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Effect of Insect Pollination on The Yield of *Sesamum indicum* (Pedaliaceae) In Maroua-Cameroon

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Abstract

From July to October 2015 and 2018 at Maroua, *Sesamum indicum* flowers were observed to determine the reproductive system, specific richness, relative frequency, and impact of the insects on the yields. 7800 and 4560 flower clusters for each studied were labeled and divided into two treatments, differentiated according to the presence or absence of protection against insect's activities. Observations were made on free-pollinated *S. indicum* flowers from September to October 2015 and 2018. The sesame is a mixed pollination plant, and the specific richness of the flowering insects was 19 species. The insects foraged the flowers of the sesame from 7:00 am to 4:00 pm, with the peak of the activity of the set of visits of them located between 9:00 am, and 10:00 am (28.59 %). These insects developed and elaborated behavior when they collected the nectar and pollen. Free-pollination produced more yield than pollination restricted with gauze bags. The influence of the insects estimated at 50.84 %, 17.22 %, 58.77 %, and 09.55 % for the fruiting rate, the number of seeds/fruit, the percentage of the average weight of seeds, and the percentage of normal seeds respectively. This study provided some knowledge about the insect's diversity of this Pedaliaceae, which can be exploited to improve fruit production. In order to improve the yield of *S. indicum*, pollinators could be protected by rational pest management tactics. Pesticide treatments should be done in the late afternoon to protect the pollinators for high seed yield or spray at a time of day when crop flowers are closed.

Keyswords: *Sesamum Indicum*, Flowers, Insects, Pollination, Yields

Introduction

In Cameroon, for more than two decades, research has been carried out on the interdependence between cultivated plants and their pollinating insects, particularly in the Center (Tchuenguem, 1993), West (Dongock *et al.,* 2004), North West (Otiobo *et al.,* 2016), Adamawa (Tchuenguem and Népidé, 2018), Littoral (Taimanga and Tchuenguem, 2018) and of the Far North (Pando *et al.*, 2019) Regions. Despite this important work, information is still lacking on the relationships between most of the plants cultivated in Cameroon and their flowering insects. However, it is known that pollinating insects consider ably increase the yields of plant species through the pollination of flowers during their foraging (Klein *et al.,* 2007).

Sesame (*Sesamum indicum* L.) is native to Asian and some African countries (Bedigian, 2003). It is one of the oldest crops in the world, cultivated for over 4,300 years in Babylon and Assyria (Hwang, 2005). Its cultivation has great economic potential, because of great demand, both nationally and internationally. The seeds, which contain about 50% oil, are the main reason for its cultivation and may be used in the food (Blal, 2013), pharmaceutical, and chemical industries (Elleuch *et al.*, 2007).

In Cameroon, the production of sesame seeds available to consumers is very low 43,498 tons/year (MINADER, 2012), is insufficient to meet demand estimated at 93,498 tones (DSCE, 2009). In fact, this country imports around 50,000 tons of sesame from Chad (Terra, 2015). It is therefore important to investigate how the production of this plant could be increased in the country.

Sesame has zygomorphic flowers with pendulous tubular corolla of 3-4 mm in length and coloring of various shades of purple-white (Georges, 1976). They occur singly or in groups of two to three in the leaf axils and are hermaphrodite (Free, 1993). The androecium consists of four stamens, two long (1.5-2.0 mm), and two short (1.0-1.5 mm), and the gynoecium has a superior ovary, multicarpelar and a long style (1.5-2.0 mm) with bifid stigma. The flower of sesame produces nectar and pollen, where attract various insect species (Free, 1993; Kamel *et al.*, 2013).

All over the world, data concerning the relationships between *S. indicum* and flowering insects are available but are insufficient. Panda *et al.* (1989) have shown that bees, Flies, and Butterflies are the predominant pollinators of *S. indicum* in India. In Brazil, Sarker (2000) reported that *Apis mellifera*, *Megachile* sp., and *Xylocopa olivacea* are the major pollinators of this plant. In Egypt, Mahfouz *et al.* (2012) and Kamel *et al.* (2013) have shown that the bees *Apis mellifera*, *A. dorsata*, *A. florea*, *Bombix priesneri,* and *Dielis collaris* are the most active pollinators on this Pedaliaceae.

In Cameroon, excepted the work carried out in Bamenda, Ngaoundéré, and Obala by Otiobo *et al.* (2016), Tchuenguem and Népidé (2018) and Pharaon *et al.* (2018) respectively, no other scientific production in this sense is available to our knowledge. It emerges from this work that insects significantly increase the yield thereof. Given the lack of scientific production on the pollination of *S. indicum* and its low yield, it is urgent to conduct additional studies in order to supplement the data already existing. In addition, according to Roubik (2000), the diversity and abundance of the pollinating entomofauna of a plant can vary in time and space. Thus, knowledge of the diversity of sesame pollinating insects should allow the breeding of potential effective pollinating insects of this Pedaliaceae.

The present work is a contribution of the knowledge of *S. indicum* pollinators in order to use their eco-systemic service in the resolution of sesame yield in Cameroon. This work has four specific objectives to: (a) determine the reproductive system; (b) identify the flowering insects of *S. indicum*; (c) assess the frequency of visits by these insects to flowers and the substances removed; (d) assess the impact of pollination by insects on fruit and seed yields. The information gained on the interaction of sesame flowers and insect floral visitors will enable farmers to develop management plans that will increase the overall quality and quantity of sesame yield.

Materials and methods

Site and biological materials

The research was carried out in Palar (10°36′16.7′′N, 14°16′ 36.5′′E and 416 m) in 2015 and Wourndé (10°38′15.7′′N, 14°18′40.4′′E and 437 m) in 2018, two localities of Maroua (Far North; Cameroon: Figure 1). These coordinates were obtained using a GARMIN Beingx 10 brand GPS. The experiments were developed each year from July to September, and fruit was harvested in October. The predominant climate in the region is Sahel-Sudanian, according to the Kuete *et al.* (1993) classification, and the average annual temperatures is 35° C and mean annual rainfall of 700 mm, with the rainy season from June to October. The choice of these observation sites is justified by the existence of peasant fields of other crops and the guarantee of safety of the experimental plots and of the observer. The plant material was represented by: (a) sesame seeds of variety S42 which were bought at the Pala market (Mayo-kebbi West; Chad); (b) the various plant species located near the experimental plots and which were in bloom at the same time as *S. indicum*. The animal material was represented by insects naturally present in the environment and which visited the flowers of *S. indicum*.

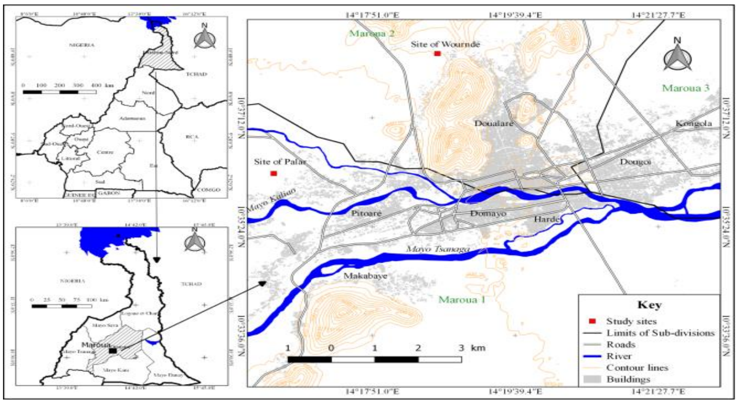

Figure 1: Map of Maroua town locating the experimental fields.

Methods

Preparation, sowing, and maintenance of the experimental plot

The experimental plot was a plot of 20 m long by 15 m wide for an area of 300 m^2. On July 13th, 2015, and July 10th, 2018, the following operations were successively carried out on the experimental plots: clearing, plowing, and training of the six sub-plots. On July 16th, 2015 in Palar and July 12th, 2018 in Wourndé, sowing was done, online on the sub-plots, at a rate of 11 lines per sub-plot. 5 seeds were sown per seed hole; the spacing was 15 cm between the rows; 60 cm on rows and 4 cm deep. The de-pairing was done when the plants had four leaves and two of the most vigorous plants were kept per hole. From germination (which occurred from July 20th, 2015, and July 18th, 2018) to the development of the first flower (September 25th, 2015, and September 22nd, 2018), hoe weeding operations were carried out regularly every two weeks. After the first weeding, the soil was amended with ash, and then the chemical fertilizer NPK (20-10-15) was according to the recommendations of Rongead (2013). From the flowering period to the ripening of the fruit, weeding was done regularly by hand, according to the recommendations of Pando *et al.* (2019).

Determination of the mating system

September 20th, 2015 and September 29th, 2018; 7800 and 4560 flowers of *S. indicum* at bud stage were labeled on 120 individual plants, and two treatments were set up as follow:

- Treatment 1 (2015) and 1' (2018), 3900 and 2280 unprotected flowers on which no insect was captured (Figure 2);
- Treatment 2 (2015) and 2' (2018), 3900, and 2280 flowers protected from insects using gauze bags (Figure 3).

Figure 2: Flowers of *Sesamum indicum* unprotected.

Figure 3: Flowers of *Sesamum indicum* bagged to prevent visitors.

Five days after the shedding of the last flower of a label, the number of pods was assessed in each treatment. The fruiting index (*Ifr*) was then calculated as described by Tchuenguem et al. (2014).

$$Ifr = (F_2 / F_1),$$

where F_2 is the number of pods formed and F_1, the number of viable flowers initially set.

The allogamy rate (*TC*) from which derives the autogamy rate (*TA*) was expressed as the difference in fruiting indexes between treatment X (unprotected flowers) and treatment Y (protected flowers):

$$TC = \{[(Ifr_X - Ifr_Y) / Ifr_X] * 100\} \text{ (Demarly, 1977)},$$

where Ifr_X and Ifr_Y are respectively the fruiting average of treatment X and treatment Y.

$$TA = 100 - TC. \text{ (Demarly, 1977)}$$

Assessment of the diversity and relative frequency of *Sesamum indicum* flowering insects in each locality

To determine the diversity of flowering insects, during the study period, the insects were caught on the flowers in free pollination and not labeled by hand and with an entomological net. In the field, the captured insects were kept in bottles containing 70% ethanol, except for Lepidoptera, which were kept in foils, according to the recommendations of Borror and White (1991). On the label corresponding to each specimen, the time, date, and place of capture were noted, and the floral products harvested or sought. For species not precisely identified in the field, they have been assigned a code and description to facilitate observations. The determination of the specimens was made at the Laboratory of Biological Sciences of the Higher Teachers' Training College of the University of Maroua using in particular reference collections and identification keys from Delvare and Arbelenc (1989), Eardley *et al.* (2010) and Pauly (2014).

To assess the frequency of visits of the different insects, observations were made every day during the flowering period on flowers of treatments 1 and 1' (unprotected), according to the 7:00-8:00 am time slots, 9:00-10:00 am, 11:00-12:00 am, 1:00-2:00 pm and 3:00-4:00 pm. On each pass, the different insects were counted on the blooming flowers. Since the insects were not marked, the cumulative results were expressed by the number of visits (Tchuenguem *et al.,* 2004). The data obtained made it possible to determine the frequency of each insect species (Fi) on the flowers of *S. indicum*, according to the following formula:

$$F_i = \{[V_i / V_T] * 100\} \text{ (Tchuenguem et al., 2004)},$$

where V_i the number of visits of the insects I on the flowers of treatment A and V_T the number of visits of all the insects on these same flowers.

To appreciate the specific richness of the diversity of flowering insects in this Pedaliaceae, the Shannon diversity indices (H) and the Piélou equitability (EQ) were calculated using the formula:

$$H = -\sum_{i=1}^{S} pi(\log_2 pi) \text{ (Dajoz, 2008) and } \mathbf{EQ} = \frac{H}{\log_2 (S)} \text{ (Dajoz, 2000)},$$

where pi = ni/N; ni: number of individuals of I (corresponding to the number of visits of i); N: total number of individuals (corresponding to the total number of visits) and S: total number of species observed. The Jacard index (*J*) was calculated to determine the similarity between the two sites:

$$J = \frac{c}{a+b-c} \text{ (Jaccard, 1908)},$$

where *a* = number of species from list 1 (record A), *b* = number of species in list 1′ (survey B), and *c* = number of species common to surveys 1 and 1′.

Détermination De L'impact Des Insectes Floricoles Sur La Pollinisation De *Sesamum indicum*

The assessment of the impact of insects on the pollination of *S. indicum* was noted throughout the study of the frequency of visits. This was to record the number of times an insect came into contact with the stigma or anther of the flower visited. This made it possible to highlight the possibilities for insect intervention in self-pollination and cross-pollination of the flowers visited (Delaplane *et al.,* 2013). To determine the different categories of pollinators, the regularity index (R) was calculated using the formula:

$$R = [(P / 100) * (f/100)]$$ (Tchuenguem, 1993),

where *P* is the percentage of insect visits, and f is the relative frequency of insect visits.

Assessment of The Effect of Insects on *Sesanum indicum* Yield

This evaluation was based on the impact of insects visiting flowers on pollination, the impact of pollination on fructification of *S. indicum* and the comparison of yield (fruiting rate, mean number of seeds/pod, mean mass of seeds and percentage of normal seeds) of treatment X (unprotected flowers) and treatment Y (protected flowers). The number of pod/plant due to the influence of foraging insects (*Fri*) was calculated by the formula:

$$Fr_i = \{[(Fr_X - Fr_Y) / Fr_X] *100\}$$ (Tchuenguem *et al.*, 2004),

where Fr_X and Fr_Y were the number of pod in treatment X and treatment Y. The fruiting rate of treatment (Fr) is:

$$Fr = [(F_2/F_1) *100]$$ (Tchuenguem *et al.*, 2004),

where F_2 is the number of pods formed and F_1, the number of viable flowers initially set. At maturity, pods were harvested from each treatment, and the number of seeds per pod counted. The mean number of seeds per pod and the percentage of normal seeds (well-developed seeds) were then calculated for each treatment. The impact of flowering insects on seed yields was evaluated using the same method as mentioned above for fruiting rate.

Data Analysis

Data were analyzed using:

- descriptive statistics (for calculating averages, standard deviations, and percentages);
- three tests: The Chi-square (χ2) for the comparison of the percentages (Schwartz, 1984); the Z test for the comparison of the averages of two samples; Student's t modified for comparison of site-specific diversity indices were calculated by the formula:

$$t = \frac{|H_1 - H_2|}{\sqrt{Var(H_1) + Var(H_2)}}$$ (Mathew *et al.*, 1998),

where H_1 and H_2 the Shannon-Wiener to sites 1 and 2, respectively.

- the XLSTAT 14.1 software.

Results and Discussion

Reproduction System of *Sesamum indicum*

The fruiting index of *S. indicum* was 0.80 (n = 300; s = 0.21), 0.42 (n = 300; s = 0.20), 0.54 (n = 300, s = 0.12) and 0.25 (n = 300, s = 0.17), respectively for treatment 1, treatment 2, treatment 1' and treatment 2'. Thus, allogamy rate was 46.89 %, and the autogamy rate was 53.11%. It appears that the results of fruits harvested show that sesame is a plant with mixed pollination, which produce fruit by both autogamy (auto and self-pollination) and crossbreeding (cross-pollination), like higher plants (Delaplane *et al.*, 2013). The significantly higher (χ^2 = 2793.60; $p < 10^{-9}$) fruit yield as a function of the self-pollination in relation to cross-pollination agrees Weiss (1983), who stated that sesame is a predominantly autogamous species, which could only and possibly present allogamy above 10%. These results contradict those of Andrade *et al.* (2014), who indicate the best performance of cross-pollination than self-pollination. This probably is due to exchange of genetic material promoted by this type of pollination, increasing the heterozygosity of the embryo formed in the seeds, which according to Mahfouz *et al.* (2012), enhances the quality and quantity of seeds produced, and anticipates the period of ripening and harvest.

Diversity and Frequency of Floral Entomofauna of *Sesamum indicum*

Table 1 shows the diversity, frequency of visits, and floral products harvested by insects observed on the flowers of *S. indicum* in Maroua.

Table 1: Diversity, frequency of visits, and floral products harvested by insects in the flowers of *Sesamum indicum* in Maroua in 2015 and 2018.

Insects			**Palar 2015**		**Wourndé 2018**		**Maroua II**	
Order	**Family**	**Genres and species**	n_1	P_1 (%)	n_2	P_2 (%)	n_t	P_t (%)
Hymenoptera	Megachilidae	*Megachile aurifera* NP	1838	**52.89**	//	//	1838	**40.99**
		Chalicodoma parieta NP	357	10.27	10	00.99	367	08.18
		Chelostoma sp. N	//	//	14	01.39	14	00.31
	Apidae	*Amegilla* sp. NP	544	15.65	//	//	544	12.13
		Xylocopa olivacea NP	//	//	458	**45.48**	458	10.21
		Apis mellifera adansonii NP	//	//	360	35.75	360	08.03
		Xylocopa torrida NP	//	//	86	08.55	86	01.92
		Xylocopa violacea N	//	//	15	01,49	15	00.33
		Xylocopa inconstans NP	111	03.19	//	//	111	02.48
	Halictidae	*Lasioglossum* sp. NP	//	//	33	03.28	33	00.21
		Crocisaspidia chandleri N	//	//	11	01.09	11	00.74
	Vespidae	*Belonogaster juncea juncea* N	196	05.64	10	00.99	206	04.59
		Polistes canadensis N	//	//	12	01.19	12	00.27
Total	**04**	**14**	**3046**	**87.65**	**1009**	**100.00**	**4055**	**90.43**
Lepidoptera	Nymphalidae	*Acraea serena* N	94	02.70	//	//	94	02.09
	Pieridae	*Eurema lactasana* N	83	02.38	//	//	83	01.85
		Catopsilia frorella N	**110**	**03.16**	//	//	110	02.48
Total	**02**	**3**	**287**	**08.26**	**//**	**//**	**287**	**06.40**
Diptera	Muscinae	*Musca domestica* N	86	02.47	**//**	**//**	**86**	01.92
Orthoptera	Tettrigoniidae	*Neoconocephalus robustus* CP	22	00.63	**//**	**//**	**22**	00.49
Coleoptera	Chrysomelidae	*Chrysomela* sp. CP	34	00.97	**//**	**//**	**34**	00.76
	03	3	**142**	**04.09**	**//**	**//**	**142**	**03.17**
Total	**09**	**19**	**3475**	**100.00**	**1009**	**100.00**	**4484**	**100.00**

n_1: number of visits to 3900 flowers in 10 days in 2015; n_2: number of visits to 2280 flowers in 9 days in 2018; n_t: number of 6180 flowers visits in 19 days in 2015 and 2018; P_1: visit percentage in 2015, P_2: visit percentage in 2018; P_t: visit percentage in 2015 and 2018 with $P_1 = (n_1/3475)*100$; $P_2 = (n_2/1009)*100$; $P_t = (n_t/4484)*100$; sp. : species not determined; //: no visit; NP: harvest of nectar and pollen; N: nectar harvest; P: pollen collection; CP: consumer of petals.

It appears from this table that five groups of pollinators visited the sesame belonging to order Hymenoptera, Diptera, Lepidoptera, Coleoptera, and Orthoptera of class insecta during the flowering period. The number of Hymenoptera was higher (90.43%), followed by Lepidoptera (06.40 %), and then both of Diptera (01.92 %), Orthoptera (00.76 %) and Coleoptera (00.49 %). The results indicate that Hymenopterans are the major pollinators visiting sesame flowers. These findings are in close agreement with Mahfouz *et al.* (2012) in Suez and Otiobo *et al.* (2016) in Bamenda, who studied the relative abundance of pollinator fauna of sesame during two successive seasons. Hymenopterans insects were higher, followed Dipterans and Lepidopterans. Also, Pharaon *et al.* (2018) in Obala reported nine species of Hymenopterans as predominant visitors of sesame flowers. Among the 3475 and 1009 visits of 11 (Palar) and 10 (Wourndé) insect species recorded on *S. indicum* flower, *Megachile aurifera* and *Xylocopa olivacea* were the most represented insects with 1838 visits (52.89%) and 458 visits (45.48%), in 2015 and 2018, respectively. However, in other parts of the world, such as Egypt and India, Mahfouz *et al.* (2012) and Sajjanar and Eswarappa (2015) reported that other bees *Apis mellifera* and *Apis dorsata*, respectively, have been reported as the main floral visitors of this crop. This could be due to the absence of this bee in those countries or its lower abundance.

The total species richness of *S. indicum*'s flowering insects was 19 in Maroua. This specific richness is far superior to that found by Tchuenguem and Népidé (2018) at Ngaoundéré, which was four species. The comparison between the two specific richness is highly significant (t = 723.81 [df = 1089; P <0.001]). This agrees the research of Roubik (2000), which revealed that the diversity and specific richness of a plant's flowering entomofauna may vary in space and time.

It appears from table 1 against that there are three categories of insects: (a) species exclusively in search of nectar which was represented by *Chelostoma* sp., *Xylocopa violacea*, *Crocisaspidia chandleri*, *Belonogaster juncea juncea, Polistes canadensis, Acraea serena, Eurema lactasana, Catopsilia frorella* and *Musca domestica*. Concerning Lepidoptera, Benachour (2008) reported in Algeria that these insects harvest only nectar on flowers. (b) Species in search of nectar and pollen were represented by: *Megachile aurifera*, *Chalicodoma parietal, Amegilla* sp., *Xylocopa olivacea, Apis mellifera adansonii, Xylocopa torrida*, and *Xylocopa inconstans*. During this flowering period, these insects that collected pollen and nectar from visited the flowers of *S. indicum*, intensely collected nectar as pollen. These findings are in conformity with the observations made by Tchuenguem and Népidé (2018), who reported that the Hymenoptera harvested intensively the nectar than pollen. (c) Species consuming petals: *Neoconocephalus robustus* and *Chrysomela* sp. It should be noted that bees visit sesame flowers for nectar and pollen, but flies, butterflies, and lady beetles visit flowers for feeding on nectar only or waiting for their preys or feeding on different parts of sesame and sometimes just for resting.

The diversity indices of Shannon-Weaver (H_1) was 2.61 at Palar and 1.97 (H_2) at Wourndé. The difference between the Shannon-Weaver diversity indices of the two sites is not significant (t = 2.03 [df = 1234; p >0.05]). Piélou's equitability (EQ) was 0.63 and 0.62, respectively, in Palar and Wourndé. The Piélou equitabilities of the two sites being very close, this would suggest that the two sites have nearly the same environmental conditions. As the Jacard index (J = 0.12) is low, the species for the two habitats compared are very different, indicating that the different habitat conditions determine a turnover of the important species. This result is in agreement with the similar findings by Pando *et al.* (2019) on *Glycine max* at Mayel-Ibbé and Wourndé.

Table 2 presents the number and percentage of days of visits of the different flowering insects from *Sesamum indicum*. It appears from this table that the frequency of each insect species is varied. We obtained three categories of frequencies: (a) frequent visitors ($f > 50$): *Megachile aurifera, Chalicodoma parietal, Amegilla* sp. and *Belonogaster juncea juncea*; (b) visitors with average frequencies ($25 < f \leq 50\%$): *Xylocopa olivacea, Apis mellifera adansonii, Xylocopa torrida, Xylocopa inconstans, Acraea serena, Eurema lactasana, Catopsilia frorella, Musca domestica, Neoconocephalus robustus, and Chrysomela* sp.; (c) Rare visitors ($f \leq 25\%$) represented by *Chelostoma* sp., *Xylocopa violacea, Lasioglossum* sp., *Crocisaspidia chandleri,* and *Polistes canadensis*. The high frequency of some species is due to their attachment to the pollen and/or nectar of *S. indicum*. The Apidae, Megachilidae, and Halictidae family's species, pollen is indispensable for their nutrition Roubik (1989). For Halictidae, this result agreed that Pando *et al.* (2019) were found on *Glycine max* flowers. However, the pollen of *S. indicum* is strongly accessible to genus *Xylocopa* except for *Xylocopa olivacea*. In addition, the attractive nature of its flowers with insects is due to the color of the flowers, which is purple, the most attractive color, according to Faegri and Piji (1971).

Table 2: Number and percentage of days of visits of the different flowering insects from *Sesamum indicum* to Maroua II in 2015 and 2018.

Insects	Palar 2015		Wourndé 2018		Maroua II	
	n₁	***f₁ (%)***	***n₂***	***f₂ (%)***	***fₜ***	***fₜ (%)***
Megachile aurifera	10	100.00	//	//	10	**52.63**
Chalicodoma parieta	10	100.00	3	33.33	13	68.42
Chelostoma sp.	//	//	3	33.33	3	15.79
Amegilla sp.	10	100.00	//	//	10	52.63
Xylocopa olivacea	//	//	9	100.00	9	47.37
Apis mellifera adansonii	//	//	9	100.00	9	47.37
Xylocopa torrida	//	//	6	66.67	6	31.58
Xylocopa violacea	//	//	4	44.44	4	21.05
Xylocopa inconstans	8	80.00	//	//	8	42.11
Lasioglossum sp.	//	//	4	44.44	3	15.79
Crocisaspidia chandleri	//	//	2	22.22	2	10.53
Belonogaster juncea juncea	10	100.00	1	11.11	11	57.89
Polistes canadensis	//	//	3	33.33	3	15.79
Acraea serena	7	70.00	//	//	7	36.84
Eurema lactasana	7	70.00	//	//	7	36.84
Catopsilia frorella	8	80.00	//	//	8	42.11
Musca domestica	5	50.00	//	//	5	26.32
Neoconocephalus robustus	5	50.00	//	//	5	26.32
Chrysomela sp.	6	60.00	//	//	6	31.58

n_1: number of days of presence of insects during N_1 observation days in 2015; n_2: number of days of presence of insects during N_2 observation days in 2018; n_t: number of days of presence of insects during N_t observation days in 2015 and 2018; f_1 (%): Relative frequency of insect visits $(n_1/N_1)*100$; f_2 (%): Relative frequency of insect visits $(n_2/N_2)*100$; f_t (%): Relative frequency of insect visits $(n_t/N_t)*100$; $N_1 = 10$, $N_2 = 9$, $N_t = 19$

Rhythm of Visits According to Time and Observation Days

Table 3 shown that the insects visited *S. indicum* flowers from 7:00 am to 4:00 pm, the daily foraging period varied with insect's species in both survey sites and the peak of activity situated between 9:00 am and 10:00 am. During this period of the day, the mean hygrometry (70.18%), along with the mean temperature (28.78°C) are high and could therefore be favorable to the high availability of nectar that attract insects (Bramel *et al.*, 2004).These conditions might partially justify the highest frequency of insect visits during that time frame.

Data in table 3 showed the foraging activity of the major insect orders visiting sesame during flowering period. Peak of foraging activity was observed in Hymenoptera, Lepidoptera and other (Diptera, Orthoptera , and Coleoptera) orders during 9-10 am, 11-12 am and 7-8 am respectively in our study. For Hymenopteran, the same observations were reported by Mahfouz *et al.* (2012) in Egypt.

The comparison among number of different bee species clearly showed that the number and foraging activity of *Megachile aurifera* higher than all bee at all five time period i.e., 7-8m, 9-10 am, 11-12 am, 1-2 pm and 3-4 pm except for *Amegilla* sp. that the most intense foraging activity at the first hour of observation (Table 3). The maximum number of *M. aurifera* was observed during 11-12 am and decreased with time during the day. This is because nectar flow is copious in the sesame crop, especially in the morning period; after that, the nectar concentration gradually diminishes due to the high temperature and the low relative humidity. Although foragers preferred warm or sunny days for the good floral activity, the negative influence of the up temperature is higher on the plant as pollen and nectar producer than on the foragers. Thus, Pesson and Louveaux (1984) shown that the temperature allows floral anthesis, and accelerates flower wilting or closing when rising. In the same order, the rainfall was documented as an environmental factor that can disrupt the floral insect activity (Bramel *et al.*, 2004). According to Kasper *et al.* (2008), when the floral products are not easily reached or when its quantity and/or quality decrease, foragers reduce their activity on flowers (to make the working energy lower than that of harvesting products energy). The Same observation was reported by Munir and Aslam (2002) in the same plant in Egypt. In fact, these insect species do not visit *S. indicum* flowers when they are poor in nectar and pollen after 4 pm. Moreover, according to Bramel *et al.* (2004), a higher temperature along with a very weak relative humidity has a negative influence on the activity of pollinators on flowers.

Table 3: Rhythm of insect visits according to observation time periods at Maroua

Insect species	Daily periods										Total
	7:00-8:00 am		9:00-10:00 am		11:00-12:00 am		1:00-2 :00 pm		3:00-4:00 pm		
	NV1	*P_1 (%)*	*NV_2*	*P_2 (%)*	*NV_3*	*P_3 (%)*	*NV_4*	*P_4(%)*	*NV_5*	*P_5 (%)*	
M. aurifera	179	09.74	425	23.12	646	35.15 *	390	21.22	198	10.77	1838
C. parieta	23	06.27	101	27.52	114	31.06*	26	07.08	103	28.07	367
Chelostoma sp.	//	//	//	//	//	//	7	50.0*	7	50.00*	14
Amegilla sp.	229	42.10*	163	29.96	74	13.60	42	07.72	36	06.62	544
X. olivacea	3	00.65	253	55.24*	59	12.88	97	21.17	46	05.89	458
A. m. adansonii	//	//	55	15.27	152	42.22*	108	30.00	45	07.50	360
X. torrida	//	//	52	60.46*	4	04.65	8	09.30	22	25.58	86
X. violacea	10	66.66*	//	//	//	//	5	33.33	//	//	15
X. inconstans	6	05.41	90	81.08*	//	//	15	13.51	//	//	111
Lasioglossum sp.	//	//	8	24.24	9	27.27*	7	21.21	9	27.27*	33
Cr. chandleri	//	//	//	//	//	//	3	2727	8	72.72*	11
B. j. juncea	8	03.88	37	17.96	48	23.30	45	21.85	68	33.01*	206
P. canadensis	//	//	//	//	5	41,66*	2	16.66	5	41.66*	12
Hymenoptera	458	11.29	1184	**29.20***	1111	27.40	755	18.62	547	13.49	4055
A. serena	13	13.83	21	22.34	29	30.85*	21	22.34	10	10.64	94
E. lactasana	3	03.61	32	38.55*	19	22.89	18	21.69	11	13.26	83
Ca. frorella	2	01.82	16	14.55	32	29.09*	31	28.18	29	26.36	110
Lepidoptera	18	06.27	69	24.04	80	**27.88***	70	24.39	50	17.42	287

Mu. domestica	33	38.37*	10	11.63	11	12.79	10	11.63	22	25.58	86
N. robustus	4	18.18	7	31.82*	3	13.64	2	09.09	6	27.28	22
Chrysomela sp.	16	47.06*	12	35.29	4	11.77	2	05.88	//	//	34
Others	53	**37.32***	29	20.42	18	12.68	14	09.86	28	19.72	142
Total	529	11.78	1282	**28.59***	1209	26.96	839	18.71	625	13.94	4484

NV1: number of visits between 7:00 am and 8:00 am; *NV2*: number of visits between 9:00 am and 10:00 am; *NV3*: number of visits between 11:00 am and 12:00 am; *NV4*: number of visits between 1:00 pm and 2:00 pm; *NV5*: number of visits between 3:00 pm and 4:00 pm; P1: percentage of visits between 7:00 am and 8:00 am; *P2*: percentage of visits between 9:00 am and 10:00 am; *P3*: percentage of visits between 11:00 am and 12:00 am; *P4*: percentage of visits between 1:00 pm and 2:00 pm; *P5*: percentage of visits between 3:00 pm and 4:00 pm ; (*): peak of activity

Impact of Insects on *Sesamum indicum* Pollination

When collecting pollen and/or nectar on flowers of *S. indicum*, insects were frequently in contact with the anthers and the stigma of visited flowers. They could therefore be directly involved in self-pollination, by putting pollen of one flower on to the stigma of the same flower. Table 4 presents the percentage of contacts between insect, anther, and stigma of *S. indicum*.

It appears on that table that all the 19 insect species that visited the flowers had contact with the anthers and/or stigmas: (a) seven of these insect species have a frequency of contact with the anthers of 100%, nine have an incidence of contact with the anthers of between 50 % ≤ f <100% and one have a frequency of contact with the anthers of between 25 % ≤ *f* < 50 %; (b) twelve of these insect species have a frequency of contact with the stigma of 100%, seven have an incidence of contact with the stigma of between 50 % ≤ *f* <100% and no have a frequency of contact with the stigma of between 25 % ≤ *f* < 50 %. Individuals of each studied bee species were seen carrying pollen of *S. indicum* from flower to flower, using the legs, mouthparts, thorax, and abdomen. Therefore, they were likely playing a positive role in geitogamy (Robertson, 1992; Delaplane *et al*., 2013) by putting the pollen of one flower to the stigma of another flower of the same plant species. The foragers passing from flower to flower on different plants were seen carrying pollen from one plant to another. They could, therefore, allowed xenogamy (Delaplane *et al*., 2013), by putting the pollen of plant species to the stigma of another plant species. Several flowering insects in general and Apoidea family, in particular, were reported as being part of the pollinating entomofauna of *S. indicum* in Cameroon by other authors such as Otiobo *et al*. (2016), Pharaon *et al*. (2018) and Tchuenguem and Népidé (2018) at Bamenda, Obala, and Ngaoundéré respectively.

Table 4: Regularity index, numbers, and percentage of insect visits in contact with the anthers and stigma of *Sesamum indicum* flowers at Maroua.

Insectes	**Maroua**			***Nt***	***NV***			
	Palar	**Wourndé**	**Total**		**Anthers**		**Stigma**	
	R2015	***R2018***	***RT***		***na***	***Pa* (%)**	***ns***	***Ps* (%)**
Megachile aurifera	0.52890	//	0.21573	1838	1789	97.33	1801	97.99
Chalicodoma parieta	0.10270	0.00329	0.05596	367	367	100.00	367	100.00
Chelostoma sp.	//	0.00463	0.00048	14	14	100.00	14	100.00
Amegilla sp.	0.15650	//	0.06384	544	544	100.00	544	100.00
Xylocopa olivacea	//	0.45481	0.04836	458	378	82.53	397	86.68
Apis mellifera adansonii	//	0.35750	0.03803	360	303	84.67	360	100.00
Xylocopa torrida	//	0.05700	0.00606	86	23	26.74	72	83.72
Xylocopa violacea	//	0.00662	0.00069	15	//	//	15	100.00
Xylocopa inconstans	0.02552	//	0.01044	111	73	65.77	101	90.99
Lasioglossum sp.	//	0.01457	0.00033	33	33	100.00	33	100.00
Crocisaspidia chandleri	//	0.00242	0.00078	11	11	100.00	11	100.00
Belonogaster juncea juncea	0.05640	0.00109	0.02657	206	108	52.43	185	89.81
Polistes canadensis	//	0.00396	0.00042	12	12	100.00	12	100.00

Acraea serena	0.01890	//	0.00769	94	67	71.27	92	97.87
Eurema lactasana	0.01666	//	0.00681	84	53	65.09	84	100.00
Catopsilia frorella	0.02528	//	0.01044	110	101	73.64	110	100.00
Musca domestica	0.01225	//	0.00505	86	59	68.61	78	90.70
Neoconocephalus robustus	0.00315	//	0.00128	22	22	100.00	22	100.00
Chrysomela sp.	0.00582	//	0.00240	34	34	100.00	34	100.00

R = (P_n/100)*(f_n/100); P_n: percentage of insect visits (Table I); f_n: Relative frequency of insect visits (n_t/19)*100; n_t: number of visits studied; *NCV*: number of contact visits, n_t': Number of total visits, n_a: number of contact anther visits; P_a: percentage of anther contact visits; n_s: number of contact stigma visits; p_s: percentage of stigma contact visits

According to table 4, the different insect species found on *S. indicum* flowers can be classified into three categories of pollinators: (a) major pollinators which are characterized by a high regulatory index (R > 0.05) and has a high pollen harvesting rate; (b) minor pollinators which are characterized by a low regulatory index (0.05 < R < 0.01). This could be explained by the low number of the species present in the experimental field, or the species were preferentially in search of nectar. (c) Occasional pollinators, which are characterized with a very weak regulatory index (R < 0.01) and absence of behavior, link to the search of pollen and/or nectar but may have a destructive attitude. All these species of insects carry out foraging activities on the flowers of *S. indicum*, thus contribute to auto pollination and/or cross-pollination. These, therefore, ensure the diversity of the species and increase the seeds yield and produce.

Assessment of The Impact of Insects on Pods and Seeds Yield

The number of fruit set, five days after anthesis of flowers, and harvested showed significant differences (p < 0.05) between treatments of pollination in Maroua (Table 5).

In Table 5, We Documented:

1. High fruiting rate of pod formation during unprotected visits compared with bagged flowers. The comparison of the fruiting rate showed that the differences observed were highly significant between treatments 1 and 2 (χ^2_{2015} = 2821.60 [df = 1 ; P < 0.001]) and treatments 1′ and 2′ (χ^2_{2018} = 203.80 [df = 1 ; P < 0.001]). The results suggest that the type of pollination effect of the fruiting rate. So, the visit by biotic pollinators is important for sesame pollination, whether increasing the efficiency of pollen transfer within the same flower or even bringing pollen from other flowers, providing cross-pollination, which was also able to set fruit in sesame. Similar results were obtained in Egypt and Brazil by Mahfouz *et al.* (2012) and Andrade *et al.* (2014) respectively. The percentage of the fruiting rate attributed to the insects was 50.84 %. Otiobo *et al.* (2016) also were obtained the corresponding result with 27.35 % in Bamenda, which is inferior. This could be due to absence or its lower abundance of the same major pollinators.

Table 5: Yield of pods and seeds of *Sesamum indicum* in different treatments at Maroua.

Parameters	Treatments (1, 1′)	Treatements (2, 2′)	Comparison (1 and 2 ; 1′ and 2′)
***Fr*2015**	80.00 %	42.00%	χ^2_{2015} = 2821.60 [df = 1 ; P < 0.001]*
***Gn*2015**	91.67 %	79.03%	χ^2_{2015} = 6.75 [df = 1 ; P < 0.05]*
***Ng*2015**	84.00 (n = 40 ; s = 3.26)	62.00 (n =40 ; s = 6.48)	Z_{2015} = 19.18 [df = 78 ; P < 0.001]*
***Mg*2015**	1.02 (n = 40 ; s = 0.021)	0.42 (n = 40 ; s = 0.02)	Z_{2015} = 33.85 [df = 78 ; P < 0.001]*
***Tf*2018**	53.97 %	24.73%	χ^2_{2018} = 203.80 [df = 1 ; P < 0.001]*

***Gn*2018**	98.13 %	92.93%	χ^2_{2018} = 2.98 [*df* = 1 ; *P* < 0.05]*
***Ng*2018**	77.90 (*n* = 40 ; *s* = 10.06)	71.48 (*n* =40; *s* =9.67)	Z_{2018} = 5.07 [*df* = 78 ; *P* < 0.05]*
***Mg*2018**	1.09 (*n* = 40; *s* = 0.011)	0.45 (*n* = 40; *s* = 0.06)	Z_{2018} = 39.05 [*df* = 78 ; *P* < 0.001]*

Fr: Fruiting rate, Ng: Number of seeds/pod, Gn: Percentage of normal seed, Mg: mass of seeds *: Significant at *P* <0.05.

2. High mean number of seeds per pod in unlimited visits compared with bagged flowers. The comparison of the mean number of seeds per pod has shown that the difference observed was highly significant between treatments 1 and 2 (Z_{2015} = 19.18 [*df* =78 ; *P* < 0,001]) and treatments 1' and 2' (Z_{2018} = 5.07 [*df* = 78 ; *P* < 0.05]). The percentage of the number seeds per pod due to the action of insects was 17.22%. In the same idea, Tchuenguem and Népidé (2018) found the corresponding result at Ngaoundéré of 29.65%.

3. The comparison of the average weight of seeds showed that the difference were high significant between treatment 1 and treatment 2 (Z_{2015} = 33.85 [*df* =78 ; *P* < 0.001]) and treatments 1' treatment 2' (Z_{2015} = 39.05 [*df* =78 ; *P* < 0.001]). The results suggest that the type of pollination affect weight. The weight of seed yield in free pollination was high than that in the pollination restricted with the gauze bag. This result agrees of that found by Andrade *et al.* (2014) in Brazil. The numeric contribution of insect pollinators on the average weight of seeds was 58.77%.

4. Higher normal seed yield for unlimited visits treatment compared with bagged flowers. The comparison of the percentage of normal seeds showed that the observed differences were highly significant between treatments 1 and 2 (χ^2_{2015} = 6.75 [*df* = 1 ; *P* < 0.05]) and treatments 1' and 2' (χ^2_{2018} = 2.98 [*df* = 1 ; *P* < 0.05]). The Normal seed yield in unprotected flowers for unlimited visits (1 and 1') was higher than that in the bagged flowers (2 and 2'). The percentage of normal seeds attributed to the influence of insects was 09.55%. This result is superior to what was obtained by Pharaon *et al.* (2018) at Obala, which was 05.02 %. The difference in this value could be explained by the presence of more pollinating species at Maroua.

Conclusion

In Maroua, *Sesamum indicum* is a mixed pollination plant, able to self-pollinate and reach levels above 53 % fruit set, but with potential for a significant increase in fruit production in the presence of biotic pollinators that promote cross-pollination. Nineteen species of insects distributed in nine families and five orders visited the flowers of *S. indicum* to harvest nectar and/or pollen. Hymenoptera were the most frequent order with 90.43%, followed by Lepidoptera (06.40 %) and others (Diptera, Orthoptera, and Coleoptera: 03.17 %). These insects foraged the flowers of this Pedaliaceae from 7:00 am to 5:00 pm, with a peak activity between 9:00 am, and 10:00 am, where 28.59% of visits are observed at that time. Comparing the yield of unlimited flowers with insect-bagged flowers, it is observed that insect visits increase the fruiting rate, the number of seeds/fruit, the average weight of seeds, and the normal seeds to 50.84 %, 17.22 %, 58.77 %, and 09.55 % respectively. The treatment of sesame plants with chemical pesticides should be avoided during the flowering period in order to benefit from the ecosystem service of pollinating insects. Pollinators could be protected by rational pest management tactics, i.e., pesticide application, if needed, should be done in the late afternoon to protect the pollinators for high seed yield or spray at a time of day when crop flowers are closed.

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Disclosure of Conflict of Interest

Authors have declared that no competing interests exist.

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