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Diversity, abundance and activity of bees in Zucchini (*Cucurbita pepo* L) crops in northern Côte d'Ivoire

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Abstract
Zucchini is booming in Korhogo in northern Côte d'Ivoire. The fecundation of this plant is ensured by pollination. However, the pollinating insects of this plant are poorly studied in Korhogo. The diversity, abundance and activities of pollinating insects were evaluated by direct observation in 2016 and 2017 on a plot of 63 m^2 set up within the vegetable patch of the Peleforo Gon Coulibaly University of Korhogo. It's appear that zucchini is pollinated by bees *Apis mellifera*, *Bombus impatiens* and *Lasioglossum interruptum*. The peak activity of these bees was between 6 and 7 am. The number of visits and the visitation time were higher on the female flowers than male flowers. The activity of these bees varied in proportion to the relative humidity and inversely proportional to the temperature. *A. mellifera* was the main pollinator of zucchini in Korhogo.

Keywords: zucchini, pollinating insects, Ivory Coast

1. Introduction
Zucchini (*C. pepo*) is growing rapidly in Korhogo in northern Côte d'Ivoire (Koné *et al*., 2018) [1]. According to FAO estimates in 2014, zucchini production in Côte d'Ivoire was 19,296 tonnes. Zucchini is a vegetable rich in protein, amino acids, minerals, vitamins and fatty acids. Zucchini seed and leaf extracts are used in the treatment of many urogenital diseases (European Medicines Agency, 2012) [2]. In Korhogo, zucchini is used as an alternative to eggplant and cabbage in the preparation of sauces (Koné *et al*., 2018) [1]. According to Erard (2002) [3], the cycle of production of zucchini is on average 45 days while that of eggplant is about 120 days. This makes this vegetable an interesting asset in the fight against food insecurity due to its short cycle. However, the increase of its production is limited by several constraints, including insufficient pollination (Walters and Taylor, 2006) [4]. Indeed, the works of Artz and Nault (2011) [5] and Enriquez *et al*. (2015) [6] showed that zucchini cannot produce fruit in the absence of pollinating insects. Zucchini is pollinated by wild bees (Enriquez *et al*., 2015) [6] and honey bee *A. mellifera* (Walters and Taylor, 2006; Frazier *et al*., 2015) [4, 7]. In addition, for Delaplane and Mayer (2000) [8], the increase in the number of bee visits on cucurbit flowers generally leads to an increase in production. However, in the sub-prefecture of Korhogo, there is very little data on the pollinating bees of zucchini. This study aims to inventory and evaluate the activities of bee species visiting zucchini flowers.

2. Material and Methods
2.1 Study site
This study was conducted over two years during rainy and dry season, on experimental plot housed in the botanical garden of Peleforo Gon Coulibaly University in the sub-prefecture of Korhogo, located between 8°26 and 10º27 N, and 5º17 and 6º19 W, 600 km from Abidjan in the north of the Côte-d'Ivoire. This sub-prefecture belongs to the Sudano-Sahelian dry tropical climate regime in which the rhythm of the seasons is regulated by the displacement of the Intertropical Front (Jourda *et al*., 2005) [9]. This climate is characterized by two seasons. The rainy season extends from May to October with a maximum of precipitation in September. The dry season lasts from November to April and is characterized by the harmattan that settles from December to February (Kouakou *et al*., 2012) [10].

2.2 Experimental device
The varieties of zucchini used are Aurore F1 and Color F1, main varieties produce in Korhogo. The experimental device was a block of Fisher, completely randomized with two (2) blocks and five (5) repetitions per blocks. Each variety of zucchini was represented by a block, with two (2) objects (T0=untreated plots, T1=plots treated with approved synthetic insecticide). Treatment was realized once a week. The blocks, were spaced 1 m apart and each made up of 5 elementary plots, making a total of 20 elementary plots for the entire system. Each elementary parcel consisted of 4 holes with gaps of 0.70 m between them. The total area of the experimental plot was 63 m^2 (9m x 7). An insecticide registered in market gardening in Côte d'Ivoire, Cypercal 50 EC (Cypermethrin) was applied once a week on the treated plots.

2.3 Assessment of the role of bees in the pollination of zucchini
In order to show the impact of bees in the pollination of zucchini, 30 female flowers were protected from the contact of bees, using muslin bags, compared to 30 other unprotected ones during each cropping cycle. Flowers were covered before anthesis, to prevent visitors of flowers from accessing them. At the end of anthesis, the flowers were discovered to let the fruit grow (Enriquez *et al*., 2015) [6].

2.4 Diversity, Abundance and Pollinator Activities

The diversity, abundance and activities of pollinators were evaluated during flowering, between 5:00 am and 11:00 am, opening time for zucchini flowers (Artz and Nault 2011, Enriquez *et al*., 2015) [5,6]. The observations took place according to the following time slots: 5H00-6H00; 6H00-7H00; 7H00-8H00; 8H00-9H00; 9:00-10:00 and 10:00-11:00 am. Using a thermo-hygrometer, mean temperature and relative humidity were recorded for each time slot. On the day of observation, five (5) open female flowers were randomly selected. During each time slot, each of the five female flowers was observed for 10 minutes. At the level of each flower, the number of individuals per species and the visit time of each individual were noted (Shuler *et al*., 2005) [11]. Bee's activities were evaluated once a week for three consecutive weeks since the start of flowering (Petersen *et al*., 2014) [12].

3. Results

3.1 Assessment of the role of bees in the pollination of zucchini

No fruit was harvested from protected flowers compared to unprotected flowers. All bagged female flowers aborted.

3.2 Diversity and relative abundance of bees

The bees collected from the male and female flowers belonged to three species. There are: *Apis mellifera* L. 1758, *Bombus impatiens* Cresson 1863, of the family Apidae, and *Lasioglossum interruptum* Panzer 1798 of the family Halictidae. These three species belong to the order Hymenoptera. Over the two years of study, a total of 3078 bees visiting the zucchini flowers were counted. The most abundant specie was *A. mellifera* with 2862 individuals, or 92.98% of the total population. It was followed by *L. interruptum* with 204 individuals or 6.63%. With a total population of 12 individuals or 0.39%, *B. impatiens* was the least common species on zucchini flowers in Korhogo. Statistical analyzes revealed a significant difference between the relative abundance of bee species ($F = 20.29$, $p < 0.05$) (Figure 1).

3.3 Daily activity of bees

3.3.1 Male flowers

On *C. pepo* male flowers, the daily activity of *A. mellifera* and *L. interruptum* varied in bell-shaped form. For these two species, the peak of activity was observed between 5 am and 6 am. Beyond this period, visits gradually decreased to cancel at 9 am for *L. interruptum* and at 10 am for *A. mellifera* (Figure 2).

3.3.2 Female flowers

As with male flowers, the daily activity of *A. mellifera* and *L. interruptum* varied in bell-shaped form. The peak activity of these two species was observed between 5 am and 6 am. The number of *L. interruptum* fell rapidly to cancel around 8 am. Activity of *A. mellifera*, decreased progressively to cancel towards 10 am. *B. impatiens* was the third specie visiting female flowers. The only identified individuals were observed between 8 am and 9 am. Outside this period, nil *B. impatiens* individuals were observed on zucchini flowers (Figure 3).

3.4 Influence of abiotic facts on the daily activity of bees

The daily activity curve of the three species of bees is superimposable to that of relative humidity. For all three bee species, when the relative humidity is high, bee activity is high. As relative humidity decreases bee activity decreases (Figure 4). There is a positive correlation between bee activity and relative humidity ($r = 0.56$). For temperature, the fluctuations are inversely proportional to the activity of the three (3) species bees.

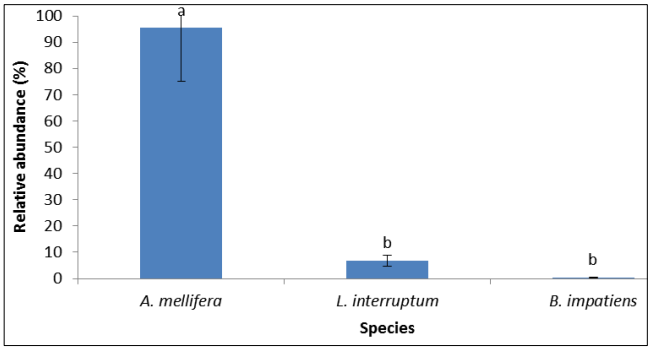

Fig 1: Relative abundance of bee species

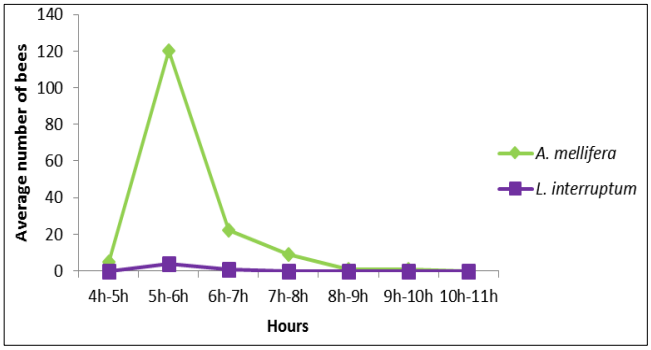

Fig 2: Daily activity of *A. mellifera* and *L. interruptum* on male flowers

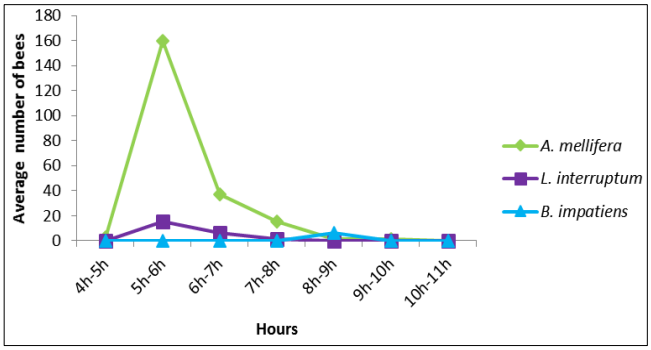

Fig 3: Daily activity of *A. mellifera*, *L. interruptum* and *B. impatiens* on female flowers

When the temperature is low, the activity of the bees is high, however when the temperature is high, the activity is low (Figure 5). There is a positive correlation between temperature and bee activity ($r = 0.55$).

3.5 Influence of the type of flower

The number of bee visits varied according to the type of flower. Out of a total of 3078 bees observed, the female flowers recorded 1780 individuals or 57.83%. Male flowers were the least visited with 1298 or 42.17% of visits. For *A. mellifera*, the greatest number of visits was recorded on female flowers with 56.11% against 43.89% of visits on male flowers. *L. interruptum* visited female flowers (79.41%)

more than male flowers (20.59%). About *B. impatiens*, highest visits were recorded on female flowers (Figure 6).

3.6 Influence of the flower type on the foraging time of bees

All bee species combined, mean visit time varied significantly by flower type ($F = 21.29$, $p < 0.05$). On average, bees spent more time on female flowers than male flowers. On female flowers the average visit times were 39.01 ± 4.1 seconds against 12.59 ± 1.5 on the male flowers. For *A. mellifera*, the mean visitation time varied significantly by flower type ($F = 26.28$, $p < 0.05$). It was higher on female flowers (32.81 ± 2.86) than on male flowers (12.21 ± 1.45). For *L. interruptum*, mean visitation time varied significantly with flower type ($F = 4.88$, $p < 0.05$). This bee spent an average of 97.98 ± 28.15 seconds on the female flowers during her visits. In contrast, on male flowers, visits lasted on average 30.8 ± 11.46 seconds (Figure 7). The visitation time of *B. impatiens* averaged 5 seconds.

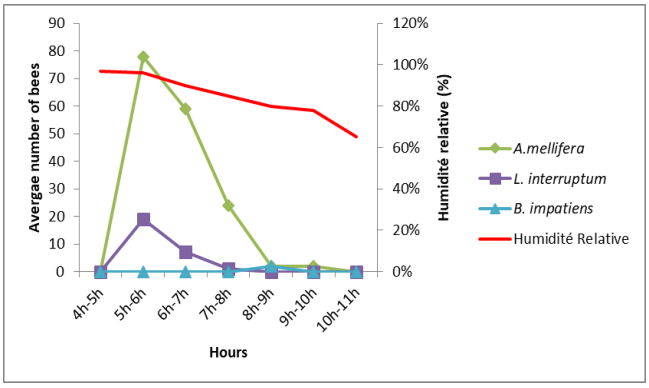

Fig 4: Variation of *A. mellifera*, *L. interruptum* and *B. impatiens* activity as a function of relative humidity

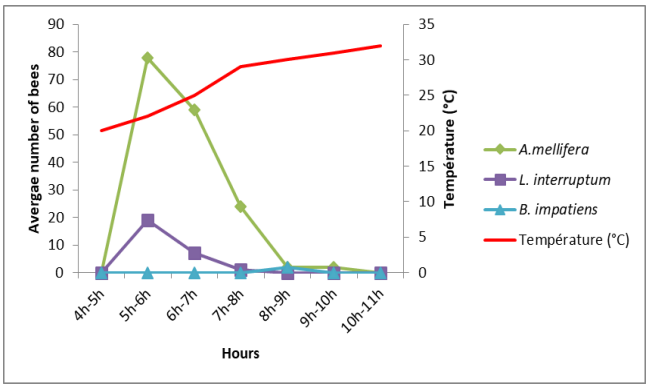

Fig 5: Variation of *A. mellifera*, *L. interruptum* and *B. impatiens* activity as a function of temperature

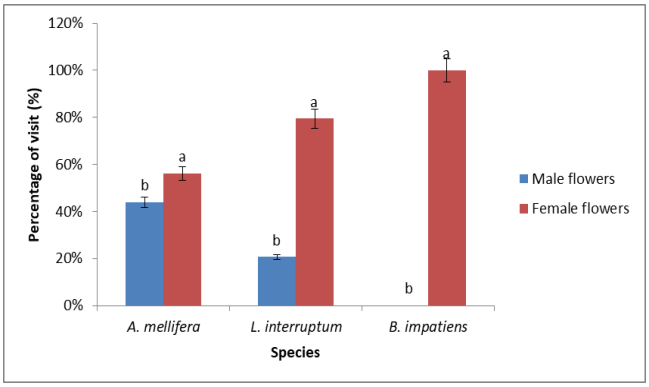

Fig 6: Influence of the type of flower on the number of bee visits

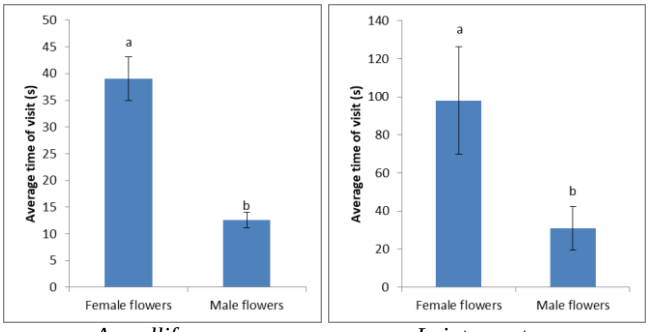

Fig 7: Average visit time of bees according to the type of flower

4. Discussion

From analyses of diversity and relative abundance of bees, it was found that the pollinators of the zucchini were *A. mellifera*, *L. interruptum* and *B. impatiens*. During the study, these species visited both male and female flowers. These results are close to those reported by Shuler *et al*. (2005) [11]. According to these authors, the main pollinators of zucchini are the bee *P. pruinosa*, *A. mellifera* and *B. impatiens*. Similar results have been obtained by Artz *et al*. (2011) [13]. In their study on zucchini, these authors collected 16 species of bees, including *P. pruinosa*, *A. mellifera* and *B. impatiens* which represented 99% of the total population. Of all three bee species observed on zucchini flowers, *A. mellifera* was the most active during study. This strong activity could be explained by the attractiveness of zucchini flowers on this bee. Indeed, the presence of bees on the flowers is in relation with intensity of the perfume emitted by the flowers (Corbet *et al*., 1991) [14]. Bees search nectar which constitutes their food. The attraction of pollinators by flowers would be function of the concentration of compounds secreted by the flowers. Pouvreau (1984) [15] showed that the concentration of sugar in oil palm flowers would be insufficient to attract the insects of the Apidae family. These insects prefer flowers whose sugar concentration of the nectar is relatively high, in particular the Cucurbitaceae (Corbet, 1997) [16]. According to Benachour and Louadi (2013) [17], cucumber flowers (Cucurbitaceae) attract many visitors, most of whom are Apidae. The high abundance of *A. mellifera* may be due to its social character. This species can form colonies of 25,000 individuals (Walters and Taylor, 2006) [4]. This ability would allow them to colonize more easily crops in flowering. The highest number visits of *A. mellifera* compared to that of the other species would be explained by his optimal foraging strategy. According to Atibita *et al*. (2015) [18], *A. mellifera* use a high number of foragers to exploit an interesting food source. This situation would result by the limitation of the visits number of other insect's species due to the occupation of most flowers by *A. mellifera*. In our study, female zucchini flowers were protected to prevent them from bees. Of all the protected flowers none gave fruit. This reflects the impact of bees in the pollination of zucchini. These results are close to those of Couto *et al*. (1990) [19]; Nicodemo *et al*. (2009) [20]. These authors have shown that in zucchini crops, no fruit can be produced in the absence of pollinating insects. On the other hand, flowers left in open pollination produced fruit in at least 32% of cases.

Daily activity of bees

For both types of flowers, the peak activity of *A. mellifera* and *L. interruptum* was between 5 am and 6 am. This could be explained by the high availability of resources collected

by these species during these periods. According to Corbet *et al*. (1991) [14] the presence of bees on flowers is function of the intensity of the fragrance emitted by the flowers. Pollinating insects search nectar which constitutes their food. Production and quality of nectar varies with time of day, flower age, species, and weather (Pouvreau 1984) [15]. Atwal (1970) [21] found that bee visits to flowers are more numerous 2 to 3 hours after sunrise. The first hours of the morning correspond to the period when the flowers produce a maximum of pollen and nectar that induce intense activity of the bees (Nepi and Pacini 1993) [22].

This important activity of the bees ensures an adequate pollination of zucchini flowers that's remaining open only one day. In addition, the end of the foraging activities of *A. mellifera* and *L. interruptum* was at 10am. This hour corresponded to the closing of zucchini flowers. The male flowers and female flowers open only one day, early in the morning to close definitively around 11H (Artz and Nault, 2011) [5]. It is known that the daily activity of foraging honeybees on flowers depends on their pollen production (Stone *et al*., 1998) [23] and nectar (Suzo *et al*., 2001) [24]. Influence of temperature and relative humidity on bee activity was evaluated. Bee's activity was proportional to the relative humidity changes. However, for temperature, the fluctuations were inversely proportional to the activity of the three (3) bee species. Similar observations have been made by Atibita *et al*. (2015) [18] on winter cherry and by Julianna and Ruffus (2010) [25] on blueberries. These authors have shown that abiotic parameters such as temperature and relative humidity influence the activity of pollinating insects. According to Blazyte-Cereskiene *et al*. (2010) [26], high temperature has a negative effect on bees foraging. Djonwangwe *et al*. (2011) [27] reported that climatic conditions influenced the foraging activity of *A. m. adansonii* on the flowers of *Vitelaria Paradoxa*. About visits, the number of bee visits varied according to the type of flower. For all pollinator species, female flowers were more visited than male flowers. This could be explained by the offer of these two types of flowers to bees. In their work on zucchini, Artz *et al*. (2011) [13] showed that pollinators of this plant have a preference for female flowers. This is because female flowers are three times more concentrated in nectar than male flowers. In addition, the nectar of female flowers is richer in sugars than that of male flowers (Couto *et al*., 1990) [19]. This study revealed that bees spent much time on female flowers than male flowers. These results are similar to those obtained by Tepedino (1981) [28]. According to this author, the bee's visits are slower on the female flowers because of the high amount of nectar and the nectary position.

5. Conclusion

Bees foraging zucchini flowers in Korhogo are *A. mellifera*, *L. interruptum* and *B. impatiens*. Of all these species encountered, *A. mellifera* was the most active on male flowers and female flowers. The peak activity of these species was between 5 am and 6 am. The activity of bees on zucchini flowers is influenced by the type of flowers and abiotic factors. Bees are more active on female flowers than on male flowers. The temperature positively influences the activity of the bees while it is inversely proportional to the fluctuation of the relative humidity.

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