

Research Article

Knowledge of Floral Biology, the Variation of Climatic Factors and the Role of Pollinators Significantly Improve the Yield of *Cucurbita maxima* Cultivation in Cameroon

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Abstract

Cucurbita maxima (Cucurbitaceae) is an edible, medicinal and vitamin-rich species. In Cameroon, although this plant is among the edible crops, knowledge about the relationship between this crop and pollinators is scarce. This study aims to describe the floral biology of *C. maxima* and the relationship between this crop and *A. mellifera* for their optimal exploitation. Observations were carried out at two sites (Torok and Vogzom) during the 2022 agricultural season. The growth stages of flowers buds and their daily appearance rate were recorded. Four treatments were defined at each site to assess the effectiveness of bee visits on pollination and fruit yield. The number of male flowers exceeded that of female flowers, with a sex ratio of 5:1 (♂/♀). The impact of climate factors on bee visits, fruit mass and diameter, and the number of mature fruits from the different treatments was evaluated. High temperatures decreased bee visits, as did rainy days. Flower visits for nectar collection were more frequent than those for pollen. Bees effectively promoted the pollination of female flowers during their foraging trips. There is a correlation between the number of bee visits received by female flowers and the fruit set rate per treatment. The production of good-quality fruit and seeds from this plant depends on optimal bee pollination activity. It is to Maintain climatic factors at a level favorable to pollinator activity by combating climate change.

Keywords

Floral Biology, *Cucurbita maxima*, *Apis mellifera*, Pollination, Pollinator Efficiency, Resilient Farming

1. Introduction

Agriculture is a sector that largely ensures food self-sufficiency and employs over 60% of the Cameroonian workforce. Cameroonian agricultural production accounts for more than half (52.3%) of the country's total agricultural output and

makes it the largest supplier of agricultural products to countries in Central Africa [1]. Despite this significant performance in agricultural production, the sector's contribution to Cameroon's GDP has gradually declined, falling from 44% in

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2004 to 17% in 2021 [2]. This decline in the agricultural sector's contribution to GDP has directly resulted in a deterioration of the population's living standards. With high rates of food insecurity in the Sudan-Sahelian zone (24.8%) in the Far North and (16.8%) in the North, while the national poverty rate stands at 37.7% [3, 4], and given the political will to preserve food self-sufficiency, one solution to these challenges would be the diversification of agricultural production, particularly the development of local edible resources [3, 5].

Cucurbita maxima, or pumpkin, is a monoecious food plant that is part of the indigenous crops of Cameroon and is cultivated in association with other crops [6-8]. It is widely cultivated as a vegetable for its fruits, seeds, and leaves, all of which are edible and rich in beta-carotene, vitamin C, and potassium, useful in the prevention of kidney stones [9]. The fruits, seeds, and leaves exhibit various pharmacological effects, such as antidiabetic, antiulcer, analgesic, nephroprotective, and anticancer effects [10]. Global squash production increased from 15,186,469.4 tonnes in 2020 to 18,306,860.06 tonnes in 2023. China is the world's leading producer, with 7,374,060.7 tonnes, representing 45% of total production. Algeria is the leading producer in Africa and 11th globally, with 455,140.94 tonnes [11]. The fruit yield of *C. maxima* varies from 20 to 30 tonnes per hectare (t/ha) in organic open-field cultivation [12]. Protecting pollinators is one of the major challenges that can optimize agricultural production, thereby reducing the impact of climate change on food security [13].

Pollination is a key factor in the sexual reproduction of many plant species. The symbiosis between insects and plants allows for the maintenance of biodiversity through cross-pollination and the sustainability of certain ecosystems [14]. Overall, the financial value of the ecological and economic services provided by bees annually is increasing; it has risen from \$153 billion, or approximately 76.5 trillion FCFA [15] to \$577 billion in 2018 [16]. Knowledge of the pollinating insects of different cultivated plants and ensuring the protection of these insect populations is therefore more than necessary to preserve our food security [13]. With regard to Cucurbitaceae, several scientific studies have confirmed their dependence on entomophilous pollinators, as their pollen grains are sticky and cannot be transported by the wind [17-23].

In Cameroon, studies have been conducted on *C. maxima*, particularly on its classification and botanical description [6, 24], as well as on the nutritional potential of its seeds [25, 26]. The distribution and cultivation practices of *C. maxima* and *Cucurbita moschata* has been studied in the Central, Eastern, and Western regions [8]. However, research on *Cucurbita maxima* is scarce, or even nonexistent, in some regions of Cameroon. This could explain the lack of reliable statistical data regarding the cultivated area and annual production of this food in the country. Given the importance of the edibility of *Cucurbita maxima* products for the population, its dependence on pollinators, and the need to diversify agricultural resources to ensure food security, further studies on *Cucurbita*

maxima are necessary to complement existing data on *Cucurbitaceae* pollinators. According to the work of Tchuenguem, [27] the flower insect fauna and its impact on pollination and crop yield can vary spatially and temporally.

The main objective of this work was to contribute to the understanding of the floral biology of *C. maxima* and the relationship between this crop and *A. mellifera* for their optimal exploitation. Specifically, it aimed to analyze the floral biology of *C. maxima*, study the activity of flower-visiting insects on this plant, and evaluate the impact of *A. mellifera* on crop yield.

2. Material and Methods

2.1. Location of Study Sites

Field observations were conducted in 2022 at Torok, located between latitude (10°0'0"N) and longitude (15°0'0"E) at an altitude of 195 m, and Vogzom, located between latitude (8°0'0"N) and longitude (15°0'0"E) at an altitude of 205 m, each covering an area of approximately 400 m² in Cameroon (Figure 1). These sites were chosen due to the high number of *Cucurbita maxima* producers, the presence of farmers' fields cultivating other crops, and the guarantee of safety for both the experimental plots and the observer.

2.2. Study of the Floral Biology of *C. maxima* in a Culture System

2.2.1. Study of the Growth Stages of *C. maxima* Flower Buds

To assess the various morphological changes and identify the different developmental stages from bud to open flower, 100 male and 60 female flower buds were randomly selected. Upon opening, the peduncle length and diameter of both male and female flowers were measured. A flower is considered open when it is fully open and developed, having reached its full potential for growth and beauty [28].

2.2.2. Study of the Flowering Rhythm of Male and Female Flowers in *Cucurbita Maxima*

From the beginning to the end of the flowering period of this plant, the number of open male and female flowers and their arrangement on the stem in each experimental plot were recorded each morning. The observations were made row by row and planting hole by planting hole, and the number of open or fully opened male and female flowers per plant per day was recorded throughout the entire flowering period [28]. This allowed researchers to assess the daily flowering rate of flowers of different sexes and deduce the sex ratio [29].

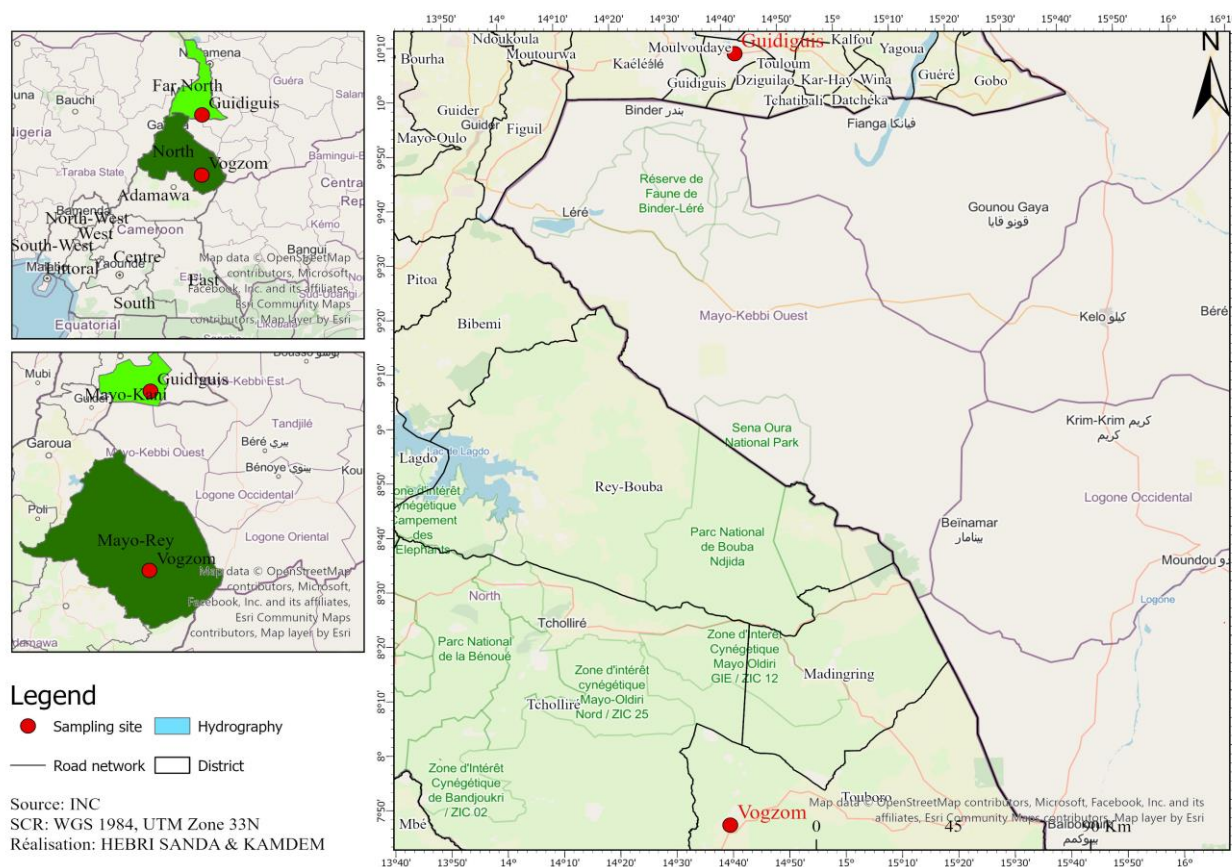


Figure 1. Location maps of study sites.

2.3. Study of Insect Activity on Flowers *Cucurbita maxima*

2.3.1. Inventory of Insect Diversity on *Cucurbita maxima* Flowers

To collect data on bee activity on *Cucurbita maxima* flowers, a randomized experimental design was used at both study sites. Twenty-four plants were randomly selected and divided into two blocks. Female flowers from these two blocks were labeled, and two treatments were defined [27]. Treatment A (T_1) (consisting of 12 plants, 30 of which female flowers were labeled and left in free pollination, i.e. accessible to floral visits by bees) and Treatment B (T_0) consisting of 30 female flowers isolated from bee visits. Observations were made daily on the flowers of *C. maxima* plants left to undergo free pollination (T_1) or block (A) between 6:00 a.m. and 1:00 p.m. (periods corresponding to the beginning and end of insect visits) during its flowering period [29]. Anthophilous insect activity was recorded during four daily time slots: 6–7:00 a.m., 8–9:00 a.m., 10–11:00 a.m., and 12–1:00 p.m. During each observation period, a single pass was made in front of each labeled plant in block A, and the number of bees on open flowers, the floral products collected, and the duration of their visits were recorded. The cumulative results of the various re-

cordings throughout the flowering period allowed for the calculation of the relative abundance and average duration of *Apis mellifera* visits per site [30]. The Shannon diversity index of two sites was deduced in order to assess species richness [31].

2.3.2. Description of the Influence of Varying Climatic Factors on Insects' Activity

Climatic factors strongly influence insect foraging activity. Temperature and humidity at the study site were recorded twice per daily observation period using a portable thermohygrometer. Average values for each observation period were calculated, and linear relationships were established with daily variations in flower-visiting insect activity. The effects of wind, sunshine, rain, and cloud cover were also noted [32].

2.4. Impact of *A. mellifera* on *Cucurbita maxima* Crop Yield

2.4.1. Pollination Efficiency

A treatment (C) was defined and subdivided into four sub-treatments. This study aimed to investigate the pollination efficiency of *C. maxima* female flowers by *Apis mellifera*. Treatment C (T_2) consisted of 10 female flowers, each receiving a single visit from *A. mellifera*; treatment C (T_3) consisted

of 10 female flowers, each receiving two simultaneous visits from *A. mellifera*; and treatment C (T_4) consisted of 10 female flowers, each receiving three simultaneous visits from *A. mellifera*. The marking, isolation, and labeling of the female flowers were carried out the day before they opened [22]. Early on the day of their opening, the mosquito netting was removed from the female flowers to allow them to be visited by *A. mellifera*. They were visited either simultaneously or consecutively by the same individual bee or by other bees. Once the

predetermined number of visits for the flower was reached, it was protected again [33]. The following day, the netting was removed to monitor the development of any potential fruit until its eventual maturity. The reappearing petals allowed for precise determination of the imminent opening of the female flowers in question [34]. The experimental setup used in this study was the same at both sites (Figure 2).

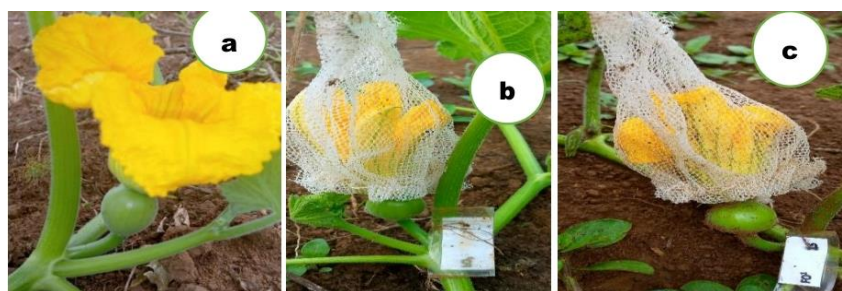


Figure 2. Free female flowers (a); isolated (b) and one that has received a visit from a bee (c).

2.4.2. Influence of Apis Mellifera Visits on the Growth Parameters and Fruit Yield of *C. maxima*

The growth parameters (diameters and masses) of the fruits under each treatment were recorded using a measuring tape and a precision electronic balance to determine the different growth phases in volume and weight. These parameters were recorded for both immature and mature fruits. The diameter (cm) and mass (kg) of immature fruits were recorded immediately upon their abortion or detachment from the mother plant [29].

Calculation of the pollination rate attributable to pollinating insects

To calculate the pollination rate attributable to pollinating insects, mature *C. maxima* fruits were weighed using kitchen scales and cut open with a machete. The total number of seeds per fruit, including both mature (SM) and immature seeds (SI), was recorded. The type of seeds allowed for the estimation of the pollination rate (P): $P (\%) = \{[(SM / ST + SI)] / 100\}$ according to Gingrass [35].

Determination of fruit set and seed maturity attributable to pollinating insects

In the 5 treatments, the fruiting rate is determined by $[(\text{number of fruits formed} / \text{number of female flowers studied}) \times 100]$. The fruiting rate (% Rfr) attributable to anthophile insects is estimated by the formula $(\%Rfr) = \{[(T_A - T_B) / T_A] \times 100\}$ where T_A and T_B are respectively the fruiting rates in treatment A (female flowers without protection) and in treatment B (female flowers protected from the floral activity of insects) [32]. Just as with the calculation of the fruiting rate due to pollinating insects, the same reasoning is applied to the calculation of

the percentage of mature seeds per fruit due to the floral activity of anthophilous insects ($\%PMS$) = $\{[(T_A - T_B) / T_A] \times 100\}$ where T_A and T_B are respectively the fruiting rates in treatments A and B [36].

Calculation of the rate of ripe fruit and abortion

All mature fruits (Fm) and all female flowers (Ff) are counted, allowing the calculation of the mature fruit percentage (%Mfr) using the following formula: $(\%Mfr) = \{(Fm / Ff) \times 100\}$. The abortion rate (%R) in each treatment was deduced using the formula: $(\%R) = 100 - (\%M)$ [37].

3. Results

3.1. Study of Floral Biology

3.1.1. Growth Stages of *C. maxima* Flower Buds

Flowering begins 30 days after seed emergence; male flower buds (σ) appear first (Figure 3M), followed by female buds (ρ) a week later (Figure 3F). It has been noted that the flower buds that originate on the main stem and primary branches are male. Subsequent branches bear a or two female flower buds and several males. Monitoring bud growth Floral observation up to full bloom performed on 160 buds Male and female floral elements allowed for identification five identical developmental stages in the male flower and female. The first stage: closed button (CB) is characterized by a small-diameter flower bud that is completely closed and green in color. Then the flower bud grows in volume as the sepals lengthen and open (second stage: sepals open, SO). The transition from stage CB to stage SO takes on average three days. This is fol-

lowed by a wider opening of the sepals, which reveals the petals which are of lemon-yellow color (third stage: small pimple yellow, SPY). The next stage is characterized by petals still closed but more elongated and colored yellow (fourth stage: large yellow bump, LYB) which is the stage preceding the blooming of the flower (fifth stage: fully opened flower, FOF). From the stage SO, the transition through each stage lasts one day, so that Six days after the appearance of the flower bud, the flower is in full bloom. The flower remains open for half a day. Then the petals wither and fall the next day. The length of the flower peduncle and the diameter of the open flower are of 10.14 ± 2.72 mm and 16.54 ± 2.26 mm for the flower's males and 8.06 ± 3.21 mm and 20.73 ± 3.43 mm for the female flowers.



Figure 3. Appearance of the first male (M) and female (F) flower buds.

3.1.2. Flowering Rhythm of Male and Female Flowers in *Cucurbita maxima*

Throughout the flowering period, it was observed that for each plant, the number of male flowers was significantly higher than the number of female flowers every day (Figure 4). Flower opening began very early (5:00 a.m.) and all flowers opened at sunrise, i.e., 6:00 a.m. The flowers gradually closed from noon until 1:00 p.m., depending on the amount of sunlight. The daily peak of male flowers was 83, while it was 32 for female flowers at both sites. The total number of male flowers obtained during the flowering of 72 pumpkin plants in this study was 1274, and the number of female flowers was 274 at Vogzom. At Torok, the figures were 1250 male flowers and 245 female flowers. The number of male and female flowers was relatively similar between the two sites. The average number of male flowers per plant was 17 and 18 at Torok and Vogzom, respectively. In contrast, the average number of female flowers per plant was 3 and 4 at Torok and Vogzom, respectively. Based on these figures, the estimated sex ratio of *C. maxima* from our experimental plots at both sites is identical, i.e., (δ/ϕ) 5:1.

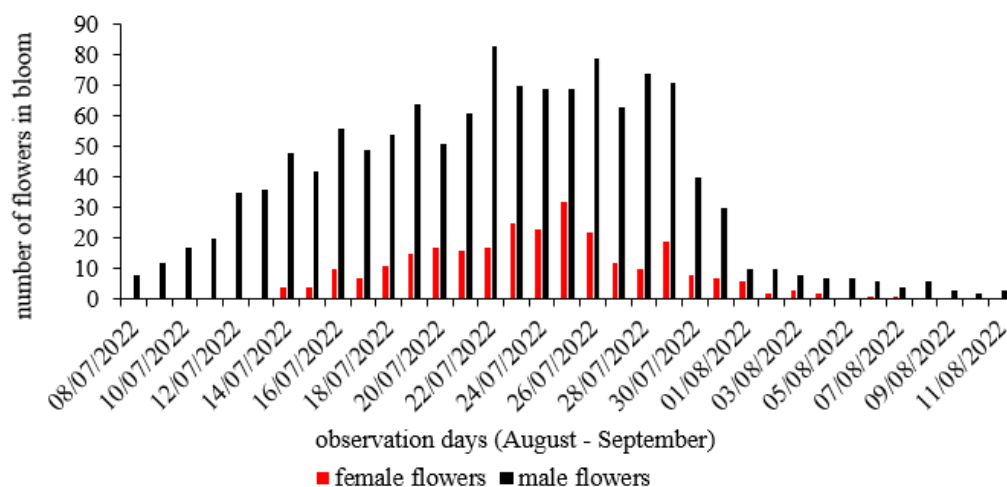


Figure 4. Rhythm of appearance of male and female flowers in *Cucurbita maxima*.

Overall, the greater the number of male flowers, the higher the probability of female flowers appearing. This variation between the two flowers led to the observation of a significant positive linear regression between male and female flowers ($y = 2.6172x + 15.711$) with a coefficient of determination $R^2 =$

0.7121 ($df = 1.2$; $p < 0.05$). This coefficient of determination indicates that 71% of the variation in the number of male flowers influences the appearance of female flowers, as illustrated by Figure 5.

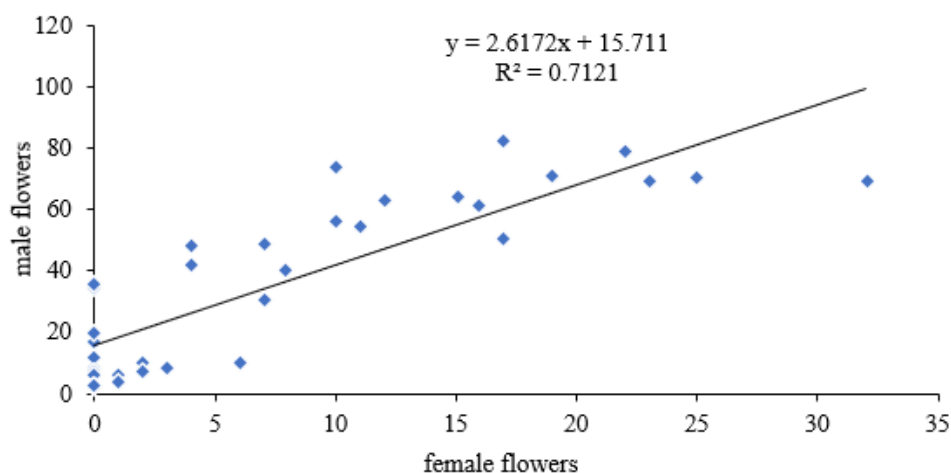


Figure 5. Linear regression between female and male flowers of *C. maxima*.

3.2. Insect Activity on *Cucurbita maxima* Flowers

3.2.1. Biological Diversity of Insects on *C. maxima* Flowers

At the order level, the flower-visiting insects of pumpkins in Vogzom are divided into three main orders of unequal importance according to their relative abundance. The order Hymenoptera is predominant with 85.58%, followed by Coleoptera and Diptera with relative abundances of 9.13% and 5.29%, respectively. The flower-visiting insects of pumpkins were grouped into four families, with Apidae being the most numerous (82.06%). Five species of anthophilous insects were recorded as active on the flowers of *C. maxima*, the honeybee *A. mellifera* being the most prominent with a relative abundance of visits of 82.06%; this bee is the only representative of the Apidae family. The other species, in descending order of relative abundance, are: *Drosophila melanogaster* (5.29%), *Monolepta intermedia* (4.93%), *Monolepta bioculata* (4.20%) and *Lasioglossum* sp. (3.52%). Overall, the flower-dwelling insect fauna of *Cucurbita maxima* at Vogzom is poorly diversified and largely dominated by *A. mellifera*.

In contrast, at Torok, the flower-visiting insect fauna of *Cucurbita maxima* is divided into three orders of unequal relative

abundance. The order Hymenoptera is dominant at 82.3%, followed by Lepidoptera and Diptera at 11.1% and 3.4% relative abundance, respectively. Regarding family, flower-visiting insects are distributed among nine families, dominated by Apidae at 77.55%. Fifteen species of anthophilous insects have been recorded as active on *C. maxima* flowers, with *A. mellifera* being dominant at 40.5% relative abundance, while *Trichostoma sjostedti* is the weakest (0.9%). Overall, the flower-visiting insect fauna of *Cucurbita maxima* at Torok is not very diverse and is dominated by *A. mellifera*. The two sites (Torok and Vogzom) are independent in terms of the number of species ($t = 34$; $df = 17$; $p\text{-value} = 0.861$) at the 5% significance level. The results of the biological diversity of the flower-visiting insect fauna associated with *C. maxima* Vogzom and Torok (Table 1).

From these relative abundances of flower-visiting insects, we can deduce the Shannon diversity index, $H' = 2.70$ (Torok) and $H' = 1.03$ (Vogzom). The difference between the Shannon diversity indices is significant (at the 5% level) between the two sites, indicating that the use of insecticides in farming practices at Vogzom has an impact on insect biodiversity. The overuse of synthetic chemicals is one of the major causes of the decline in insect biodiversity, despite the crucial role insects play in maintaining and functioning the ecosystem.

Table 1. Relative abundances of flower-visiting insects of *C. maxima* at Torok and Vogzom.

Flower-growing insects			Far North		North	
			Torok		Vogzom	
Order	Family	Genus and Species	n_1	$f_1(\%)$	n_2	$f_2(\%)$
Hymenoptera	Apidae	<i>Apis mellifera</i>	528	40.5	1349	82.06

Flower-growing insects			Far North		North	
			Torok		Vogzom	
Order	Family	Genus and Species	<i>n</i> ₁	<i>f</i> ₁ (%)	<i>n</i> ₂	<i>f</i> ₂ (%)
Total Hymenoptera		<i>Xylocopa inconstant</i>	261	19.7	-	-
		<i>Xylocopa pubescens</i>	195	14.7	-	-
		<i>Xylocopa olive tree</i>	42	3.2	-	-
		Total Apidae	4	1026	77.55	1349
	Halictidae	<i>Liporichs collaris</i>	42	3.2	--	
		<i>Lasioglossum sp.</i>	-	-	58	3.52
		<i>Trinchostoma sjostedti</i>	12	0.9	-	-
	Total Halictidae	3	54	4.1	-	-
	Megachilidae	<i>Megachile aurifera</i>	51	3.9	-	-
	Total Megachilidae	1	51	3.9	58	3.52
	4	8	1089	82.3	1407	85.58
Coleoptera	Chrysomelidae	<i>Monolepta intermedia</i>	-	-	81	4.93
		<i>Monolepta bioculata</i>	-	-	69	4.2
	Total Chrysomelidae	2	-	-	150	9.13
Total Coleoptera	1	2	-	-	150	9.13
Diptera	Syrphidae	<i>Phytomia sp.</i>	15	1.1	-	-
		1 sp.	12	0.9	-	-
	Total Syrphidae	2	27	2.00	-	-
	Drosophilidae	<i>Drosophila melanogaster</i>	18	1.4	87	5.29
	Total Drosophilidae	1	18	1.4	87	5.29
	Total Diptera	2	3	45	3.40	87
Lepidoptera	Nymphalidea	<i>Hypolimnas misippus</i>	48	3.6	-	-
	Total Nymphalidea	1	48	3.6	-	-
	Papilionidea	<i>Papilio demodecus</i>	30	2.3	-	-
	Total Papilionidea	1	30	2.3	-	-
	Pieridae	<i>Eurema exima</i>	21	1.6	-	-
		<i>Catopsilia florella</i>	15	1.1	-	-
	Total Pieridae	2	36	2.7	-	-
	Acraeidae	<i>Acraea acerata</i>	33	2.5	-	-
	Total Acraeidae	1	33	2.5	-	-
	Total Lepidoptera	4	5	147	11.1	-
Total	10	18	1323	100.0	1644	100.0

3.2.2. Influence of Climatological Parameters on Bee Activity

Overall, insect abundance on *C. maxima* flowers is correlated with the number of flowers open per day. This observation shows that visits become more frequent as the number of open flowers increases (Figure 6). There is a positive and significant correlation between the number of open flowers and

visitor abundance at both Torok ($r = 0.93$; $df = 14$; $P < 0.001$) and Vogzom ($r = 0.92$; $df = 4$; $P > 0.01$). This correlation highlights the high attractiveness of the floral products of this cucurbit to pollinators. However, the abundance of flower visits by flower-visiting insects is influenced by temperature and humidity depending on the observation days. Floral visits are most frequent in the morning, when the temperature is low (18 °C) and the relative humidity is high (80.36%). Visits by flower-visiting insects gradually decrease as the temperature rises and the relative humidity falls. Overall, insect visits to flowers are strongly correlated with variations in air temperature and relative humidity.

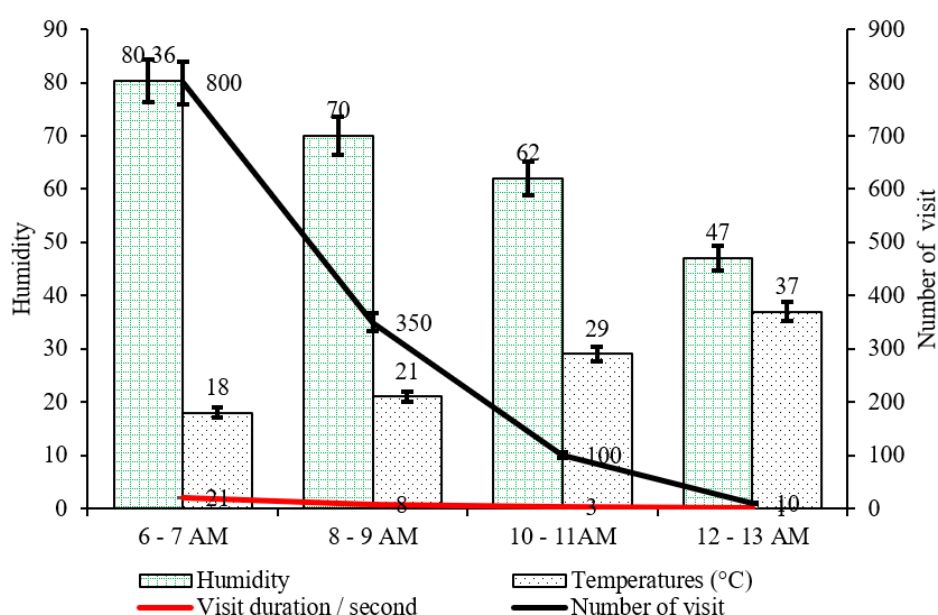


Figure 6. Insect visits as a function of open flowers (a) and abiotic factors (b).

3.3. Impact of *A. mellifera* on the Yield of *Cucurbita maxima*

3.3.1. Impact of *Apis mellifera* on the Pollination of *Cucurbita maxima*

During their visits to *Cucurbita maxima* flowers, bees actively collected nectar and pollen at both study sites. After careful observation of *Apis mellifera* behavior during pollen and/or nectar collection on the flowers of this plant, it was found that these bees frequently came into contact with the anthers (male flowers) and the stigma (female flowers). The highest number of individuals simultaneously active on a single open flower averaged 3 (3.26 ± 1.45) for *A. mellifera* at both sites (Figure 7).



Figure 7. *Cucurbita maxima* flower being foraged by several worker bees.

3.3.2. Influence of Bee Visits on the Growth Parameters and Fruit Yields of *C. maxima*

The average fruit diameter ranged from 16.57 cm in treatment T₁ to 0.52 cm in treatment T₃ and 6.26 cm in treatment T₄. The average fruit mass was 4.26 kg in treatment T₁, 0.11 kg in treatment T₃, and 0.87 kg in treatment T₄. The abortion rate was 90% in T₃, 80% in T₄, and 100% in treatments T₀ and T₂. The growth parameters of the formed fruits increased with the number of bee visits to the female flowers; the more bee visits to the female flowers, the better the characteristics of the formed fruits, and vice versa (Figure 8).

The fruit set rate was 93.33% in treatment A, 0% in treatment B, and 43.33% in treatment C. The overall comparison indicated a significant difference between treatments A, B, and C ($\chi^2 = 52.77$; $df = 2$; $P < 0.05$) and between treatments A and C ($\chi^2 = 17.33$; $df = 1$; $P < 0.05$). The average number of mature seeds per fruit was 255.31 ± 51.02 in treatment A, 15 in treatment C, and zero in treatment B, as no fruit formed.

The pollination rate was deduced from the ratio of the total number of mature seeds (TNMS) obtained to the total number of seeds (TNS), which reflects the total number of ovules obtained. This rate was $[(6521/6638) \times 100] = 98.23\%$ in treatment (A) and 35.71% in treatment (C). This high rate in treatment (A) indicates that good pollination leads to good fertilization and increased yields of high-quality seeds and fruit in *C. maxima*. Although the flowers in treatment (A) received significant bee visits, it was observed that some female flowers failed to develop fruit, with a rate of approximately 7.14% (Table 2). The statistical test reveals the existence of a highly significant difference between the rate of fruiting of the treatments (T₀, T₁, T₃ and T₄) with χ^2 global = 189.45; $df = 4$; $P < 0.001$ at a significance level of 5%.

Overall, the larger the fruit, the higher the number of mature seeds it generally contains. There is a significant positive regression equation between fruit mass and diameter ($y = 102.57x + 36.418$) with a coefficient of determination $R^2 = 0.89$ ($df = 1.19$; $p < 0.05$). This coefficient of determination indicates that 89% of the variation in the number of mature seeds influences the resulting fruit mass (Figure 9).

Table 2. Evaluation of *C. maxima* yield according to treatments at Torok and Vogzom.

Parameters studied	Treatment A		Treatment B		Treatment C	
	Torok	Vogzom	Torok	Vogzom	Torok	Vogzom
Number of female flowers studied	30	30	30	30	30	30
Number of fruits formed	23	28	0	0	8	13
Fruits formed (%)	76.67	93.33	0	0	26.66	43.33
Number of ripe fruits obtained	12	26	0	0	0	2 ***
Ripe fruit (%)	52.17	92.85	0	0	0	15.38
Total number of seeds obtained	3028	6638	0	0	0	84
Number of mature seeds	2925	6521	0	0	0	30
Mature seeds obtained (%)	96.60	98.23	0	0	0	35.71
Number of immature seeds	103	117	0	0	0	54
Immature seeds obtained (%)	3.40	1.76	0	0	0	64.28
Average number of mature seeds /fruits	252 ± 38	255 ± 51	0	0	0	15
Abortion rate (%)	48.00	7.15	100.00	100.00	100.00	85.00

Legend: ***: flowers having received three visits (T₄).

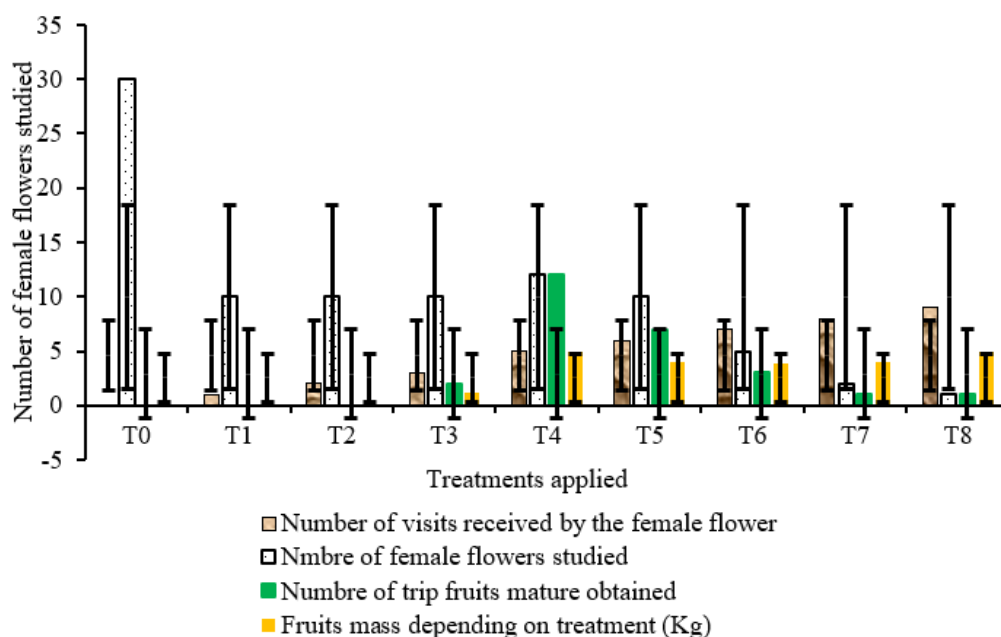


Figure 8. Influence of the number of bee visits on the formation and mass of fruit formed by treatment.

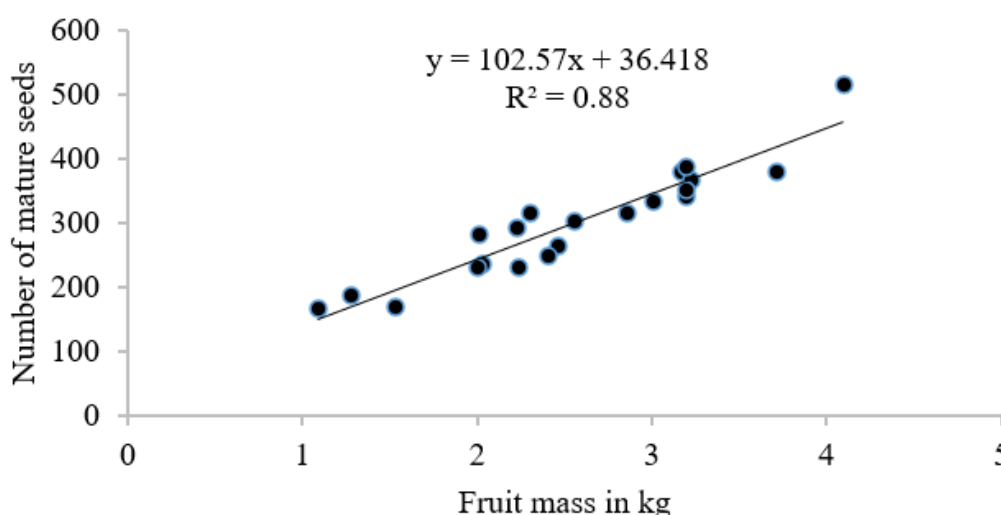


Figure 9. Linear relationship between fruit mass and number of mature seeds.

4. Discussion

The results of the experiments reveal that there are five identical growth stages of male and female flower buds until they open. The first flowers to open are male, and the female flowers appear seven days later. The flowers of *C. maxima* have a short lifespan; they open very early and close at 1 p.m. Similar results were observed in another species of the Cucurbitaceae family, *Cucumis melo* L. var. *agrestis*, in Ivory Coast and *Cucurbita. moschata* in Dang (Ngaoundere) [28, 29]. The sex ratio of *Cucurbita maxima* based on observations from this study was ($\delta/\varphi = 5:1$), which is contrary to that obtained by Dje [28] on *Cucumis melo* L. var. *agrestis*, which was

($\delta/\varphi = 15:3$), while Nepi & Paccini [38] obtained a sex ratio of ($\delta/\varphi = 9:7$) on *Cucurbita* these species all belong to the Cucurbitaceae family. The difference in sex ratio between these species could be due to ecological and environmental factors, such as soil fertility, rainfall, and the local varieties of species cultivated by farmers. The significant variation in the number of female and male flowers from one plant to another is explained by the action of enzymes (gibberellin, abscisic acid, ethylene, and auxins) which, according to Odet's findings, regulate the proportions of flowers of different sexes in Cucurbitaceae [39].

Cucurbita maxima cultivation in Vogzom comprises 3 orders, 4 families, and 5 species, while in Torok it consists of 3 orders, 9 families, and 15 species. The insect fauna associated

with this crop is sparse in Vogzom and less diverse in Torok. Studies by other authors on other cultivated and wild *Cucurbitaceae* species mention a relatively high number of flower-visiting insect species. Azo'o, recorded 38 insect species on *Citrullus lanatus*, 35 on *Cucumeropsis mannii* and 14 on *Abelmoschus esculentus* flowers in Yaounde [36]. Although overall the flower insect fauna varies from one plant species to another, the overuse of chemicals by farmers would also be a cause of the decrease in insect biodiversity in the context of this study.

Among the insect species identified at the two sites, *A. mellifera* was the most abundant, with a relative abundance of 82.06% and 40.5% at Vogzom and Torok, respectively. Honeybees are known to be the main flower-visiting insects for other *Cucurbitaceae* species, as documented by several authors [23, 35, 37, 40-44].

The activity of *Apis mellifera*, particularly on the flowers of *C. maxima*, is predominant at 6–7 a.m. and ends around midday (12–1 p.m.), marked by the closing of the flowers of this plant. It is known in the literature that pumpkin flowers, like other *Cucurbitaceae* species, have an ephemeral lifespan [45, 46] of which they produce a lot of nectar in the morning from anthesis onwards. Thus, the peak of bee activity on the flowers of plant species is generally correlated with the greater availability of floral resources such as nectar and pollen [46, 47]. The results of this study also show a significant positive correlation between the daily flowering rate and the number of flower visits by insects in general, and Apidae in particular. These results corroborate those of several other authors [31, 47, 48]. The rate of bee visits gradually decreases as the temperature rises, and the same is true during rainy days. To benefit from the ecosystem services provided by bees, it is essential to combat climate change in order to maintain temperatures at a level favorable to their activities. Climate change significantly affects pollinator activity and consequently the production of many crops, thus causing famine and poverty according to Potts [49].

During their foraging visits to the staminate flowers of *C. maxima*, *A. mellifera* would contact the anthers and passively collect pollen using their thick fur. Passing over the female flowers, the foragers would come into contact with the stigmas and induce pollination of the visited female flowers. In entomophilous plants, including *C. maxima* [50, 51], pollination requires pollen transfer via the bees' integument. Pollen retained by the bees' hairs is the most suitable for pollination [52]. The peak activity of honeybees at the moment flowers open (6-7 a.m.) coincides with the period of stamen maturity and stigma receptivity in *Cucurbitaceae* [53]. Indeed, in *Cucurbitaceae*, when the flowers are open, the pollen is dehiscent and the stigma receptive for at least two hours [53].

The optimal yield of mature fruits and grains was obtained in treatment T₁ (free flowers), while it was zero in treatment T₀ (female flowers isolated from bee activity) at both sites. Optimal yield is determined by the number of bee visits re-

ceived by the female flowers of *Cucurbita maxima*. These results are consistent with those of Phillippe and Pablo [54, 55]: the more pollen grains a female flower receives, the greater its potential to develop into a large fruit containing numerous mature seeds. These correlations explain why effective entomophilous pollination leads to the production of good-quality fruit. Conversely, poor pollination of female flowers results in reduced yield and poor-quality fruit. This explains why, in treatments T₂, T₃ and T₄, at both Vogzom and Torok, the abortion rate is high and the ripening fruits are of poor quality. The production of this *Cucurbitaceae* depends on the optimal pollination activity of *A. mellifera*, according to the results of several authors' work [17, 19-21]. The average mass of fruit from female flowers left for open pollination (T₁) in this study was 4.26 kg, which is lower than that of Isuta & Mallowa [56] who obtained up to 7.5 kg. Cordova, mentions that several factors can influence agricultural yield, including: variety, climate, cropping system, soil characteristics, and pollinator availability [18].

5. Conclusion

The first flower buds are male, and there are five identical developmental stages in the male flower. as in the female flower from bud to open flower. The estimated value of the sex ratio of *C. maxima* from experimentation is (σ/ϕ) 5:1. The flower-visiting insect fauna associated with *C. maxima* cultivation in Vogzom, as in Torok, is generally not very diverse. Pollination of the female flowers is primarily ensured by *Apis mellifera*, which played a significant role in the fertilization of this crop. During their foraging, the bees moved from flower to flower, transferring pollen grains from the male flowers to the stigmas of the female flowers, thus fertilizing the ovaries. Experiments showed that the more frequently a female flower receives bee visits, the more pollen it receives, and the greater the fruit and seed production. Therefore, the minimum number of bee visits to the female pumpkin flowers that guarantees crop yield is, on average, three visits in this study. All 30 female flowers protected from insect visits (T₀) systematically aborted. Overall, honeybee activity determines the fruit and seed production of *Cucurbita maxima*. The average weight of T₁ fruits from experimental fields at two sites, each with an area of 400 m², was 4.26 ± 1.12 kg. One of the important factors for optimizing the production of high-quality fruit and seeds of *C. maxima* is combating the decline of pollinating insects by avoiding the use of chemical pesticides in cultivation practices and promoting beekeeping.

Abbreviations

GDP	Gross Domestic Product
Rfr	Fruiting Rate
MS	Mature Seeds
Fm	Mature Fruits

Ff	Female Flowers
AR	Abortion Rate
Mfr	Mature Fruit
SI	Immature Seeds
CB	Closed Button
SO	Sepals Open
SPY	Small Pimple Yellow
LYB	Large Yellow Bump
FOF	Fully Opened Flower
TNS	Total Number of Seeds
TNMS	Total Number of Mature Seeds

Conflicts of Interest

The authors declare no conflicts of interest.

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