Pollination Efficiency of *Dactylurina Staudingeri* (Hymenoptera : Apidae) on *Vernonia Amygdalina* (Asteraceae) Florets at Dang (Ngaoundéré, Cameroon)

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Abstract: *To evaluate the impact of Dactylurina staudingeri on fruit and seed yields of Vernonia amygdalina, its foraging and pollination activities were studied in Ngaoundéré for two seasons, December 2016 - January 2017 and December 2017 - January 2018. Observations were made on 540 capitula divided in four treatments : two differentiated according to the presence or absence of capitula protection regarding insect visits ; the third protected and uncovered when florets were opened, to allow D. staudingeri visits and the fourth with capitula uncovered then rebagged without any visit. Bee's daily rhythm of activity, its foraging behaviour on florets and its pollination activity were evaluated. Results showed that, on V. amygdalina florets, D. staudingeri intensely collected pollen only. The mean foraging speed was 18.48 florets per minute. Data obtained allow the classification of this Asteraceae as a highly polliniferous stingless bee plant. Through its pollination efficiency on V. amygdalina, D. staudingeri has increased the fruiting rate by 23.12 % and 16.34 %, and the percentage of normal achenes by 32.79 % and 33 %, respectively in 2017 and 2018. Hence, conservation of D. staudingeri colonies close to V. amygdalina fields is recommended to improve its fruit production.*

Keywords: *Dactylurina staudingeri, Vernonia amygdalina, florets, pollination efficiency, yields.*

1. INTRODUCTION

Pollination, the transfer of pollen grains to the stigma of the plant gynoecium is a crucial step in the sexual reproduction of flowering plants [1]. These last exhibit enormous variations in their mode of pollination [2]. The most common way plants attract animals to visit their flowers is by providing food such as nectar, pollen or oils [1]. *Vernonia amygdalina* (Asteraceae), commonly called « bitter leaf » is a shrub with petiolate green leaf of about 6 mm diameter and elliptic shape [3]. It has been used in Nigeria [4] as an ingredient to prepare Nigerian (ogbono soup) or Cameroon (ndolè) dish after reduction of its bitter taste through soaking several time in water or by boiling [5]. The leaves have found relevance in traditional folk medicine as anthelmintic, antimalarial and laxative [3].

Meliponi culture is the practice of keeping stingless bees [6, 7]. These insects have been known for their honey and pollen production; their role as the providers of ecosystem services is also recognized [6, 8]. They are important for the pollination of many flowering plants in tropical ecosystems [8, 9]. *Dactylurina staudingeri* is a stingless bee originated from Ethiopia [10]. It is also found at Makokou and Belinga, a village near Cameroon and middle Congo [11]. It is the only stingless bee found in Cameroon with exposed nests who are often fixed on tree branches at a height above 4 m [12]. This social bee produces honey with a quality different to that of honeybee [10, 11]. It is known to visit some plant species such as *Zea mays* at Yaoundé [13], *Endosiphon prumiloides* at Campo [14], *Jatropha* sp. at Kinshasa [11], *Cucumeropsis manii* at Yaoundé [15] and *Vigna unguiculata* in Ghana

[16]. None of this research examines in detail the foraging activity or the pollinating efficiency of *D. staudingeri*.

The study of the foraging activity of *Apis mellifera* on the bitter leaf flowers have been made at Ngaoundéré by [17]. The honeybee, although recognized as the best pollinator [1], has for some years been confronted with natural or man-made threats [18, 19] thus endangering this species. With the growing pressure on the environment, and the associated loss of honey bees, there is the need for additional pollinator species to be used in agriculture to maintain resilience in food production and yields improvement [20]. For this, the stingless bees have proved to be an alternative [21, 22].

The general objective of this work is to contribute to the understanding of the relationships between *V. amygdalina* and *D. staudingeri*, for their optimal management. Specific objectives are to : determine the place of *D. staudingeri* in the *V. amygdalina* floral entomofauna ; study the activity of this stingless bee on florets of this Asteraceae ; evaluate the impact of the flowering insects including *D. staudingeri* on pollination and seed production of the bitter leaf ; estimate the meliponicultural value of *V. amygdalina* ; determine the pollination efficiency of *D. staudingeri* on *V. amygdalina*.

2. MATERIAL AND METHODS

2.1. Material

2.1.1. Study Site and Biological Material

The experiment was carried out from November 2016 to January 2017 and from November 2017 to January 2018 at Dang, during the flowering periods of *V. amygdalina*, within the experimental fields of the Unit for Apply Apidology (latitude: 7°42.264 N; longitude: 13°53.945 E; altitude: 1106 m a. s. 1.) of the Faculty of Science, University of Ngaoundéré. This region belongs to the high altitude Guinean savannah agro-ecological zone [23]. The climate is characterized by a rainy season (April to October) and a dry season (November to March), with an annual rainfall of about 1500 mm. The mean annual temperature is 22 °C, while the mean annual relative humidity is 70 % [24]. The vegetation was represented by crops, ornamental plants, hedge plants and native plants of savannah and gallery forests. The plant material was represented by *V. amygdalina* cultivated in the University Campus. The *D. staudingeri* individuals of the experimental station came from the nests located at the bee house of the Unit for Apply Apidology.

2.2. Methods

2.2.1. Determination of the Place of Dactylurina Staudingeri in the Floral Entomofauna of Vernonia Amygdalina

On 19th December 2016, 240 *V. amygdalina* capitula from ten plants with florets at bud stage were labelled (24 capitula per plant) among which 120 were left unattended (treatment 1) and 120 were protected using gauze bags net to prevent insect visitors (treatment 2) [25]. Similarly, on 28th December 2017, 240 *V. amygdalina* capitula with florets at bud stage were labelled (24 capitula per plant) among which 120 were left unattended (treatment 5) and 120 were protected using gauze bags net to prevent insect visitors (treatment 6).

From 20th December 2016 to 8th January 2017 and from 29th December 2017 to 17th January 2018, observations were made every day on capitula of treatments 1 and 5, at 6 - 7 h, 8 - 9 h, 10 - 11 h, 12 - 13 h, 14 - 15 h and 16 - 17 h. For each of these time slots, the different insects encountered on the blooming florets were counted [26]. Cumulative results were expressed as the number of visits [27]. Data on the frequency of visits of the various identified flowering insects have made it possible to determine the place of *D. staudingeri* in the anthophilous entomofauna of *V. amygdalina*. The frequency of visits of the insect *i* on *V. amygdalina* florets (Fi) was calculated using the following formula: $Fi = \{[Vi \ / \ Vt] * 100\}$, where Vi is the number of visits for insect *i* on unprotected capitula and Vt the number of visits of all insects on the same capitula [26].

Specimens of all insect taxa, excluded *Apis mellifera* and *D. staudingeri* were caught using insect net on unlabeled capitula and conserved in 70 % ethanol, excluding butterflies that were preserved dry [28], for subsequent taxonomic identification.

2.2.2. Study of the Activity of Dactylurina Staudingeri on the Florets of Vernonia Amygdalina

In addition to the determination of the flower visiting insect frequency, direct observation of the foraging activity of *D. staudingeri* on florets was made in the field. The floral products (nectar or pollen) sampled by this bee were noted during the same dates and time slots as for the duration of visits, based on its foraging behavior. Nectar foragers were expected to extend their proboscis to the base of the corolla and the stigma, while pollen gatherers were supposed to scratch the anthers with mandibles and legs [28].

The duration of visits per floret was recorded during the following daily time frames: 7 - 8 h, 9 - 10 h, 11 - 12 h, 13 - 14 h, 15 - 16 h and 17 - 18 h. The abundances of foragers (highest number of individuals simultaneously in activity per floret, per capitula and per 1000 florets) [27] and the foraging speed (number of florets visited per minute) [29] were registered at the same dates and daily time frames as for the duration of visits. The abundances of foragers per floret and per capitula were noted following the direct counting. For the abundance per 1000 florets (A_{1000}), the number of individuals of *D.staudingeri* was counted on a known number of florets at the moment *x*. The abundance per 1000 florets was then calculated using the formula $A_{1000} = [(A_x / F_x)*1000]$, where F_x and A_x are respectively the number of opened florets and the number of foragers effectively counted on these florets at time *x* [26]. The foraging speed (*Fs*) was calculated using the formula $Fs = (Fl / du)*60$, where *du* is the duration (sec) given by a stopwatch, and *Fl*, the number of florets visited during *du* [26]. During each daily period of investigation, a mobile thermo-hygrometer installed in the shade, was used to register the temperature and the relative humidity of the station every 30 min, from 6 h to 18 h, during the entire flowering period [26].

2.2.3. Evaluation of the Meliponicultural Value of Vernonia Amygdalina

The meliponicultural value of *V. amygdalina* was assessed using data on its flowering intensity and the degree of attractiveness of *D. staudingeri* foragers with respect to its nectar and pollen.

2.2.4. Evaluation of the Effect of Insects Including Dactylurina Staudingeri on Vernonia Amygdalina Production

In parallel to the constitution of treatments 1, 2, 5 and 6, 600 capitula with florets at bud stage were protected in 2016 and 2017, to form two treatments:

- Treatment 3 in 2016 or 7 in 2017: 400 capitula protected using gauze bag nets to prevent insect or any other organism visits and destined to be visited exclusively by *D. staudingeri*. As soon as the first florets were opened on each capitulum of treatments 3 and 7, the gauze bag was removed and this capitulum was observed for up to 10 minutes ; the capitula visited once by *D. staudingeri* was marked and then protected once more ;

- Treatment 4 in 2016 and 8 in 2017: 200 capitula protected using gauze bag nets and destined to be uncovered then rebagged without the visit of insects or any other organism. As soon as the first florets were opened on each capitulum of treatments 4 and 8, the gauze bag was removed and this capitulum was observed for up to 10 minutes, while avoid insect or any other organism visits.

At the maturity, achenes were harvested and counted from each treatment. The fruiting rate and the percentage of normal achenes were then determined for each treatment.

The evaluation of the effect of insects including *D. staudingeri* on *V. amygdalina* production was based on the impact of flowering insects on pollination, the impact of pollination on *V. amygdalina* fruiting, and the comparison of fruiting rate and percentage of normal (that is well developed) achenes of treatments 1, 2, 4, 5, 6 and 8. For each year, the fruiting rate due to the foraging insects including *D. staudingeri* (*Fri*) was calculated using the following formula : $Fri = \{[(FX + Eg) - FY / (FX + Eg)] *100\}$, where *FX* and *FY* are the fruiting rates in treatment *X* (capitula left in free pollination) and treatment *Y* (capitula protected from all insect visits), and *Eg* the effect of the gauze bag net who can be calculated using the formula $Eg = FY - FZ$, where *FZ* is the fruiting rate in treatment *Z* (capitula protected then unbagged and rebagged without insect or any other organism visit).

Finally, $Fri = \{[(FX - FZ) / (FX + FY - FZ)] *100\}$

The fruiting rate of a treatment (*F*) is $F = [(b/a)*100]$, where *b* is the number of achenes formed and the number of viable florets initially set [27].

The impact of flower visiting insects including *D. staudingeri* on normal achenes was evaluated using the same method as mentioned above for the fruiting rate.

2.2.5. Assessment of the Pollination Efficiency of Dactylurina Staudingeri on Vernonia Amygdalina

The contribution of *D. staudingeri* in the fruiting rate and the percentage of normal achenes were calculated using data of treatments 3 and 4 for 2016 and those of treatments 7 and 8 for 2017.

For each observation year, the contribution of *D. staudingeri* in the fruiting rate (*FrD*) was calculated using the following formula: $FrD = \{[(FD - FZ) / FD] * 100\}$, where *FD* is the fruiting rate in treatment *D* (capitula visited exclusively by *D. staudingeri*) [30].

The impact of *D. staudingeri* on normal achenes was evaluated using the same method as mentioned above for the fruiting rate.

2.2.6. Data Analysis

Data were subjected to descriptive statistics (means, standard deviations and percentages), Student's *t*-test for the comparison of the mean of two samples, Pearson correlation coefficient (*r*) for the study of association between two variables, chi-square ($\chi 2$) for the comparison of percentages and ANOVA (*F*) for the comparison of means of more than two samples using Microsoft Excel 2010.

3. RESULTS AND DISCUSSION

3.1. Place of *Dactylurina Staudingeri* in the Flower Entomofauna of *Vernonia Amygdalina*

Amongst the 2037 and 1772 visits of 25 and 27 insect species recorded on *V. amygdalina* capitula in 2016/2017 and 2017/2018 respectively, *D. staudingeri* ranked second accounting for 15.46 % and 15.86 % of all visits. The first place was occupied by *Apis mellifera* in the first (30.34 %) and the second (35.21 %) seasons respectively (Table 1). The weak frequency of the visit of *D. staudingeri* on *V. amygdalina* florets compare to that of *A. mellifera* could be explained by the strategies adopted by the honey bee that consist of recruiting a great number of workers for the exploitation of an interesting food source [31]. Besides, during our investigations, the number of honeybee colonies (76) in the study site was much higher than that of *D. staudingeri* (3).

Table1. *Insects registered on Vernonia amygdalina florets in 2017 and 2018, number and percentage of visits of different insects.*

	Insects		2017		2018		2017/ 2018	
Order	Family	Genus and species	*n1*	*P1* (%)	*n2*	*P1* (%)	*Nt*	*Pt* (%)
Coleoptera	Scarabeidae	(sp. 1) (ne)	22	1.08	5	0.28	27	0.71
		(sp. 3) (ne)	25	1.23	15	0.85	40	1.05
Diptera	Calliphoridae	*Calliphora* sp. (ne)0	44	2.16	7	0.40	51	1.33
	Muscidae	*Musca domestica* (ne, po)	38	1.86	12	0.68	50	1.31
Hymenoptera	Apidae	*Apis mellifera* (ne, po)	618	30.34	624	35.21	1242	32.60
		Braunsapis sp. (ne, po)	177	8.69	124	7	301	7.90
		Ceratina sp. (ne, po)	124	6.08	85	4.80	209	5.48
		Dactylurina staudingeri (po)	315	15.46	281	15.86	596	15.64
		Meliponula ferruginea (ne, po)	80	3.93	89	5.02	169	4.43
		Xylocopa olivacea (ne)	89	4.37	41	2.31	130	3.41
		Xylocopa inconstans (ne)	31	1.52	24	1.35	55	1.44
	Halictidae	*Crossisaspidia chandleri* (ne)	32	1.57	23	1.30	55	1.44
		Lipotriches notabilis (ne)	9	0.44	20	1.13	29	0.76
		(1sp.) (ne)	-	-	19	1.07	19	0.50
	Megachilidae	*Chalicodoma cincta* (ne)	71	3.48	32	1.81	103	2.70
		Chalicodoma rufipes (ne)	80	3.93	29	1.64	109	2.86
		Megachille torrida (ne)	32	1.57	13	0.73	45	1.18
		(1sp.) (ne)	12	0.59	23	1.30	35	0.92
	Sphecidae	*Philanthus triangulum* (ne, pr)	23	1.13	28	1.58	51	1.34
	Vespidae	*Belonogaster juncea* (ne)	81	3.98	90	5.08	171	4.49
Hemiptera	Pentatomidae	(sp.1) (ne)	14	0.69	-	-	14	0.36

	Pyrrhocoridae	*Dysdercus voelkeri* (ne)	30	1.47	10	0.56	40	1.05
Lepidoptera	Acraeidae	*Acraea acerata* (ne)	28	1.37	39	2.20	67	1.76
	Nymphalidae	*Hypolimnas misippus* (ne)	31	1.52	50	2.82	81	2.12
	Papilionidae	*Papilio demodocus* (ne)	10	0.49	11	0.62	21	0.55
		Graphium pylades (ne)	21	1.03	32	1.81	53	1.39
	Pieridae	*Catopsilia florella* (ne)	-	-	16	0.90	16	0.42
TOTAL		**27 species**	**2037**	**100**	**1772**	**100**	**3809**	**100**

n1: number of visits on 1648 florets in 19 days ; n2 : number of visits on 1559 florets in 20 days ; nt : total number of visits ; sp. : undetermined species ; P1, P2, Pt : percentages of visits :

P1 = (n1/2037)*100; P2 = (n2/1772)*100; Pt = (nt /3809)*100 ; ne : nectar collection ; po: pollen collection ; pr : predator of Apis mellifera.

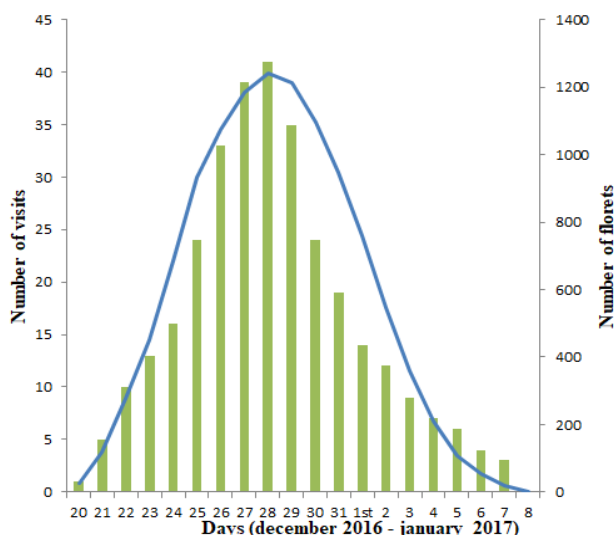### 3.2. Activity of *Dactylurina staudingeri* on *Vernonia amygdalina* florets

On *V. amygdalina* florets, individuals of *D. staudingeri* were seen intensively collected the pollen (Figure 1). No nectar harvest visit was observed.

Figure1. *Dactylurina staudingeri collecting pollen on a floret of Vernonia amygdalina*

The strong attractivity of the pollen of this Asteraceae with respect to *D. staudingeri* could be partly explained by the availability and the quality of this food as well as the best time to harvest it at the level of the florets [31].

Dactylurina staudingeri visits were numerous on *V. amygdalina* plants when the number of opened florets was highest (Figure 2). Furthermore, we found a positive and highly significant correlation between the number of *D. staudingeri* visits and the number of *V. amygdalina* opened florets in 2016/2017 ($r = 0.95$; $df = 15$; $P < 0.01$) (Figure 2, A) as well as in 2017/2018 ($r = 0.45$; $df = 21$; $P < 0.05$) (Figure 2, B).
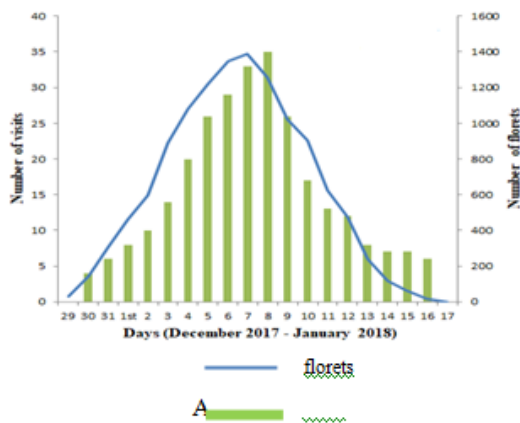
Figure2. *Seasonal variation of the number of Vernonia amygdalina opened florets and the number of Dactylurina staudingeri visits on these organs in 2016/2017 (A) and 2017/2018 (B) at Dang*

Dactylurina staudingeri was active on *V. amygdalina* florets from 6 h to 17 h in 2016/2017 and 2017/2018, with a peak of visits between 12 h and 13 h (Figure 3). This daily period probably correspond to that of the highest availability of pollen on florets of this Asteraceae. Indeed, the period of daily activity of many flowering insects on a given plant species depends on the availability of pollen [32] or nectar [33] in its flowers.
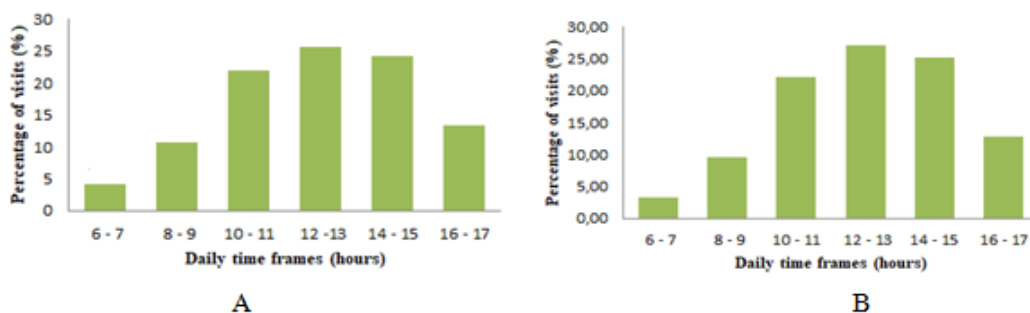

Figure3. *Variation of the number of Dactylurina staudingeri visits on Vernonia amygdalina capitula according to the daily time frames in 2016/2017 (A) and 2017/2018 (B) at dang*

This activity could also be conditioned by some climatic factors [32]. In 2016/2017, the correlation was not significant between the number of *D. staudingeri* visits and the temperature ($r = 0.81$; $df = 4$; $P > 0.05$) and between the same number of visits and the relative humidity ($r = - 0.83$; $df = 4$; $P > 0.05$) (Figure 4, A). Equally, in 2017/2018, the correlation was not significant between the number of *D. staudingeri* visits and the temperature ($r = 0.90$; $df = 4$; $P > 0.05$) and between the same number of visits and the relative humidity ($r = - 0.94$; $df = 4$; $P > 0.05$) (Figure 4, B).
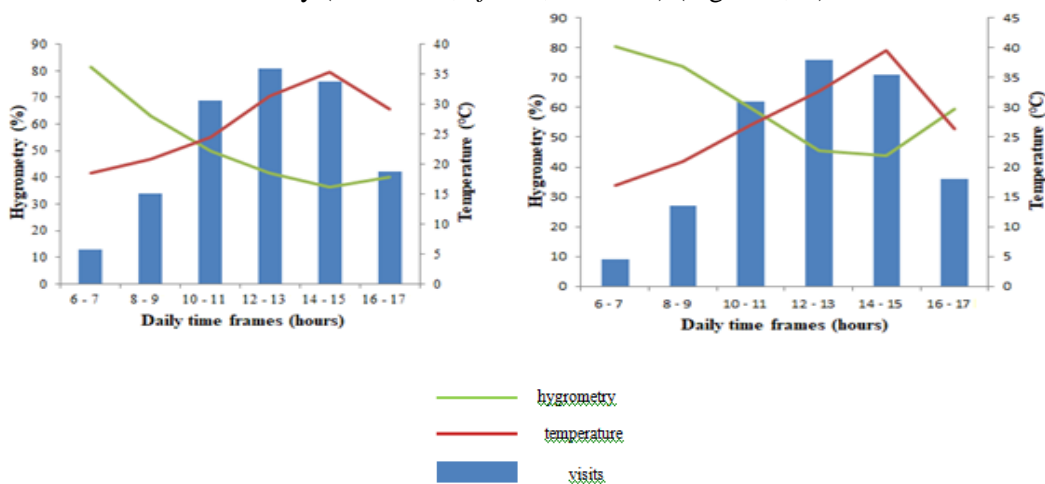

Figure4. *Variations of the temperature, the hygrometry and the number of Dactylurina staudingeri visits on the florets of Vernonia amygdalina according to the daily time frames in 2016/2017 (A) and 2017/2018 (B) at Dang.*

In 2016/2017, the highest mean number of *D. staudingeri* individuals simultaneous in activity was 1/floret ($n = 96$; $s = 0$), 1.33 per capitula ($n = 70$; $s = 0.47$; $min = 1$; $max = 2$) and 129.27 per 1000 florets ($n = 79$; $s = 47.79$; $min = 60$; $max = 300$). In 2017/2018, the corresponding figures were 1/floret ($n = 91$; $s = 0$), 1.23 per capitula ($n = 62$; $s = 0.42$; $min = 1$; $max = 2$) and 118.73 per 1000 florets ($n = 86$; $s = 29.13$; $min = 71$; $max = 205$). For the abundance per 1000 florets, the difference between the two means (129.27 and 118.73) was highly significant ($t = 11.00$; $df = 163$; $P < 0.001$). For the two cumulated years the mean number of individuals of *D. staudingeri* was 1.28 per capitula and 124 per 1000 florets.

The high abundance of *D. staudingeri* individuals per 1000 florets, and the positive and significant correlation between the number of *V. amygdalina* florets and the number of *D. staudingeri* visits, highlight the attractiveness of *V. amygdalina* pollen for *D. staudingeri*. This could be explained by the availability and the accessibility of this floral product. As has been pointed out for *Trigona iridipennis*, *D. staudingeri* could be able to recruit a great number of workers for the exploitation of a good food source [34].

The mean duration of *D. staudingeri* visit per *V. amygdalina* floret was 3.97 sec ($n = 61$; $s = 1.87$) in 2016/2017 and 4 sec ($n = 53$; $s = 2.27$) in 2017/2018 respectively. The difference between these two means is not significant ($t = 0.4$; $df = 112$; $P > 0.05$). For the two cumulated years, the mean duration of a flower visit was 3.98 sec.

In the *V. amygdalina* field, an individual of *D. staudingeri* visited between 9 and 48 florets per minute in 2016/2017 and between 9 and 27 florets per minute in 2017/2018. The mean foraging speed was 19.57 florets per minute ($n = 63$; $s = 6.21$; $min = 9.47$; $max = 48$) in 2016/2017 and 17.40 florets per minute ($n = 71$; $s = 7.79$; $min = 8.57$; $max = 27.27$) in 2017/2018. The difference between these two means is highly significant ($t = 10.13$; $df = 132$; $P < 0.001$). This difference would be explain by the accessibility and availability of pollen or the distance separating the flowers visited during the various foraging trips. For the two cumulated years, the mean foraging speed was 18.48 florets per minute.

3.3. Meliponicultural Value of *Vernonia Amygdalina*

During the flowering period of *V. amygdalina*, we noted a well elaborated activity of *D. staudingeri* foragers at the level of its flowers: high abundance of workers per 1000 florets, good daily and seasonal frequency of visits, good pollen harvest and fidelity of the foragers to the florets during foraging bouts. These data highlight the good attractiveness of the pollen and the unattractiveness of the nectar of this plant for *D. staudingeri*. Therefore, *V. amygdalina* is a highly polliniferous stingless bee plant. This Asteraceae could thus be grown to help stabilize *D. staudingeri* colonies during the dry season and increase pollen production as a stingless bee hive product.

3.4. Impact of Flowering Insects Including *Dactylurina Staudingeri* on *Vernonia Amygdalina* Production

During nectar or pollen harvest on *V. amygdalina*, foraging insects always shook florets and regularly contacted anthers and stigma, increasing self - pollination and/or cross - pollination possibilities of this plant species.

Table 2 gives the fruiting rate and the percentage of normal achenes in the different treatments of *V. amygdalina*. It appears from this table that :

- The fruiting rates were 85.62 %, 37.60 %, 68.82 %, 52.91 %, 82.49 %, 38.91 %, 62.43 % and 52.23 % in treatments 1 to 8 respectively. The differences between these eight percentages are highly significant ($\chi^2 = 1507.51$; $df = 7$; $P < 0.001$). Two - to - two comparisons showed that the difference observed is highly significant between treatments 1 and 2 ($\chi^2 = 791.29$; $df = 1$; $P < 0.001$) as well as between treatments 5 and 6 ($\chi^2 = 592.58$; $df = 1$; $P < 0.001$). Consequently, in 2016/2017 and 2017/2018, the fruiting rate of exposed capitula (treatments 1 and 5) was higher than that of capitula bagged during their flowering period (treatments 2 and 6).

- The percentages of normal achenes were 66.05 %, 44.13 %, 51.79 %, 34.81 %, 67.57 %, 46.86 %, 56.08 % and 37.57 % in treatments 1 to 8 respectively. The differences between these eight percentages are highly significant ($\chi^2 = 4438.37$; $df = 7$; $P < 0.001$). Two - to - two

comparisons showed that the difference observed was highly significant between treatments 1 and 2 (χ^2 = 83.61 ; *df* = 1 ; *P* < 0.001) as well as between treatments 5 and 6 (χ^2 = 69.03 ; *df* = 1 ; *P* < 0.001). Hence, in 2016/2017 and 2017/2018, the percentage of normal achenes from exposed capitula was higher than that from capitula bagged during their flowering period.

In 2016/2017, the numeric contribution of the anthophilous insects including *D. staudingeri* in the fruiting rate and the percentage of normal achenes of *V. amygdalina* were 46.52 % and 41.44 % respectively. In 2017/2018, the corresponding figures were 43.75 % and 39.03 % respectively. For the two cumulate years, the numeric contributions of flowering insects were 45.13 % and 40.23 % for the fruiting rate and the percentage of normal achenes respectively.

Table2. *Fruiting rate and percentage of normal achenes according to different treatments of Vernonia amygdalina in 2016/2017 and 2017/2018 at Dang*

Treatments	Years	NC	NFS	TNA	FR (%)	NNA	% NA
1 (Uc)	2016/2017	120	1648	1411	85.62	932	66.05
2 (Pc)		119	1585	596	37.60	263	44.13
3 (Bcvd)		188	2399	1651	68.82	855	51.79
4 (Bcwv)		91	1064	563	52.91	196	34.81
5 (Uc)	2017/2018	118	1559	1286	82.49	869	67.57
6 (Pc)		117	1393	542	38.91	254	46.86
7 (Bcvd)		138	1725	1077	62.43	604	56.08
8 (Bcwv)		87	1055	551	52.23	207	37.57

Uc : unprotected capitula ; Pc : protected capitula ; Bcvb : capitula bagged then uncovered and exclusively visited by Dactylurina staudingeri ; Bcwv : capitula bagged, then uncovered and rebagged without insect or any other organism visit ; NC : number of capitula ; NFS : number of florets studies ; TNA : total number of achenes ; FR : fruiting rate ; NNA : number of normal achenes ; % NA : percentage of normal achenes.

3.5. Pollination Efficiency of *Dactylurina Staudingeri* on *Vernonia Amygdalina*

During pollen harvest on the bitter leaf florets, workers of *D. staudingeri* always came into contact with anthers and stigma, increasing the possibilities of *V. amygdalina* pollination. With this pollen, they flew frequently from floret to floret. The percentage of the total number of visits during which forager bees came into contact with the stigma of the visited florets was   90 % in 2016/2017, 95 % in 2017/2018 and 92.5 % for the two cumulated seasons.

The fruiting rate due to *D. staudingeri* was 68.82 % in 2016/2017, 62.43 % in 2017/2018 and 65.62 % for the two cumulate years. The difference was highly significant between treatments 3 and 4 (χ^2 = 80.88 ; *df* = 1 ; *P* < 0.001) as well as between treatments 7 and 8 (χ^2 = 28.11 ; *df* = 1 ; *P* < 0.001). Hence, in 2016/2017 and 2017/2018, the fruiting rate of capitula protected and visited exclusively by *D. staudingeri* was higher than that of capitula protected, uncovered and re-protected without insect or any other organism visit.

The percentage of normal achenes due to *D. staudingeri* was 51.79 % in 2016/2017, 56.08 % in 2017/2018 and 53.93 % for the two cumulate years. The difference was highly significant between treatments 3 and 4 (χ^2 = 48.50 ; *df* = 1 ; *P* < 0.001) as well as between treatments 7 and 8 (χ^2 = 49.98 ; *df* = 1 ; *P* < 0.001). These results pointed out that capitula visited by *D. staudingeri* have the highest number of normal achenes compare to those protected then uncovered and rebagged without the visit of insect or any other organism.

In 2016/2017, the numeric contribution of *D. staudingeri* via a single floret visit were 23.11 % for the fruiting rate and 32.78 % for the percentage of normal achenes. In 2017/2018, the corresponding figures were 16.34 % and 33.01 % respectively. For the two cumulate years, the numeric contribution of *D. staudingeri* on the fruiting rate and the percentage of normal achenes were 19.72 % and 32.89 % respectively.

4. CONCLUSION

From our observations, *Vernonia amygdalina* is a plant species that highly benefits from pollination by insects among which *Dactylurina staudingeri* is one of the most important and harvested pollen. The comparison of achenes production of capitula visited exclusively by *D. staudingeri* with that of the capitula protected from insects then uncovered and reprotected without the visit of insect or any

other organism underscores the capacity of this stingless bee in increasing fruits production as well as achenes quality. Data on the foraging activity of *D. staudingeri* enable the classification of *V. amygdalina* among the highly polliniferous stingless bee plant. Thus, *V. amygdalina* plant could be grown or protected to increase pollen production as a stingless bee hive product and stabilize *D. staudingeri* colonies during the dry season.

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