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### Comparative study of the foraging activity of *Apis mellifera* L. (Hymenoptera: Apidae) on the capitula of *Helianthus annuus* in two agroecological zones of Cameroon

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#### Abstract

The impact of *Apis mellifera* L. (Hymenoptera: Apidae) foraging on *Helianthus annuus* yields in two agroecological zones of Cameroon (the Sudanese-Sahelian zone and the humid equatorial zone) was assessed. The observations were made in Maroua and Yaoundé, during the 2019 and 2020 rainy seasons. In each agroecological zone, 480 flower heads were labeled and 2 treatments carried out, one left in open pollination and the other isolated from insects. The diversity of flower-visiting insects, their foraging behavior, regularity index, fruiting rate, and the percentage of achenes with seeds were evaluated. The results showed that *A. mellifera* was the most abundant floral visitor at both sites. In Maroua, out of 16 species of insect visitors recorded on the flowers of *H. annuus*, *A. mellifera* accounted for 41.40% of 5715 visits, while in Yaoundé, 11 species of flower-visiting insects were recorded, and *A. mellifera* accounted

for 51.38% of 3400 visits. In Maroua as in Yaoundé, *A. mellifera* harvested nectar intensively (80.85% and 85.19% of the foraging time). The mean abundance per 1000 flowers of *A. mellifera* on the capitula of *H. annuus* was  $107.5 \pm 9.90$  and  $138.95 \pm 39.05$  respectively in Maroua and Yaoundé. The regularity index of *A. mellifera* and the flowers of *H. annuus* was 0.41 (Maroua) and 0.51 (Yaoundé). Flower-visiting insects, including *A. mellifera*, increased the fruiting rate, the number of achenes with seeds and the percentage of achenes with normal seeds was 37.91%, 17.20% and 30.22% respectively. The corresponding values (Yaoundé) were 49.65%, 19.07% and 43.63%. In Maroua as in Yaoundé, the conservation of colonies of *A. mellifera* near plantations of *H. annuus* is recommended to increase yields of this plant.

**Keywords:** *Apis Mellifera*, *Helianthus Annuus*, Agroecological Zones, Pollination Services, Yields, Central Africa

#### 1. Introduction

The sunflower, *Helianthus annuus* is native to North America (Plant Biosafety Office, 2005) [33]. This crop is ideal for cultivation in any season in tropical climates because of its wide adaptability, drought tolerance, short life cycle, and photo and thermal insensitivity (Krishna *et al.*, 2014) [21]. It is cultivated primarily for its seeds, which yield the world's second most important source of edible oil (Plant Biosafety Office, 2005; Dwivedi and Sharma, 2014) [33, 12]. The seed oil, shoots, and herb tincture have been employed for anti-inflammatory, antipyretic, diuretic, stimulant and vermifuge purpose (Dwivedi and Sharma, 2014) [12]. Florets produce of nectar and pollen and are visited by insects (Vimla *et al.*, 2013) [41]. In Kenya, Honeybee pollination increases sunflower seed yield by 30% and oil content by more than 6% (Kasina *et al.*, 2007) [19]. They are important flower visitors not only in Kenya (Kasina *et al.*, 2007) [19] but also, in Turkey (Oz *et al.*, 2009) [30], Cameroon (Tchuenguem *et al.*, 2009; Faïbawa *et al.*, 2018; Egono *et al.*, 2018; Douka *et al.*, 202) [37, 15, 14, 10], Israël (Pisanty *et al.*, 2013) [32], India (Vimla *et al.*, 2013) [41] and Sudan (Osman and Siham, 2015) [29]. Non-*Apis* bees are also known to visit sunflower and have been reported to improve crop yield by enhancing efficiency of *Apis mellifera* (De Grandi and Watkins, 2000; Greenleaf and Kremen, 2006) [8, 16]. In Africa in general and in Cameroon in particular, the demand for sunflower seed oil is very high whereas seed yields are weak, primarily due to lack of awareness of importance of the role played by pollinators. (Kasina *et al.*, 2007, Nderitu *et al.*, 2008; Oz *et al.*, 2009; Vimla *et al.* 2013; Pysanty *et al.*, 2013; Osman and

Siham, 2015) [19, 28, 30, 41, 32, 29]. Before our work in Cameroon, the relationship between flower-visiting insects and sunflower are those of Tchuenguem et al. (2009) [37], Faïbawa et al. (2018) [15], Egono et al. (2018) [14] and Douka et al. (2021) [10] carried out in Ngaoundéré, in the High Guinean savannah ecoregion. These studies found that out of 50 species of insects that visited the florets of this plant, *A. mellifera* was the most frequent visitor. However, given that floral visitors and the impact of insects on pollination and plant yields can vary over time and space (Roubik, 2000; Tchuenguem, 2005) [34, 36], additional studies need to be carried out in novel agroecological zones. Broadly, this study seeks to contribute to our understanding of the plant-pollinator relationships between *H. annuus* et *Apis mellifera* L., with the goal of improving pollination management and increasing yields in two of Cameroon's agroecological zones. The four specific objectives include : (a) determining the diversity, relative frequency, and species richness of flower-visiting insects of *H. annuus*; (b) recording the foraging behaviour of *Apis mellifera* on the florets of this Asteraceae; (c) evaluating the impact of *A. mellifera* behaviour on the pollination of *H. annuus* in open vs. bagged treatments, (d) evaluating the impact of floral visitors including *Apis mellifera* on yield.

## 2. Materials and Methods

### 2.1 Study site and biological material

Investigations were carried out on two plots centered on points with the following geographical coordinates: latitude: 10°62' N; longitude: 14°33' E; altitude: 400 m (Maroua) and latitude 3° 51' North, longitude 11° 30' East and altitude 740 m (Yaounde). Maroua belongs to the Sudano-Sahelian ecological zone. The climate is of the Sudano-Sahelian type, characterized by two seasons: a long dry season (November to May) and a short rainy season (June to October); rainfall varies between 400 and 1100 mm; annual temperature oscillates between 29 and 38°C, with a daily thermal amplitude varying between 6 and 7°C (Kueté et al., 1993) [22]. Yaoundé belongs to the agro-ecological forest zone (dense tropical rainforest) with a bimodal rainfall regime. This zone enjoys a sub-equatorial climate, characterized by two alternating rainy seasons (March-June and August-November) and two dry seasons (November-March and June-August), and an annual rainfall of 1.7 to 2 m, spread over 150 days (Letouzey, 1985) [23]. The study station is a rectangular plot 23 m long by 14 m wide, with a surface area of 322 m<sup>2</sup>. The number of *A. mellifera* colonies close (500 m) to the experimental field was one each in Maroua and Yaoundé. Plant material consisted of *Helianthus annuus* seeds purchased from the Institute for Agricultural Research for Development in Yaoundé.

## 2.2 Methods

### 2.2.1 Plot establishment

During the rainy seasons from May to September 2019 and 2020 in Maroua and during the short rainy seasons from March to June 2019 and 2020 in Yaounde, the experimental plots were cleared, demarcated, and ploughed. Sowing was carried out in 9 rows of 20 holes per subplot, with 4 achenes per hole. The spacing between two holes was 40 cm within rows and 50 cm between rows.

### 2.2.2 Diversity, relative frequency, and specific richness of floral visitors to *Helianthus annuus*

In 2019 and 2020 in both Maroua and Yaoundé, observations were made on *H. annuus* flower heads every one day after three consecutive days, during four time slots: 08:00 - 09:00; 11:00 - 12:00; 13:00 - 14:00; 15:00 - 16:00. For each of these time slots, the various insects found on the flower heads were counted. As the insects were not marked, the cumulative results were expressed as the number of visits (Tchuenguem, 2005) [36]. Floral visitor counts enabled us to determine the place of insects in the anthophilous entomofauna of *H. annuus*. The percentage of visits by insect *i* to *H. annuus* florets ( $F_i$ ) was calculated using the formula below:  $F_i = \{[(V_i)/ VI] * 100\}$ , where  $V_i$  is the number of visits by insect *i* to the flowers of the open-flowering treatment and VI is the total number of visits by all insects to the same flowers (Tchuenguem et al. 1997) [38]. To assess the specific richness and diversity of *H. annuus* flowering insects in the two (02) study areas, Shannon's diversity index (H) (Dajoz, 2008) [7] and Piéou's equitability (EQ) (Dajoz, 2000) [6] were calculated using the formulas :

$$H = - \sum_{i=1}^S p_i (\log_2 p_i) \quad EQ = \frac{H}{\log_2(S)}$$

or  $p(i) = n(i)/N$ ;  $n(i)$ : 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 Jaccard index (Jaccard, 1908) [17] (J) was calculated:

$$J = \frac{c}{a+b-c}$$

where a = number of species from list a (survey A); b = number of species from list b (survey A'); c = number of species common to both surveys A and A'. This index was used to determine the similarity between the two sites.

### 2.2.3 Behavior of *Apis mellifera* on the florets of *Helianthus annuus*

#### 2.2.3.1 Floral products harvested

In both Maroua and Yaounde, the aim was to find out whether insects gathered nectar, pollen, or both from a flower. A bee that plunges its mouth parts or head deep into the corolla of a flower is a nectar gatherer; when it scratches the anthers with its mandibles and legs, it is a pollen gatherer. The pollen collected can be seen on the transport organs, notably in the corbiculae, or on the collecting hairs of the hind legs (Borror & White, 1991) [3]. Pollen harvesting can be active (when pollen is taken up) or passive (when, while collecting nectar, pollen accumulates on the insect's integument and is then collected in its storage organs) (Jousselin & Kjellberg, 2001) [18]. The floral products collected were systematically noted when recording the duration of visits per flower (Tchuenguem, 2005) [36]. Abundance is expressed by the greatest number of individuals simultaneously active on a flower or on 1000 flowers (Tchuenguem, 2005) [36].

#### 2.2.3.2 Relative abundance of *Apis mellifera* on the flowers of *Helianthus annuus*

In both Maroua and Yaoundé, abundance per flower was

recorded following direct counts. For abundance per 1000 flowers, individuals of each species were counted on a known number of opened flowers; abundance per 1000 flowers (A1000) was then calculated using the following formula:  $A1000 = [(Ax / Fx) \times 1000]$ , where Fx and Ax are respectively the number of opened flowers and the number of foragers counted on these flowers at time x (Tchuenguem, 2005) [36]. Data were recorded on the same dates as were the frequency of visits, at a rate of at least five readings for each bee when activity allowed.

### 2.2.3.3 Visitation length per capitulum of *Helianthus annuus*

The aim was to measure the time it takes a bee to collect a product (nectar and/or pollen) from a flower. As soon as a bee lands on a flower, the chronometer (brand SEIKO 11, made in China), previously set to zero, was triggered. It was not stopped until the bee left the flower. The time measured by the chronometer (in seconds) corresponds to the duration of the visit (Tchuenguem, 2005) [36]. Data on the duration of nectar and pollen collection visits were recorded separately, on the same dates and at the same times as forager abundance, at a rate of at least five readings per daily period and for *A. mellifera* studied, activity permitting, at the Maroua and Yaoundé sites.

### 2.2.4 Evaluation of the impact of *Apis mellifera* on the pollination of *Helianthus annuus*

For each year of the study and for each study site, the impact of flower-feeding insects, including *A. mellifera*, on pollination was assessed by studying the duration of visits. During nectar or pollen collection, the number of times a forager contacted the stigma of the flower visited was recorded. The regularity index (r) was calculated according to the formula:  $r = Pi \times fi$  where Pi is the percentage of visits and fi the frequency. This approach highlights the potential role of these insects in pollination.

### 2.2.5 Evaluation of the impact of insect visitors including *Apis mellifera* on the yield of *Helianthus annuus*

#### 2.2.5.1 Reproductive system

During the 2019 and 2020 rainy seasons in Maroua and the 2019 and 2020 short rainy seasons in Yaoundé, treatments 1 and 2, 3 and 4, 7 and 8, 9 and 10 were carried out as soon as flower buds appeared (Fig 1).

In Maroua, on August 22, 2019, 240 flowers at bud stage were tagged on 30 *Helianthus annuus* plants and two treatments were set up: treatment 1 made up of 120 flower buds (Fig 2) and treatment 2 made up of 120 flower buds protected from insects using gauze bags (Fig 3). On August 18, 2020, the same procedure was repeated. In Yaoundé on May 12, 2019 and May 20, 2020, the same experiment was carried out with the same number of flower buds.

At harvest, in both Maroua and Yaoundé, the number of fruits formed was counted on plants from each treatment. For each treatment, the fruiting index (Ifr) was calculated according to the following formula:  $Ifr = (Fy / Fx)$  where Fx is the number of flowers initially borne and Fy is the number of fruits formed (Tchuenguem, 2005) [36]. For each season of observation, the difference between the fruiting index in the treatment with flowers left to pollinate freely and that in the treatment with flowers protected from insects is used to calculate the rates of allogamy (TC) and autogamy in the broad sense (TA), according to the following formulas (Tchuenguem *et al.*, 2001) [39].  $TC = \{[(IfrX - IfrY) / IfrX] \times 100\}$ , where IfrX and IfrY are the average fruiting indices in

the treatment with flowers left open to pollination and in the treatment with flowers protected from insects respectively.  $TA = [100 - TC]$ .



Fig 1: Partial view of the experimental plots of *Helianthus annuus*



Fig 2: Capitula of *Helianthus annuus* in the open pollination treatment



Fig 3: Capitulum of *Helianthus annuus* protected from insect visits by a gauze bag.

#### 2.2.5.2 *Helianthus annuus* yields

Three parameters were selected for the study of fruit and grain yields: fruiting rate; number of seeds per flower head; and percentage of normal seeds, i.e. well-developed seeds. Assessment of the impact of insects including *A. mellifera* on yields was based on a comparison of fruit and seed yields from the open-flowering treatment (treatment a) and yields from the insect-protected flowering treatment (treatment b) (Tchuenguem, 2005) [36]. The percentage (Pfi) of fruiting due to the influence of flowering insects was calculated using the formula:  $Pfi = \{[(Fa - Fb) / Fa] \times 100\}$  where Fa and Fb are the fruiting rates in treatments a and b respectively. For treatment x, the fruiting rate (Fx) is:  $Fx = [(number\ of\ fruits/number\ of\ flowers) \times 100]$  (Tchuenguem, 2005) [36]; the percentage (Pgi) of the number of seeds per fruit due to the influence of flowering insects was calculated using the formula:  $Pgi = \{[(ga - gb) / ga] \times 100\}$  where ga and gb are the average number of seeds per fruit in treatments a and b respectively (Tchuenguem, 2005) [36]; the percentage (Pni) of normal seeds due to the influence of floricultural insects was calculated using the formula:  $Pni = \{[(gna - gnb) / gna] \times 100\}$  where gna and gnb are the percentages of normal seeds from treatments a and b respectively (Tchuenguem, 2005) [36].

### 2.2.6 Capture and identification of insect visitors

In both the Sudano-Sahelian and Equatorial zones, insects other than *A. mellifera* were captured on the capitulum (if possible, 3 to 10 specimens per insect species). Captures were made by hand, using tweezers or an insect net. Captured insects were preserved in flasks containing 70% ethanol, except for Lepidoptera and Odonata, which were preserved dry in glassine envelopes as recommended by Borror & White (1991) [3]. For each specimen, the time and date of capture were noted. Species not identified in the field were given a code and a brief description. Specimens were identified by, using specialist literature (Delvare & Aberlenc, 1989; Borror & White, 1991; Eardley *et al.*, 2010) [9, 3, 13].

### 2.2.7 Data analysis

Data analysis was carried out using: descriptive statistics (to calculate means, standard deviations and percentages); four tests: Student's t-test (to compare the means of two samples); Chi-square ( $\chi^2$ ) (to compare percentages); ANOVA (F) (to compare the means of more than two samples); Pearson's correlation coefficient (r) (to study linear relationships between two variables).

## 3. Results

### 3.1 Diversity, relative frequency, and specific richness of floral visiting insects to *Helianthus annuus*

In Maroua, during the 2019 and 2020 rainy seasons (May to September) respectively, 2967 and 2748 visits by 15 and

16 floral visitors were recorded on 120 *H. annuus* flowers per site. *Apis mellifera* was the most insect visitor with 41.40% and 41.41% of visits respectively (Table 1). In Yaoundé, during the short rainy seasons (March to June) 2019 and 2020 respectively, 1824 and 1576 visits by 10 and 11 insect species, respectively, were recorded on 120 *H. annuus* flowers. *Apis mellifera* was the most frequent insect visitor with 50.00% and 52.98% of visits to *H. annuus* flowers per site (Table 2). Across both agro-ecological zones, *A. mellifera* was the most frequent insect visitor, with visitation frequencies ranging from 75% to 100% (Tables 3 and 4). The Shannon-Weaver diversity index (H) was 2.58 and 2.39 respectively in 2019 in Maroua and Yaoundé. The difference observed between the Shannon-Weaver diversity indices of the two (02) sites in 2019 was non-significant ( $t = 0.77$ ;  $p > 0.05$ ). The Shannon-Weaver diversity index (H) was 3 and 1.75 respectively in 2020 at Maroua and Yaoundé. The difference between the Shannon-Weaver diversity indices of the two sites in 2020 was highly significant ( $t = 3.03$ ;  $p < 0.001$ ). Piéluou's equitability (EQ) was 0.66 and 0.75 in 2019 and 2020 respectively at Maroua. For the two cumulative years, Piéluou's equitability (EQ) was 0.70. The Jacard index calculation was  $J = 0.88$  at Maroua. Piéluou equitability (EQ) was 0.72 and 0.51 in 2019 and 2020 respectively in Yaoundé. For the two cumulative years, Piéluou's equitability (EQ) was 0.65. The calculation of Jacard's index was  $J = 0.92$  in Yaoundé (Table 5).

**Table 1 :** Insects recorded on the capitula of *Helianthus annuus* in 2019 and 2020 in Maroua: number and percent visits per insect species

Ordre	Famille	Insectes Genre, Espèce, Sous-espèce	2019		2020		2019/2020	
			$n_1$	$p_1$ (%)	$n_2$	$p_2$ (%)	$n_T$	$p_T$ (%)
	Calliphoridae	1 sp.	42	1.41	67	2.43	109	1.90
Diptera	Muscidae	<i>Musca domestica</i>	66	2.22	71	2.58	137	2.39
	<b>Apidae</b>	<b><i>Apis mellifera</i> (ne, po)</b>	<b>1226</b>	<b>41.4</b>	<b>1138</b>	<b>41.41</b>	<b>2364</b>	<b>41.40</b>
		<i>Amegilla</i> sp. (ne)	260	8.78	224	8.15	484	8.47
		<i>Chalicodoma cincta</i> (ne, po)	258	8.71	211	7.67	469	8.21
Hymenoptera		<i>Xylocopa olivacea</i> (ne, po)	497	16.78	391	12.22	888	15.55
	Halictidae	<i>Lipotriches collaris</i> (ne, po)	61	2.05	82	2.98	143	2.50
	Megachilidae	<i>Megachile</i> sp. 2 (ne)	86	2.9	77	2.8	163	2.85
		<i>Megachile</i> sp. 3 (po)	107	3.61	89	3.23	196	3.43
	Vespidae	<i>Belonogaster juncea</i> (ne)	77	2.6	85	3.09	162	2.83
		<i>Synagris cornuta</i>	68	2.3	55	2	123	2.15
Lepidoptera	Pieridae	<i>Catopsilia florella</i>	72	2.43	81	2.94	153	2.67
		<i>Eurema</i> sp. (ne)	85	2.87	69	2.51	154	2.69
	Acraeidae	<i>Acraea acerata</i>	57	1.98	41	1.49	98	1.71
Neuroptera		(1 sp.) (ne)	0	0	67	2.43	67	1.17
Mantodea	Mantidae	<i>Mantis religiosa</i>	5	0.16	0	0	5	0.08
	<b>Total</b>	<b>Visites</b>	<b>2967</b>	<b>100</b>	<b>2748</b>	<b>100</b>	<b>5715</b>	<b>100</b>
		<b>Espèces</b>	<b>(15 espèces)</b>		<b>(16 espèces)</b>		<b>(16 espèces)</b>	

visits to 120 flower heads for 20 days in 2019;  $n_2$ : number of visits to 120 flower heads over 20 days in 2020;  $n_T$ : total number of visits to 240 flower heads in 40 days; *sp.* : species not determined;  $P_1, P_2, p_T$ : percentages of visits:  $P_1 = (n_1 / 2967) * 100$ ;  $P_2 = (n_2 / 2748) * 100$ ;  $p_T = (n_T / 5715) * 100$ ; Comparison of the percentages of *Apis mellifera* visits (2019/2020): ( $\chi^2 = 0.02$ ;  $df = 1$ ;  $P > 0.05$ ); *ne*: nectar; *po*: pollen.

**Table 2:** Insects recorded on the flower heads of *Helianthus annuus* in 2019 and 2020 in Yaoundé, number and percentage of visits by different insects

Ordre	Famille	Insectes Genre, Espèce, Sous-espèce	2019		2020		2019/2020	
			$n_1$	$p_1$ (%)	$n_2$	$p_2$ (%)	$n_T$	$p_T$ (%)
	<b>Apidae</b>	<b><i>Apis mellifera</i> (ne, po)</b>	<b>912</b>	<b>50</b>	<b>835</b>	<b>52.98</b>	<b>1747</b>	<b>51.38</b>
		<i>Amegilla</i> sp. (ne)	102	5.6	74	4.69	176	5.17
		<i>Chalicodoma cincta</i> (ne, po)	255	13.98	197	12.5	452	13.29
Hymenoptera		<i>Xylocopa inconstans</i> (ne, po)	101	5.53	88	5.58	189	5.55
		<i>Xylocopa lugubris</i> (ne)	75	4.11	52	3.3	127	3.73
	Halictidae	<i>Lipotriches collaris</i> (ne, po)	53	2.9	36	2.28	89	2.61
	Megachilidae	<i>Megachile</i> sp. 2 (ne)	65	3.56	42	2.66	107	3.14

		<i>Megachile</i> sp. 3 (po)	109	5.97	89	5.64	198	5.82
	Vespidae	<i>Belonogaster juncea</i> (ne)	67	3.67	33	2.09	100	2.94
Lepidoptera	Pieridae	<i>Eurema</i> sp. (ne)	85	4.66	79	5.01	164	4.82
Neuroptera		(1 sp.) (ne)	0	0	51	3.23	51	1.5
	<b>Total</b>	<b>Visites</b>	<b>1824</b>	<b>100</b>	<b>1576</b>	<b>100</b>	<b>3400</b>	<b>100</b>
		<b>Espèces</b>	<b>(10 espèces)</b>		<b>(11 espèces)</b>		<b>(11 espèces)</b>	

visits to 120 flower heads for 20 days in 2019;  $n_2$ : number of visits to 120 flower heads over 20 days in 2020;  $n_T$ : total number of visits to 240 flower heads in 40 days;  $sp.$ : species not determined;  $P_1, P_2, pr$ : percentages of visits:  $P_1 = (n_1 / 1824) * 100$ ;  $P_2 = (n_2 / 1576) * 100$ ;  $P_T = (n_T / 3400) * 100$ ; Comparison of the percentages of *Apis mellifera* visits (2019/2020): ( $\chi^2 = 3.01$ ;  $df = 1$ ;  $P > 0.05$ );  $ne$ : nectar;  $po$ : pollen

**Table 3:** Frequency of insects recorded on the flower heads of *Helianthus annuus* in 2019 and 2020 in Maroua

Insectes	2019		2020		2019/2020		Comparaisons
	$n_1$	$f_1$ (%)	$n_2$	$f_2$ (%)	$n_T$	$f_T$ (%)	
<b>Genres et espèces</b>	$n_1$	$f_1$ (%)	$n_2$	$f_2$ (%)	$n_T$	$f_T$ (%)	
<i>Apis mellifera</i>	12	100	12	100	24	100	<b>100 ≥ F (%) ≥ 75 Insectes plus fréquents</b>
<i>Xylocopa olivacea</i>	12	100	12	100	24	100	
<i>Amegilla</i> sp.	10	83.33	8	66.66	18	75.00	
<i>Chalicodoma cincta</i>	10	83.33	6	50.00	16	66.66	<b>75 &gt; F (%) ≥ 25 Insectes fréquents</b>
<i>Musca domestica</i>	7	58.33	5	41.66	12	50.00	
<i>Megachile</i> (sp. 2)	6	50.00	5	41.66	11	45.83	
<i>Megachile</i> (sp. 3)	8	66.66	3	25.00	11	45.83	
<i>Acraea acerata</i>	6	50.00	4	33.33	10	41.66	
Calliphoridae (1 sp)	6	50.00	3	25.00	9	37.50	
<i>Lipotriches collaris</i>	5	41.66	4	33.33	9	37.50	<b>25 &gt; F (%) ≥ 10 Insectes occasionnels</b>
<i>Eurema</i> (sp. 1)	5	41.66	3	25.00	8	33.33	
<i>Catopsilia florella</i>	3	25.00	3	25.00	6	25.00	
<i>Belonogaster juncea</i>	5	41.66	2	16.66	7	29.16	<b>0 &gt; F (%) &gt; 10 Insectes rares</b>
<i>Synagris cornuta</i>	4	33.33	1	8.33	5	20.83	
<i>Mantis religiosa</i>	2	16.66	0	0	2	8.33	
Neuroptera (1 sp.)	0	0	1	8.33	1	4.16	
	<b>12</b>		<b>12</b>		<b>24</b>		

$n_1$ : number of days of observations in 2019;  $n_2$ : number of days of observations in 2020;  $n_T$ : total number of days of observations;  $sp.$ : species not determined;  $f_1, f_2, f_T$ : frequency of visits:  $f_1 = (n_1 / 12) * 100$ ;  $f_2 = (n_2 / 12) * 100$ ;  $f_T = (n_T / 24) * 100$ .

**Table 4:** Frequency of insects recorded on the flower heads of *Helianthus annuus* in 2019 and 2020 in Yaoundé

Insectes	2019		2020		2019/2020		Comparaisons
	$n_1$	$f_1$ (%)	$n_2$	$f_2$ (%)	$n_T$	$f_T$ (%)	
<b>Genre et espèce</b>	$n_1$	$f_1$ (%)	$n_2$	$f_2$ (%)	$n_T$	$f_T$ (%)	
<i>Apis mellifera</i>	12	100	12	100	24	100	<b>100 ≥ F (%) ≥ 75 Insectes plus fréquents</b>
<i>Chalicodoma cincta</i>	12	100	12	100	24	100	
<i>Amegilla</i> sp. 1	10	83.33	11	91.66	21	87.50	
<i>Xylocopa inconstans</i>	11	91.66	10	83.33	21	87.50	<b>75 &gt; F (%) ≥ 25 Insectes fréquents</b>
<i>Xylocopa lugubris</i>	8	66.66	6	50.00	14	58.33	
<i>Megachile</i> sp. 3	7	58.33	6	50.00	13	54.16	
<i>Lipotriches collaris</i>	5	41.66	3	25.00	8	33.33	
<i>Megachile</i> sp. 2	6	50.00	4	33.33	10	41.66	
<i>Belonogaster juncea</i>	5	41.66	4	33.33	9	37.50	
<i>Eurema</i> sp. 1	5	41.66	4	33.33	9	37.50	<b>25 &gt; F (%) ≥ 10 Insectes occasionnels</b>
Neuroptera (1 sp.)	0	0	5	41.66	5	20.83	
	<b>12</b>		<b>12</b>		<b>24</b>		

$n_1$ : number of days of observations in 2019;  $n_2$ : number of days of observations in 2020;  $n_T$ : total number of days of observations;  $sp.$ : species not determined;  $f_1, f_2, f_T$ : frequency of visits:  $f_1 = (n_1 / 12) * 100$ ;  $f_2 = (n_2 / 12) * 100$ ;  $f_T = (n_T / 24) * 100$

**Table 5:** Specific richness, diversity and similarity depending on location of study of *Helianthus annuus*

Années	Maroua			Yaoundé		
	Shannon (H')	Pielou (E)	Indice de Similarité de Jaccard (J)	Shannon (H')	Pielou (E)	Indice de Similarité de Jaccard (J)
2019	2.58	0.66	0.88	2.39	0.72	0.92
2020	3	0.75		1.75	0.51	
2019/2020	2.82	0.70		2.25	0.65	

### 3.2 Behavior of *Apis mellifera* on the flowers of *Helianthus annuus*

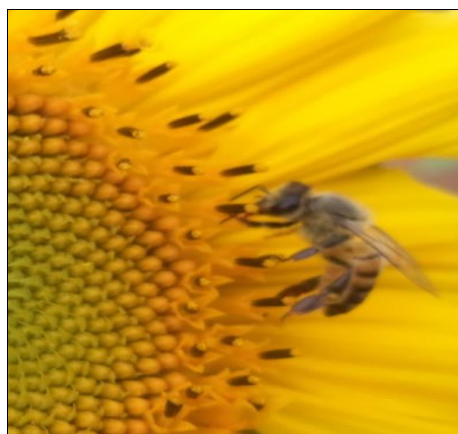
#### 3.2.1 Floral products harvested

*Apis mellifera* collected nectar and pollen from *Helianthus annuus* flowers (Table 6, Fig 2). Of 1946 and 1125 *A. mellifera* visits recorded in 2019 and 2020 in Maroua, 78.51% and 87.55% were devoted exclusively to nectar harvesting, 12.74% and 2.48% exclusively to pollen

harvesting. The difference in nectar harvesting between years is highly significant ( $\chi^2 = 51.52$ ;  $ddl = 1$ ;  $P > 0.001$ ); In Yaoundé, of 1112 and 1135 *A. mellifera* visits recorded in 2019 and 2020, 90.02% and 81.69% were devoted exclusively to nectar harvesting, 2.25% and 3.78% exclusively to pollen harvesting. The difference in nectar harvesting between years is highly significant ( $\chi^2 = 35.42$ ;  $ddl = 1$ ;  $P > 0.001$ ).

**Table 6:** Floral products harvested by *Apis mellifera* on the flowers of *Helianthus annuus* in 2019 and 2020 in Maroua and Yaoundé

site	Années	Nombre total de visites	Visites de récolte de nectar		Visites de récolte de pollen		Visites de récolte de nectar et pollen	
			Nombre	%	Nombre	%	Nombre	%
Maroua	2019	1946	1498	78.51	248	12.74	200	10.27
	2020	1125	985	87.55	28	2.48	112	9.95
	<b>Total</b>	<b>3071</b>	<b>2483</b>	<b>80.85</b>	<b>276</b>	<b>8.98</b>	<b>312</b>	<b>10.15</b>
Yaoundé	2019	1112	1001	90.02	25	2.25	86	7.73
	2020	1535	1254	81.69	58	3.78	223	14.53
	<b>Total</b>	<b>2647</b>	<b>2255</b>	<b>85.19</b>	<b>83</b>	<b>3.14</b>	<b>309</b>	<b>11.67</b>



*Apis mellifera* recolte le nectar



*Apis mellifera* recolte le pollen

**Fig 2:** *Apis mellifera* collecting nectar and pollen from the florets of *Helianthus annuus*

**Table 7:** Abundance of *Apis mellifera* per 1000 flowers on *Helianthus annuus* in 2019 and 2020 in Maroua and Yaoundé

Site	Années	N	Mean Abundance per 1000 fleurs (AMF)				Comparaison des moyennes
			M	s	Mini	maxi	
Maroua	2019	80	119.12	8.59	22	182	$F = 49.77; ddl = 3; P < 0.001; THS$ $Am_{(2019)}/Am_{(2020)} : t = 94.53; P < 0.001; THS$
	2020	80	95.37	11.21	6	134	
Yaoundé	2019	90	145.88	19.99	36	258	$F = 32.09; ddl = 3; P < 0.001; THS$ $Am_{(2019)}/Am_{(2020)} : t = 14.27; P < 0.001; THS$
	2020	90	132.02	58.11	86	176	
Maroua	2019	<b>160</b>	<b>107.24</b>	<b>9.90</b>	<b>14</b>	<b>182</b>	$t = 392.19; P < 0.001; THS$ $t = 187.99; P < 0.001; THS$
Yaoundé	2020	<b>180</b>	<b>138.95</b>	<b>39.05</b>	<b>36</b>	<b>258</b>	

AMF : Mean Abundance per 1000 Flowers; t : Student; F : Fisher; Am : *Apis mellifera*; THS : Très Hautement Significative.

**3.2.2 Relative abundance of *Apis mellifera* on the flowers of *Helianthus annuus***

The highest number of simultaneous individuals active on a flower head was 2.52 (n = 35; s = 0.18) in Maroua and 3.02 (n = 35; s = 0.02) in Yaoundé (Table 7). The table shows that the Mean Abundance per 1000 Flower (AMF) of *A. mellifera* on *H. annuus* flower heads ranged from 95.37 ± 11.21 (2020) to 119.12 ± 8.59 (2019) in Maroua and Yaoundé, the Mean Abundance per 1000 Flower (AMF) of *A. mellifera* on *H. annuus* flower heads ranged from 132.02 ± 58.11 (2020) to 145.88 ± 19.99 (2019).

**3.2.3 Duration of the visit**

In Maroua, the mean duration of an *A. mellifera* visit per *H. annuus* floret (MVD) was 5.87 sec ± 2.52 (2019) and 5.07 sec ± 2.41 (2020); the difference between these two means is highly significant (t = 15.14; P < 0.001); for the two cumulative years, MVD = 5.47 sec ± 2.46; the Mean Visit Duration of *A. mellifera* per floret for nectar harvest (MVDN) of *H. annuus* was 4.15 sec ± 1.90 (2019) and 4.90 sec ± 1.28 (2020); the difference between these two values is highly significant (t = -9.14; P < 0.001); for the two cumulative years, MVDN = 4.52 sec ± 1.59. The Mean Visit Duration of *A. mellifera* per floret for pollen harvesting (MVDP) of *H. annuus* was 3.30 sec ± 1.41 (2019) and 2.98

sec ± 3.88 (2020); the difference between these two figures is not significant (t = 1.96; P > 0.05); for the two cumulative years, MVDP = 3.14 ± 2.64 sec (Table 8).

In Yaoundé, the Mean Visit Duration (MVD) of *A. mellifera* per floret of *H. annuus* was 4.42 sec ± 0.82 (2019) and 2.89 sec ± 0.97 (2020); the difference between these two means is highly significant (t = 87.01; P < 0.001); for the two years combined, MVD = 3.65 sec ± 0.89; the Mean Visit Duration of *A. mellifera* per floret for Nectar harvesting (MVDN) of *H. annuus* was 3.95 sec ± 0.98 (2019) and 2.72 sec ± 0.18 (2020); the difference between these two values is highly significant (t = 45.29; P < 0.001); for the two cumulative years, MVDN = 3.33 sec ± 0.58; the Mean Visit Duration of *A. mellifera* per floret for pollen harvesting (MVDP) of *H. annuus* was 2.80 sec ± 0.22 (2019) and 1.90 sec ± 1.66 (2020); the difference between these two figures is highly significant (t = 10.54; P < 0.001); for the two cumulative years, MVDP = 2.35 ± 0.94 sec (Table 9).

**Table 8:** Duration of *Apis mellifera* visits by *Helianthus annuus* flower in 2019 and 2020 in Maroua

Année	Aliments Récoltés	n	Durée d'une visite par fleur (sec)			
			m	s	Mini	maxi
2019	Nectar	40	4.15	1.90	1	11
	Pollen	35	3.30	1.41	1	9

	Nectar + pollen	14	10.17	4.27	1	21
		<b>89</b>	<b>5.87</b>	<b>2.52</b>	<b>1</b>	<b>21</b>
	Nectar	50	4.98	1.28	1	15
	Pollen	20	2.98	3.88	1	12
2020	Nectar + pollen	9	7.27	2.07	1	17
		<b>79</b>	<b>5.07</b>	<b>2.41</b>	<b>1</b>	<b>17</b>
	Nectar	90	4.56	1.59	1	15
	Pollen	55	3.14	2.64	1	12
Total	Nectar + pollen	23	8.72	3.16	1	21
		<b>168</b>	<b>5.47</b>	<b>2.48</b>	<b>1</b>	<b>21</b>

**Table 9:** Duration of *Apis mellifera* visits by *Helianthus annuus* flower in 2019 and 2020 in Yaoundé

Année	Aliments	n	Durée d'une visite par fleur (sec)			
			Récoltés	m	S	Mini
2019	Nectar	60	3.95	0.98	1	16
	Pollen	25	2.80	0.22	1	6
	Nectar + pollen	15	6.52	1.31	1	19
		<b>100</b>	<b>4.42</b>	<b>0.82</b>	<b>1</b>	<b>19</b>
2020	Nectar	50	2.72	0.18	1	5
	Pollen	45	1.91	1.66	1	8
	Nectar + pollen	11	4.05	1.09	1	11
		<b>106</b>	<b>2.89</b>	<b>0.97</b>	<b>1</b>	<b>11</b>
Total	Nectar	110	3.33	0.58	1	16
	Pollen	70	2.35	0.94	1	8
	Nectar + pollen	26	5.28	1.20	1	19
		<b>206</b>	<b>3.65</b>	<b>0.90</b>	<b>1</b>	<b>19</b>

### 3.3 Impact of *Apis mellifera* on the pollination of *Helianthus annuus*

In both Maroua and Yaoundé (Tables 10 and 11), *Apis mellifera* was the major pollinator (Ir) with a high regularity index ( $Ir > 0.05$ ) and a high percentage of visits with stigma contact ( $p\% > 100\%$ ).

### 3.4 Impact of floral visitor including *Apis mellifera* on the yields of *Helianthus annuus*

#### 3.4.1 Reproductive system

In Maroua, the fruiting index was 0.95 for treatment 1; 0.10 for treatment 2; 0.92 for treatment 7 and 0.05 for treatment 8. Thus, for the year 2019, TC = 89.47% (Allogamy rate) and TA = 10.47% (Autogamy rate) and in 2020, TC = 94.56% and TA = 05.43%. For both study periods, TC = 92.01% and TA = 07.97%.

In Yaoundé, the fruiting index was 0.89 for treatment 3; 0.07 for treatment 4; 0.80 for treatment 9 and 0.03 for treatment 10. Thus, for the year 2019, TC = 92.13% (Allogamy rate) and TA = 7.86% (Autogamy rate) and in 2020, TC = 96.25% and TA = 03.75%. For both study periods, TC = 94.19% and TA = 05.80%.

Independent of the study area, *H. annuus* has a mixed allogamous - autogamous reproductive regime, with allogamy predominating (Table 12).

**Table 11:** Regulation index, number and percentage of visits with stigmatic contact on *Helianthus annuus* flowers in 2019 and 2020 in Maroua

Insectes	pv (%)			f (%)			Ir			NPV								
	2019	2020	T.	2019	2020	T.	2019	2020	T.	N			Ve			pc (%)		
										2019	2020	T.	2019	2020	T.	2019	2020	T.
<i>Apis mellifera</i>	41.40	41.41	41.40	100	100	100	0.4140	0.4141	0.4140	30	30	30	30	60	100	100	100	
<i>Xylocopa olivacea</i>	16.78	12.22	15.55	100	100	100	0.1678	0.1222	0.1450	30	30	30	30	60	100	100	100	
<i>Amegilla</i> sp.	8.78	8.15	8.47	83.33	66.66	75.00	0.0731	0.0543	0.0637	30	30	30	30	60	100	100	100	
<i>Chalicodoma cincta</i>	8.71	7.67	8.21	83.33	50.00	66.66	0.0725	0.0383	0.0554	30	30	30	30	60	100	100	100	
<i>Musca domestica</i>	2.22	2.58	2.39	58.33	41.66	50.00	0.0129	0.0107	0.0118	30	30	18	15	33	60.00	50.00	55.00	
<i>Megachile</i> (sp. 2)	2.90	2.80	2.85	50.00	41.66	45.83	0.0145	0.0116	0.0130	30	30	30	30	60	100	100	100	
<i>Megachile</i> (sp. 3)	3.61	3.23	3.43	66.66	25.00	45.83	0.0240	0.0080	0.0160	30	30	30	30	60	100	100	100	
<i>Acraea acerata</i>	1.98	1.49	1.71	50.00	33.33	41.66	0.0099	0.0049	0.0074	30	30	5	1	6	16.66	3.33	9.99	
<i>Calliphoridae</i> (1 sp)	1.41	2.43	1.90	50.00	25.00	37.50	0.0070	0.0060	0.0065	30	30	10	7	17	33.33	23.33	28.33	
<i>Lipotriches collaris</i>	2.05	2.98	2.50	41.66	33.33	37.50	0.0085	0.0099	0.0092	30	30	30	30	60	100	100	100	
<i>Eurema</i> (sp. 1)	2.87	2.51	2.69	41.66	25.00	33.33	0.0119	0.0062	0.0090	30	30	3	2	5	39.44	6.66	23.05	
<i>Catopsilia florella</i>	2.43	2.94	2.67	25.00	25.00	25.00	0.0060	0.0074	0.0067	30	30	3	1	4	10.00	3.33	06.66	
<i>Belonogaster juncea</i>	2.60	3.09	2.83	41.66	16.66	29.16	0.0108	0.0051	0.0079	30	30	0	0	0	0	0	0	
<i>Synagris cornuta</i>	2.30	2.00	2.15	33.33	8.33	20.83	0.0076	0.0016	0.0047	30	30	0	0	0	0	0	0	
<i>Mantis religiosa</i>	0	2.43	1.17	16.66	0	8.33	0	0	0	05	0	0	0	0	0	0	0	
<i>Nevroptera</i> (1 sp)	0.16	0	0.08	0	8.33	4.16	0	0	0	0	30	0	0	0	0	0	0	

**Table 11:** Regulation index, number and percentage of visits with stigmatic contact on *Helianthus annuus* flowers in 2019 and 2020 in Yaoundé

Insectes	pv (%)			f (%)			Ir			NPV								
	2019	2020	T.	2019	2020	T.	2019	2020	T.	n		Ve			pc (%)			
										2019	2020	2019	2020	T.	2019	2020	T.	
<i>Apis mellifera</i>	50.00	52.98	51.38	100	100	100	0.5000	0.5298	0.5149	30	30	30	30	60	100	100	100	
<i>Chalicodoma cincta</i>	13.98	12.50	13.29	100	100	100	0.1398	0.1250	0.1324	30	30	30	30	60	100	100	100	
<i>Amegilla</i> sp. 1	5.60	4.69	5.17	83.33	91.66	87.50	0.0466	0.0429	0.0344	30	30	30	30	60	100	100	100	
<i>Xylocopa inconstans</i>	5.53	5.58	5.55	91.66	83.33	87.50	0.0506	0.0466	0.0486	30	30	30	30	60	100	100	100	
<i>Xylocopa lugubris</i>	4.11	3.30	3.73	66.66	50.00	58.33	0.0273	0.0165	0.0219	30	30	30	30	60	100	100	100	
<i>Megachile</i> sp. 3	5.97	5.64	5.82	58.33	50.00	54.16	0.0348	0.0282	0.0315	30	30	30	30	60	100	100	100	
<i>Lipotriches collaris</i>	2.90	2.28	2.61	41.66	25.00	33.33	0.0120	0.0057	0.0088	30	30	25	11	36	83.33	36.66	59.99	
<i>Megachile</i> sp. 2	3.56	2.66	3.14	50.00	33.33	41.66	0.0178	0.0088	0.0133	30	30	30	30	60	100	100	100	
<i>Belonogaster juncea</i>	2.67	2.09	2.94	41.66	33.33	37.50	0.0111	0.0069	0.0090	30	30	00	00	00	00	00	00	
<i>Eurema</i> sp. 1	4.66	5.01	4.82	41.66	33.33	37.50	0.0194	0.0166	0.0180	30	30	05	07	12	16.66	23.33	19.99	
<i>Nevroptera</i> (1 sp.)	0	2.23	1.50	0	41.66	20.83	0	0.0092	0.0046	0	30	00	00	00	00	00	00	

**Ir** = (Pv/100) × (f/100); **Pv (%)**: percentage of visits; **f**: relative frequency of visits; **Ve**: studied visits; **NPV**: number and percentage of visits with stigmatic contact; **n**: number of visits with stigmatic contact; **pc (%)**: percentage of visits with stigmatic contact; **T**: Total.

**Table 12:** Mode of reproduction of *Helianthus annuus* in 2019 and 2020 in Maroua and Yaoundé

Sites	Traitements	NFE	NAF	IF	TC	TA
Maroua <sub>2019</sub>	1	120	56810	0,95	89.47 %	10.47 %
	2	120	3155	0,10		
Maroua <sub>2020</sub>	7	120	19976	0,92	94.56 %	05.43 %
	8	120	5477	0,05		
<b>Total</b>					<b>92.01 %</b>	<b>07.97 %</b>
Yaoundé <sub>2019</sub>	3	120	58049	0,89	92.13 %	07.86 %
	4	120	1724	0,04		
Yaoundé <sub>2020</sub>	9	120	18216	0,80	96.25 %	03.75 %
	10	120	1336	0,03		
<b>Total</b>					<b>94.19 %</b>	<b>05.80 %</b>
<b>Total cumulé</b>					<b>93.10 %</b>	<b>6.98 %</b>

IF: Fruiting Index, NFE: Number of Blooming Florets, NAF: Number of Achenes Formed, TA: Autogamy Rate, TC: Allogamy Rate.

**3.4.2 Fruiting rate**

In Maroua, in 2019 and 2020, for both rainy seasons. The average fruiting rate was 93.99% in treatments 1 and 7 (flowers left to pollinate freely and foraged by all flowering insects) and 7.90% in treatments 2 and 8 (flowers protected from flowering insects); the difference between these two percentages is very highly significant ( $\chi^2 = 160411.98$ ; ddf = 1;  $P < 0.001$ ). In Yaoundé, in 2019 and 2020, for both rainy seasons. The average fruiting rate was 85.29% in treatments 3 and 9 (flowers left to pollinate freely and foraged by all flowering insects) and 4.12% in treatments 4 and 10 (flowers protected from flowering insects); the difference between these two percentages is very highly significant ( $\chi^2 = 111006.35$ ; ddf = 1;  $P < 0.001$ ) (Table 13).

**3.4.3 Achenes with seed**

In Maroua, the average percentage of achenes with seeds was 92.42% in treatments 1 and 7 (open pollination treatment and foraged by all insects) and 58.30% in treatments 2 and 8 (flowers protected from insects); the difference between these two percentages is highly

significant ( $\chi^2 = 14485.45$ ; ddf = 1;  $P < 0.001$ ). In Yaoundé, the average percentage of achenes with seeds was 93.78% in treatments 3 and 9 (open pollination treatment and foraged by all insects) and 71% in treatments 4 and 10 (flowers protected from insects); the difference between these two percentages is highly significant ( $\chi^2 = 7579.96$ ; ddf = 1;  $P < 0.001$ ) (Table 13).

**3.4.4 Normally developed seeds**

In Maroua, the percentage of achenes with normal seeds was 95.31% in treatments 1 and 7 (open pollination treatment and foraged by all insects) and 25.31% in treatments 2 and 8 (flowers protected from insects); the difference between these two percentages is highly significant ( $\chi^2 = 31613.84$ ; ddf = 1;  $P < 0.001$ ). In Yaoundé, the percentage of achenes with normal seeds was 87.38% in treatments 3 and 9 (open pollination treatment and foraged by all insects) and 14.30% in treatments 4 and 10 (flowers protected from insects); the difference between these two percentages is highly significant ( $\chi^2 = 11503.00$ ; ddf = 1;  $P < 0.001$ ) (Table 13).

**Table 13:** Fruiting rate, average number of achenes per flower head and percentage of normal seeds according to *Helianthus annuus* treatments in Maroua and Yaoundé in 2019 and 2020

Années	Tts	NCE	NFE	NTAF	TF (%)	NAG	% AG	NAN	% AN
2019 Maroua	T1	120	59643	56810	95.25	54918	96.66	53090	96.67
	T2	120	31238	3155	10.10	1596	50.58	385	24.12
2020 Maroua	T7	120	21612	19976	92.43	17793	89.05	16720	93.96
	T8	120	98930	5477	5.90	3148	57.47	858	27.25
<b>Total</b>	<b>T1 et 7</b>	<b>240</b>	<b>81255</b>	<b>76786</b>	<b>93.99</b>	<b>72711</b>	<b>92.85</b>	<b>69810</b>	<b>95.31</b>
	<b>T2 et 8</b>	<b>240</b>	<b>130168</b>	<b>8632</b>	<b>7.90</b>	<b>4744</b>	<b>54.02</b>	<b>1243</b>	<b>25.68</b>
2019 Yaoundé	T3	120	64714	58049	89.70	57192	98.52	51200	89.52
	T4	120	39632	1724	4.35	960	55.68	151	15.72
2020 Yaoundé	T9	120	22519	18214	80.88	15725	86.33	13405	85.24
	T10	120	34256	1336	3.90	814	60.92	105	12.89
<b>Total</b>	<b>T3 et 9</b>	<b>240</b>	<b>87233</b>	<b>76263</b>	<b>85.29</b>	<b>72917</b>	<b>92.42</b>	<b>64605</b>	<b>87.38</b>
	<b>T4 et 10</b>	<b>204</b>	<b>73880</b>	<b>3060</b>	<b>4.12</b>	<b>1774</b>	<b>58.30</b>	<b>256</b>	<b>14.30</b>

Tts: Treatments; NCE: Number of flower heads studied; NFE: number of flagships studied; NTAF: number of achenes per flower head; TF: fruiting rate; NAG: number of achenes with seeds; % AG: percentage of achenes with seeds; NAN: number of normal achenes; % AN: percentage of normal achenes

**3.5 Impact of *Apis mellifera* on the yield of *Helianthus annuus***

In Maroua, the contributions of *A. mellifera* to the fruiting rate of *H. annuus* were 37.00% (2019) and 38.83% (2020). The percentages of achenes with seeds due to *A. mellifera* were 19.73% (2019) and 14.68% (2020). The contribution of *A. mellifera* to the percentage of achenes with normal seeds was 31.06% (2019) and 29.39% (2020). For the two seasons combined, the contributions of *A. mellifera* to the

fruiting rate, number of achenes with seeds and percentage of achenes with normal seeds were 37.91%, 17.20% and 30.22% respectively. In Yaoundé, the contributions of *A. mellifera* to the fruiting rate of *H. annuus* were 50.41% (2019) and 48.89% (2020). The percentages of achenes with seeds due to *A. mellifera* were 23.03% (2019) and 15.12% (2020). The contribution of *A. mellifera* to the percentage of achenes with normal seeds was 43.67% (2019) and 43.60% (2020). For the two seasons combined, the contributions of



*A. mellifera* to the fruiting rate, number of achenes with seeds and percentage of achenes with normal seeds were 49.65%, 19.07% and 43.63% respectively.

#### 4. Discussion

##### 4.1 Diversity, relative frequency and specific richness of floral visiting insects to *Helianthus annuus*

In the two agro-ecological zones (Maroua and Yaoundé), we inventoried 16 and 11 species of pollinating insects on *H. annuus* flowers respectively. *Apis mellifera*, ranked 1st with 41.40% and 51.38 % respectively in Maroua and Yaoundé ( $X^2 = 86.40$ ;  $p < 0.001$ ). *Apis mellifera*, was the most frequent insect on *Helianthus annuus* flowers in Yaoundé. The similarity indices and specific richness in the two agro-ecological zones are variable. These results are in line with observations made in Cameroon by Tchuenguem *et al.* (2009) [37] and Egono *et al.* (2018) [14] and in Sudan by Ahmed *et al.* (1989) [1], which showed the honey bee to be the most frequent insect visitor on *H. annuus* florets, attracted by the floral products presented by *Helianthus annuus* flowers.

##### 4.2 Behavior of *Apis mellifera* on the flowers of *Helianthus annuus*

*Apis mellifera* harvests mainly nectar and to a lesser amount, pollen, on *Helianthus annuus* flowers in both Maroua and Yaoundé. Our data do not agree with those obtained by Egono *et al.* (2018) [14] in Dang (Cameroon), who reported that these bees harvest mainly pollen and less so nectar from *H. annuus* flowers. This high nectar collection in *H. annuus* by *A. mellifera*, would be explained by the high availability of nectar, the attractiveness and accessibility of this floral product vis-à-vis these bees as well as their nutritional needs during the flowering period of this plant species. On *H. annuus* flowers in Maroua, the highest number of *A. mellifera* workers simultaneously active per 1000 flowers was  $107.24 \pm 9.90$ . In Yaoundé, this abundance was  $138.95 \pm 39.05$  and  $32.90 \pm 19.01$ . The high abundances per 1000 flowers recorded highlight the high attractiveness of the floral products of *H. annuus* and *A. mellifera*. The high abundance may be linked to their ability to recruit large numbers of foragers to exploit an attractive food source (Louveaux, 1984) [26]. On *H. annuus* flowers in Maroua, *A. mellifera* visited a floret for an average of  $5.47 \pm 2.48$  seconds, while in Yaoundé, visits lasted  $3.64 \pm 0.90$  seconds. The duration of a nectar collection visit was significantly higher than that of a pollen collection visit ( $P < 0.001$ ), irrespective of the study site. This difference could be explained by the accessibility of each of the floral products, as reported by Tchuenguem (2005) [36] in *Callistemon rigidus* and *Syzygium guineense* var. *macrocarpum*: pollen is produced by the anthers, which are located below the stamen threads and are easily accessible to bees, whereas nectar is found in the corolla tube between the base of the style and the stamens, making it difficult to access. Visit duration also varies with nectar availability. *Apis mellifera* stays longer on nectar-rich flowers than on nectar-poor ones (Chittka *et al.*, 2003, Egono *et al.*, 2019; Douka *et al.*, 2021; 2022) [5, 14, 10, 11].

##### 4.3 Impact of *Apis mellifera* on the pollination of *Helianthus annuus*

During the foraging activity of *A. mellifera* on *H. annuus* flowers in Maroua and Yaoundé, *Apis mellifera* transported

pollen from floret to floret, using its legs, mouthparts, thorax, and abdomen. These parts were always in contact with the stigmas of the foraged flowers. This stigmatic contact would increase the probability of pollination through the deposition of pollen on the stigmas. In this way, visit frequencies accompanied by high regularity indices were likely to play a positive role in geitonogamy (Klin khamer & de Jong, 1993) [20], by placing pollen from one flower on the stigma of another flower on the same plant. As bees moved from flower to flower on different plants, they were able to transport pollen from one plant to another. They therefore facilitate xenogamy (Lobreau-Callen & Coutin, 1987) [25], by placing the pollen of one plant on the stigma of another plant's flower. In this way, we have noted the increase in pollination possibilities for this plant.

##### 4.4 Reproductive systems and yields of *Helianthus annuus*

*Helianthus annuus* in Maroua and Yaoundé, was studied in two rainy seasons 2019 and 2020; showing that *H. annuus* has a mixed allogamous-autogamous reproductive system, with allogamy predominating. This result (autogamy: 6.98%; allogamy: 93.10%) agrees with that obtained by Tchuenguem *et al.* (2009) [37] and Engono *et al.* (2018) [18] in Ngaoundéré, which indicates the predominance of allogamy over autogamy. This predominance of allogamy is thought to be due to self-incompatibility in these plant species (Chapman & Reiss, 1992) [4]. As fruiting is mainly dependent on pollination intensity (Mc Gregor, 1976) [27], the significant increase in yields in the presence of *A. mellifera* is the consequence of this bee's foraging activity on the pollination of the flowers of the plant studied. Work by other researchers on other plants has shown that *A. mellifera* plays a positive and significant role in fruit and/or seed yields thanks to its pollinating activity: *Actinidia deliciosa* (Vaissière *et al.*, 1996) [40]; *Adenocalymma bracteatum* (Almeida-Soares *et al.*, 2010) [2]; *Allium cepa*, *Passiflora edulis* and *Cucumis melo* (Philippe, 1991) [31]; *Allium cepa* (Tchindébé & Tchuenguem, 2014) [35].

#### 5. Conclusion

Thus, it was observed that flowering insects including *A. mellifera* enhance the productivity of *H. annuus*. High relative visitation frequencies along with the high potential for pollen transport observed suggest that sunflower pollination can benefit highly from *A. mellifera* visits. The comparison of seeds set of protected flowers with that of visited by all insects including *A. mellifera* underscores the value of these insects in increasing pod and seed set as well as seed quality. This study demonstrates the importance of pollinator visits to all yield parameters in *H. annuus* cultivation, and suggests that efforts to safeguard or increase pollinator abundance, in particular that of *A. mellifera* colonies in proximity to sunflower fields, can be important for ensuring high yields.

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