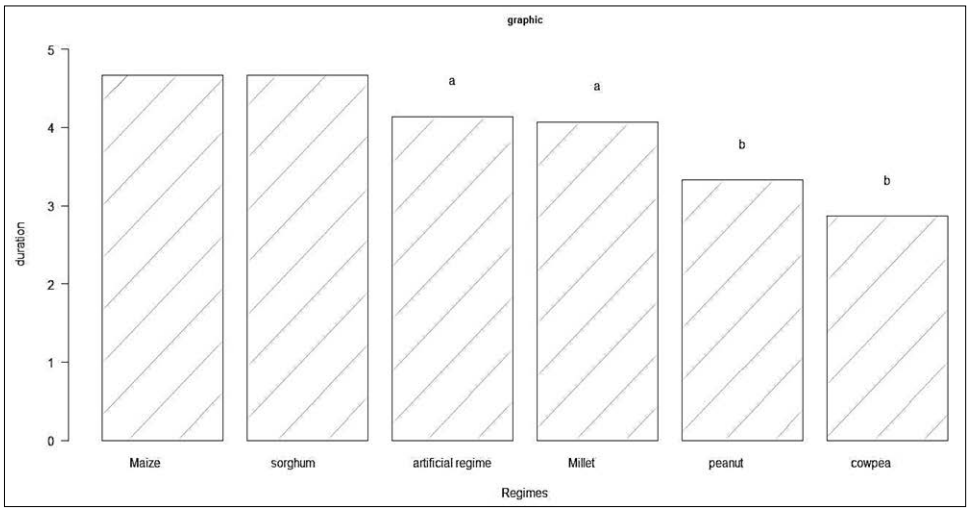

Fig 5: Bar diagram of the structure of the mean duration of the pupae stage of *Spodoptera frugiperda* on different food sources

Adult stage

Statistical analysis of the results shows that there is a significant difference in the duration of the adult stage of *Spodoptera frugiperda* on the different food sources in our study (P = 1.25e-05). The duration is shorter when the larvae are fed with cowpea (2.86 ± 0.21) and peanut (3.33 ± 0.21) leaves (table 6 and figure 3). The test of structuring of means yielded 2 statistically different groups (Figure 4).

Table 6: Duration (Average, standard error, Min, Max) in days of the adult stage of *Spodoptera frugiperda* on different food sources

Regimes max.	Average	Std. Error		Min.
Peanut	3.33	0.23	2	5
Maize	4.66	0.21	4	6
Mil	4.06	0.22	3	5
Cowpea	2.86	0.21	2	4
Artificial regime	4.13	0.37	2	6
Sorghum	4.6	0.31	2	6

P = 1.25e-05 ***; Df Error = 84

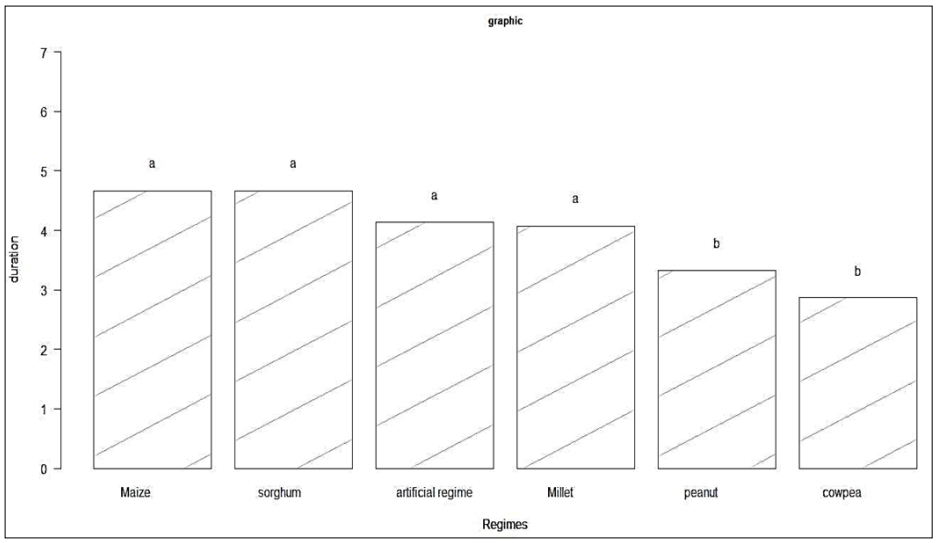

Fig 6: Bar diagram of the structure of the mean duration of the adult stage of *Spodoptera frugiperda* on different food sources

Weight of pupae

Statistical analysis of the results shows that there is a significant difference in the weight of the pupae of the Fall Armyworm on the different food sources in our study (P-value = 2e-16). Pupae whose larvae are fed with cowpea leaves have the lowest weight (0.138 ± 0.008) and those whose larvae are fed on the basis of the artificial diet the highest weight (0.281 ± 0.002) (Table 7). The test of structuring of means yielded 3 statistically different groups (Figure 4).

Table 7: Weight (Average, standard error, min, max) in grams of the pupal stage of *Spodoptera frugiperda* on different food sources

Regimes	Average	Std. Error	Min.	Max.
Peanut	0.177	0.001	0.13	0.20
Maize	0.173	0.006	0.14	0.20
Millet	0.147	0.009	0.10	0.19
Cowpea	0.138	0.008	0.09	0.17
Artificial regime	0.281	0.002	0.27	0.29
Sorghum	0.167	0.007	0.13	0.20

F-value = 52.17; Df Error = 54; P-value = 2e-16 ***

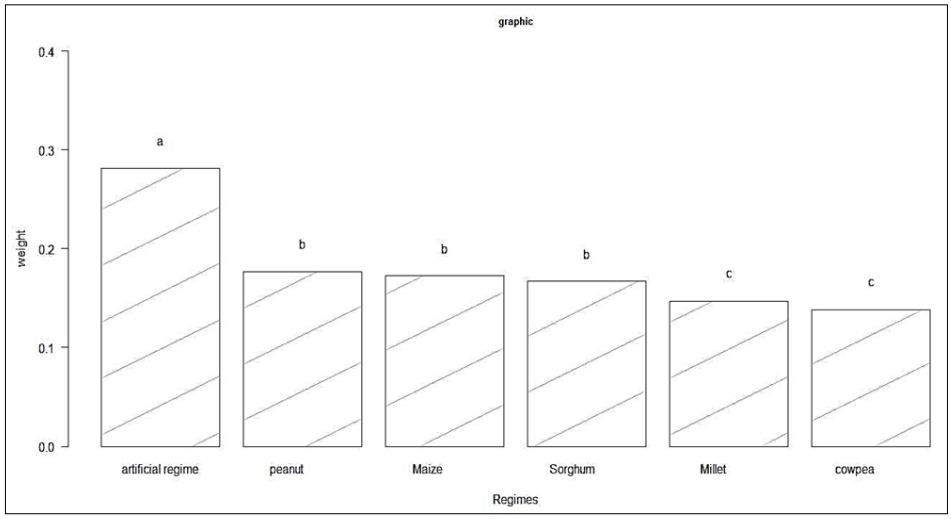

Fig 7: Bar diagram of the structure of the average weights of the chrysalis of *Spodoptera frugiperda* on different food sources

Larva-adult duration

Statistical analysis of the results shows that there is a significant difference (P-value = 2e-16) in the duration of the larva-adult of *Spodoptera frugiperda* between the different food sources on which the study is based (Table 8). The duration is longer for the larvae fed with cowpea leaves (29.66 ± 0.55) and peanuts (28.93 ± 0.21). The mean structuring test allowed to obtain 6 groups statistically different (figure 5).

Table 8: Duration (Average, standard error, min, max) in days of the larva-adult cycle of *Spodoptera frugiperda* fed on different food sources

Regimes	Average	Std. Error	Min.	Max.
Peanut	28.33	0.21	28	29
Maize	21.66	0.21	21	22
Millet	22.33	0.21	22	23
Cowpea	29.66	0.55	28	31
Artificial regime	23.00	0.36	22	24
Sorghum	20.83	0.16	20	21

P-value = 2e-16 ***; Df = 5; Df error = 30

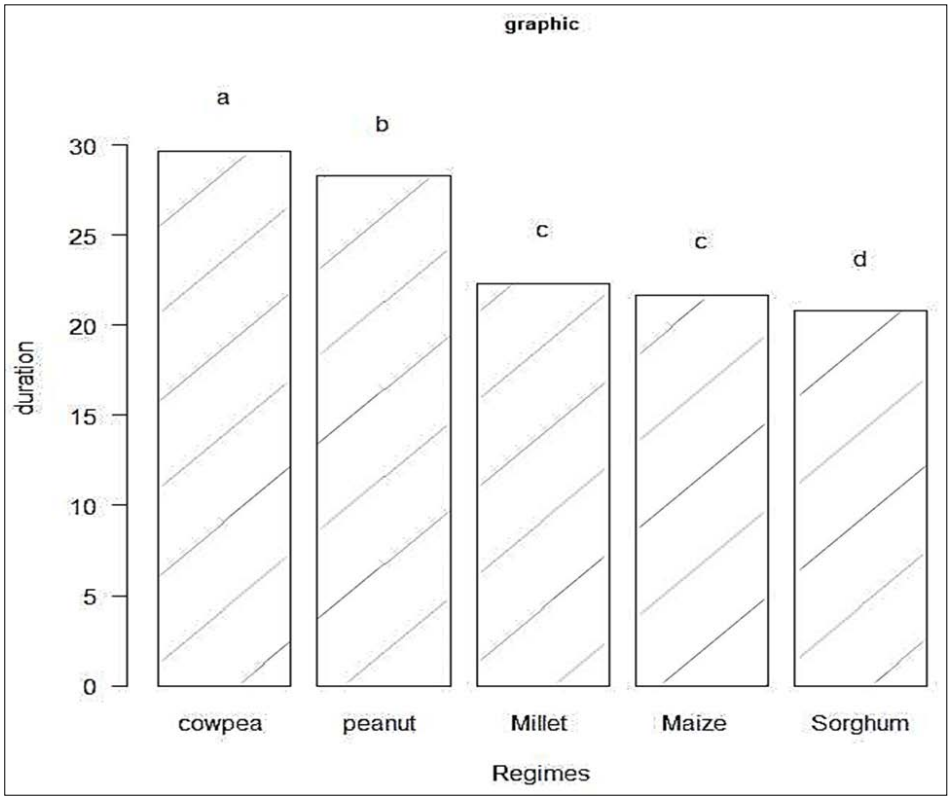

Fig 8: Bar diagram of the structure of the mean durations of the larva-adult cycle of *Spodoptera frugiperda* on different food sources

Sex ratio

The sex ratio (number of emerged females/number of males) is 0.69 ± 0.10.

Table 9: Sex ratio of *Spodoptera frugiperda* according to diet

Régimes	Sex-ratio
Millet	0.67
Maize	0.83
Sorghum	0.17
Artificial regime	0.67
Cowpea	0.25
Peanut	0.60

Discussion

The present study is intended as a contribution to a better knowledge of the biology of *Spodoptera frugiperda*. It is based on the use of five plant species cultivated in Senegal and an artificial diet as a source of food for the larvae.

The results obtained from the incubation of *Spodoptera frugiperda* eggs show that the hatching of the latter occurs after a period varying between 2.25 ± 0.25 and 2.75 ± 0.47 days of incubation under conditions of laboratory with a temperature of 26 °C ± 2 °C and a relative humidity of 70 ± 10%. These results are different from those obtained by [11] who had an incubation period of 5 ± 1 days with a temperature of 25 °C. This difference can be explained by factors such as temperature [12, 13] and to a lesser extent humidity [14]. Thus, under natural conditions, the incubation time of *Spodoptera frugiperda* eggs varies with the time of year and can range from 2 to 10 days [14].

Statistical analysis of the results by ANOVA shows that there is a significant difference (P-value = 2.334e-13) in the duration of the larval stage of *Spodoptera frugiperda* between the different food sources on which the study is based. It is longer when the larvae are fed with cowpea and peanut leaves. According to [15], the development of insects is generally influenced by the nutritional quality of the plant host on which the first instar larvae fed. And since the nutritional quality can vary from plant to plant, we can say that the fact that the larval stage of caterpillars fed on cowpea

leaves is 8 longer; 9.06; 8.53 and 8.4 days than those fed with leaves of millet, corn, sorghum and the artificial diet, respectively, is related to a difference between the nutritional quality of different host plants. The same goes for larvae fed on peanut leaves, the larval stage of which is 5.47; 6.53; 6; 5.87 days longer than those fed with leaves of millet, maize, and sorghum and on the basis of the artificial diet, respectively. Cowpea and peanut are therefore the least suitable plants among the 5 plants evaluated for the larval development of *Spodoptera frugiperda* since the larvae fed with these two plants (cowpea and peanut) have a longer larval stage than the others (millet, corn sorghum and artificial diet). This extension of the duration of the larval stage when the larvae are fed with cowpea and peanut leaves is a compensatory action of the larvae in order to reach the adequate weight to pupate [16]. The duration of the larval stage has an impact on the larva-adult duration. Thus, for the larvae fed with plants of low nutritional quality (cowpea and peanut) for *Spodoptera frugiperda* the duration of the larva-adult stage is longer (Table 9).

Our study to compare the biological characteristics of this pest when fed on different host species cultivated in Senegal mainly in the rainy season is crucial for a better understanding of the infestation of these host plants by *Spodoptera frugiperda*. Host plant nutritional factors in addition to moth immigration and emigration may contribute to changes in population dynamics of *Spodoptera frugiperda* in an agricultural landscape [17]. The fact that the larval stage of Fall Armyworm (FAW) is longer when the larvae are fed cowpea and peanut leaves may help reduce *Spodoptera frugiperda* populations. Indeed, the longer the duration of the larval stage, the greater the risk of the larvae being attacked by predators.

Conclusion

These results has shown that the cycle length of *Spodoptera frugiperda* is influenced by the diet. Thus, among the various food sources (millet, sorghum maize, peanut, cowpea and artificial diet) concerned by the present study, niébé and peanut were the least favourable for the development of *Spodoptera frugiperda* with a larva-adult stage duration of 29.66 ± 0.55 and 28.93 ± 0.21 days respectively, unlike millet, maize, sorghum and artificial diet which were more favourable to the development of *S. frugiperda* with respectively a duration of the larva-adult stage of 22.33 ± 0.21; 21.67 ± 0.21; 20.83 ± 0.16 and 23.00 ± 0.36 days. However, despite the fact that the cowpea and peanut were not adequate for the development of the fall armyworm, the larvae of the latter could cause damage to these two plant species because they could still allow them to complete their development cycle.

Moreover, *S. frugiperda* used in our trials was originally collected in maize, corresponding to the maize strain. Other *S. frugiperda* strains might lead to different results which still need to be studied in future research projects

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