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Combined effects of *Chalicodoma rufipes* (Hymenoptera: Megachilidae) activity and compost on pollination and production of *Phaseolus vulgaris* pink variety (Fabaceae) at Dang (Ngaoundéré, Cameroon)

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Abstract

To evaluate synergistic effect of compost and *Chalicodoma rufipes* on *Phaseolus vulgaris* pink variety production, during the 2018 and 2021 cropping seasons, experiments were carried out in a complete randomized block design with three treatments: subplots applied with compost; subplots applied with NPK-fertilizer; subplots applied neither with compost, nor with NPK. Other four treatments were made up of 1220 labelled flowers comprising two differentiated according to the presence or absence of protection of flowers from insect visits; the third with flowers protected and uncovered when flowers were opened, to anable *C. rufipes* visit and the fourth with flowers uncovered and reprotected from insect or any other organism visits. *Chalicodoma rufipes*'s daily rhythm of activity, its foraging behavior on flowers and its pollination efficiency were evaluated. Results reveal that compost significantly increased (P < 0.001) *P. vulgaris* plants biomass. Among the 13 insect species recorded on *P. vulgaris* flowers, *C. rufipes* ranked second with 20.84% of the 782 visits. *Chalicodoma rufipes* intensely harvested nectar. The combined effet of *C. rufipes* and compost notably increased the fruiting rate, the number of seeds per pod and the percentage of normal seeds by 49.4%, 13.8% and 26.63%, respectively. Hence, application of compost and preservation of *C. rufipes* nests close to *P. vulgaris* fields is recommended to improve production of this important crop legume in the region.

Keywords: *Chalicodoma rufipes*; *Phaseolus vulgaris*; Pollination efficiency; Compost; Production

1. Introduction

Phaseolus vulgaris or common beans is an annual plant originated from South and Central America [1]. Worlwidely, it is ranked 10th among grown vegetables and first among legumes consumed as pulses (excluding soybean) ahead of peas, chickpeas and broad beans [2]. In Cameroon, *P. vulgaris* is cultivated as vegetable and can be consumed raw or cooked; pods are sold fresh (green beans) or transformed into flour, while stems and leaves are used to feed livestock [3]. The color of flower can vary from pink, white to purple depending on different varieties [3], while flowers produce nectar and pollen that attract insects [4, 5]. *P. vulgaris* pods contain 1 to 12 seeds [2], and its flowers were reported to produce fewer seeds per pod in the absence of efficient pollinators in the United States of America [6]. In Maroua a study conducted by Douka and Tchuenguem [5] revealed that *Apis mellifera* visits *P. vulgaris* (Red and Small seeds variety) flowers for nectar and pollen collection and increase the fruiting rate by 55.32%, the number of seeds/pod by 19.10% and the percentage of normal seeds by 7.71%. In Chad study by Mainkete *et al.* [7] pointed out that *X. olivacea* provoke a significant increase of the fruiting rate (52.27%), the number of seeds/pod (30.79%), as well as the percentage of normal seeds (84.03%) of *P. vulgaris* (Large White Seeds variety). Cross-pollination of *P. vulgaris* by insects has generally been observed in such an autogamous/allogamous plant [5, 6, 7].

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Agricultural soil degradation has become a significant global problem [8, 9], with a negative impact on crops growth and yields, but can be permanently alleviated by effective and long-term regeneration of soil fertility through improved soil structure, or increased organic matter rate [10]. It is expected that organic soil amendments could be cheaper and more beneficial for maximizing crop yield in the context of the high cost of mineral fertilizers [11]. Studies on the synergistic effects of compost and pollinating insects on growth and crops production at Dang has been highlighted by Mohamadou *et al.* [12], Djakbé *et al.* [13], respectively on *Glycine max* and *Physalis minima*. No previous research has been reported on the relationships between *P. vulgaris* pink variety, *C. rufipes* and compost, although the activity and diversity of flowering insects of a plant species may vary with varieties [4]. The general objective of this work was to contribute to the understanding of the relationships between *P. vulgaris* (pink variety), *C. rufipes* and compost, for their optimal management. The following specific objectives were persued: (a) evaluate the impact of the compost on nodulation and biomass of *P. vulgaris*; (b) determine the place of *C. rufipes* in the *P. vulgaris* floral entomofauna; (c) study the activity of this Megachilidae on this Fabaceae flowers; (d) evaluate the impact of one *C. rufipes* visit on pollination, pods and seeds production of *P. vulgaris;* (e) evaluate the impact of compost on *P. vulgaris* production; (f) evaluate the combined effect of compost and one flower visit of *C. rufipes* on production of this plant species.

2. Material and methods

2.1. Experimental site description and biological material

The experiment was carried out from August to October 2018 and from August to October 2021 at Dang, during the *Phaseolus vulgaris* flowering periods, within the experimental fields of the Unit for Applied Apidology (latitude: 7°42.264 N; longitude: 13°53.945 E; altitude: 1106 m a. s. 1.) of the Faculty of Science, University of Ngaoundere, in the Adamaoua region of Cameroon. This region belongs to the high altitude Guinean savannah agro-ecological high altitude zone [14]. 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 % [15]. The vegetation near the *P. vulgaris* field has various unmanaged and cultivated species. The experimental plant material was represented by *P. vulgaris* pink variety. *P. vulgaris* seeds used originated from the Unit for Applied Apidology. *C. rufipes* individuals visiting the experimental station were from the natural population nested in the vicinity of the fields.

2.2. Sowing and weeding

From August to September 2018 and 2021, experimental plots were delimited, ploughed and divided into nine subplots, each measuring 42 m^2. Three subplots were applied with compost (T_C), three with chemical NPK-fertilizer (T_{NPK}) and three other left applied neither with compost, nor with NPK-fertilizer (control subplot (T_0)). Three seeds were sown per hole on 7 lines per subplot, for a total of 8 holes per line. Holes were separated 50 cm from each other, while lines were 75 cm apart. Weeding was performed manually as necessary to maintain plots weeds-free. Compost was produced in the Composting Unit of the Faculty of Science of the University of Ngaoundere. Approximately 1000 g of compost were applied per hole, as a layer before sowing. The NPK-fertilizer used was the commonly used formula 20:10:10 by growers, purchased from a local phytosanitary store. It was applied 10 days after sowing, at a rate of 10 g within the rhizosphere of each plantlet.

2.3. Determination of the reproduction mode of *Phaseolus vulgaris*

On September 20th 2018 in control subplots, 240 common beans flowers at bud stage were labelled (15 plants per subplot) among which 120 were left unattended (treatment 1), while 120 were protected using gauze bags net to prevent insect visitors (treatment 2) [16]. In similar subplots, on September 22nd 2021, *P. vulgaris* with flowers at bud stage were labelled (15 plants per subplot) among which 120 were left unattended (treatment 3), whereas 120 were protected using gauze bags net to prevent insect visitors (treatment 4). At the end of each cropping season, the number of fruits formed in each treatment was assessed at harvest. For each treatment, the podding index (*Pi*) was then calculated as described by Tchuenguem *et al*. [17] (2009a): $Pi = FB / FA$, where *FB* is the number of fruits formed and *FA* the number of viable flowers initially set. The allogamy rate (*Alr*) from which derives the autogamy rate (*Atr*) was expressed as the difference in podding indexes between treatment with unprotected flowers (treatment 1 or 3) and treatment with protected flowers (treatment 2 or 4): $Alr = [(PiA - PiB) / PiA] * 100$, where *PiA* and *PiB* are respectively the podding indexes in unprotected flowers (treatments 1 or 3) and in protected flowers (treatments 2 or 4). $Atr = 100 - Alr$.

2.4. Evaluation of nodulation and biomass of *Phaseolus vulgaris* in response to inoculation

At 60 days after planting (DAP), 15 plants per subplot (45 plants per treatment) were randomly selected and their nodules were harvested, counted, and stored in labeled envelopes before they were sun-dried and weighed [18]. Plants samples wrapped in newspapers, were also dried in a KA-1000 brand oven at 72 °C for 12 hours and weighed using a Sartorus brand electronic scale (accuracy 0.01). The collection of nodules from different plants was carried out according to the techniques recommended by Vincent (1970) [19] and Somasegaran & Hoben (1994) [20]. This involved digging at 15 cm around the plant and at 20 cm depth to extract the plant and its root system, removing soil by hand at the root level, without damaging the nodules, then washing nodules in tap water before counting.

2.5. Determination of the place of *Chalicodoma rufipes* in the *Phaseolus vulgaris* floral entomofauna

From 23rd September to 9th October 2018 and from 26th September to 12nd Octobrer 2021, observations were made every day on flowers of treatments 1 and 3, respectively, between 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, different insects encountered on the blooming flowers were counted [21]. Cumulative results were expressed as the number of visits [22]. Data on the visits frequency of various identified flowering insects were used to determine the place of *C. rufipes* in the anthophilous entomofauna of *P. vulgaris*. The visits frequency of insect *i* on *P. vulgaris* flowers (Fi) was calculated using the following formula: $Fi = \{[Vi / Vt] * 100\}$, where Vi is the number of visits for insect *i* on unprotected flowers and Vt the number of visits of all insects on the same flowers [21]. Specimens of all insect taxa, excluding *Apis mellifera* and *C. rufipes* were caught using insect net on unlabeled flowers and conserved in 70 % ethanol, excluding butterflies that were preserved dry [23, 24], for subsequent taxonomic identification.

2.6. Study of the activity of *Chalicodoma rufipes* on *Phaseolus vulgaris* flowers

In each subplot (T_C, T_{NPK} and T_0), in addition to the determination of the flower visiting insect frequency, direct observation of the foraging activity of *Chalicodoma rufipes* on flowers was separately made in the experimental field. Floral products (nectar or pollen) harvested by *C. rufipes* during each flower visit were registered based on its foraging behavior. Nectar foragers were expected to extend their proboscis to the base of the corolla and the stigma, while pollen gathered were scratched from anthers using their mandibles and legs [23, 24].

In the morning of each sampling day, the number of opened flowers labelled was counted. Whereas the visits frequency, the duration of individual flower visits were recorded (using a stopwatch) at least six times: 7-8 h, 9-10 h, 11-12 h, 13-14 h, 15-16 h and 17-18 h. Moreover, the number of pollinating visits which is defined as the visits with contact between the bees and stigma [25], the abundance of foragers (highest number of individuals foraging simultaneously per flower and per 1000 flowers) [26] and the foraging speed (number of flowers visited by individual bee per minute) [25] were recorded during the same dates and daily periods as for the registration of the visits duration.

The abundance of foragers per flower was recorded following the direct counting. For the abundance per 1000 flowers (C_{1000}), the number of *C. rufipes* individuals was counted on a known number of flowers at time *x*. The abundance per 1000 flowers was calculated using formula: $C_{1000} = [(Ax / Fx) * 1000]$, where Fx and Ax are respectively the number of flourished flowers and the number of *C. rufipes* individuals counted at time *x* [21].

The disruption of the activity of foragers by competitors or predators and the attractiveness exerted by other plant species on *C. rufipes* were assessed. During each daily period of investigation, a mobile thermo-hygrometer installed in the shade was used to register the temperature and the relative humidity at the site after every 30 min [21].

2.7. Assessment of the impact of compost on *Phaseolus vulgaris* production

On the compost subplots, 240 flowers were labeled and protected to form treatments 5 (2018) and 6 (2021) (like those of treatments 2 and 4). The comparison of production (fruiting rate, mean number of seeds per pod and percentage of normal seeds) of treatments 2 and 5 for the first year, 4 and 6 for the second year were assessed as influenced by compost on common bean plants.

For each observation year, the impact of compost on the fruiting rate (FrC) was calculated using the following formula: $FrC = \{[(FrA – FrB) / FrA] * 100\}$, where FrA and FrB are the fruiting rates in treatments 5 or 6 and in treatments 2 or 4, respectively.

The fruiting rate of a treatment (*Fr*) was expressed as: $Fr = [(b/a) * 100]$, where *b* is the number of fruits formed and *a* the number of viable flowers initially set [22]. The impact of compost on the mean number of seeds per pod and the percentage of normal seeds were evaluated using the same method as mentioned above for the fruiting rate.

2.8. Impact of Flowering Insects Including *Chalicodoma rufipes* on *Phaseolus vulgaris* production

Concimitantly to the constitution of treatments 1, 2, 3 and 4, 300 flowers at bud stage were protected in 2018 and 2021 in control subplots, to form four treatments:

- Treatment 7 in 2018 and treatment 8 in 2021: 200 flowers at bud stage were protected using gauze bag nets to prevent insect or any other organism visits and destined to be exclusively visited once by *C. rufipes*. When flowers were opened, the gauze bag was removed and flowers were observed for up to 10 minutes; flowers visited once by *C. rufipes* were marked and then reprotected;
- Treatment 9 in 2018 and 10 in 2021: 100 flowers protected using gauze bag nets and destined to be uncovered then rebagged without the visit of insects or any other organism. When the flowers were opened, the gauze bag was removed and flowers were observed for up to 10 minutes, while avoiding insect or any other organism visits and then reprotected.

For each investigation year, this evaluation was based on the impact of flowering insects on pollination, the impact of pollination on *P. vulgaris* fruiting, and the comparison of production (fruiting rate, mean number of seeds per pod and percentage of normal seeds) of treatment 1, 2, 3, 4, 9 and 10. For each year, the fruiting rate due to the foraging insects including *C. rufipes* (*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 (flowers left in free pollination) and treatment Y (flowers protected from all insect visits), and *Eg* the effect of the gauze bag net, calculated using the formula $Eg = FY - FZ$, where FZ is the fruiting rate in treatment Z (flowers protected then unbagged and rebagged without insect or any other organism visit).

Finally, $Fri = \{[(FX - FZ) / (FX + FY - FZ)] * 100\}$ [27].

The fruiting rate of a treatment (*Fr*) was calculated as: $Fr = [(b/a) * 100]$, where b is the number of achenes formed and the number of viable florets initially set [13].

The impact of flower visiting insects including *C. rufipes* on the mean number of seeds per pod and on the percentage of normal seeds was evaluated using the same method as mentioned above for the fruiting rate.

2.9. Assessment of the pollination efficiency of *Chalicodoma rufipes* on *Phaseolus vulgaris*

The contribution of *C. rufipes* in the fruiting rate, the mean number of seeds per pod and the percentage of normal seeds were calculated using data of treatments 7 and 9 for 2018 and those of treatments 8 and 10 for 2021. For each observation year, the contribution of *C. rufipes* in the fruiting rate (*FrCh*) was calculated using the following formula: $FrCh = \{[(Frz - Fry) / Frz] * 100\}$, where *Frz* and *Fry* are the fruiting rates in treatment 7 or 8 (flowers visited once by *C. rufipes*) and in treatments 9 or 10 (flowers bagged, uncovered and rebagged without visits of insect or another organism), respectively [13].

At maturity, pods were harvested from treatment 7, 8, 9 and 10 and the number of seeds per pod was counted. The mean number of seeds per pod and the percentage of normal seeds were then calculated for each treatment. The impact of *C. rufipes* on the number of seeds per pod and on the percentage of normal seeds was determined using the above method as mentioned for the fruiting rate.

2.10. Assessment of the cumulative action of *Chalicodoma rufipes* and compost on *Phaseolus vulgaris* production

As for treatment 7 and 8, on compost subplots 200 flowers at bud stage were protected using gauze bag nets in 2018 (treatment 11) and 2021 (treatment 12) to prevent insect or any other organism visits and destinated to be exclusively visited once by *C. rufipes*. To evaluate the impact of both compost and *C. rufipes* on *P. vulgaris* production, the comparison of production (fruiting rate, mean number of seeds per pod and percentage of normal seeds) between treatment 11 or 12 with those of treatment 9 or 10 was assessed.

The cumulative contribution of *C. rufipes* and compost on common bean fruiting rate, mean number of seeds per pod and percentage of normal seeds was calculated using data of treatments 9 and 11 for 2018 and 10 and 12 for 2021. For each observation year, cumulative contribution of *C. rufipes* and compost on the fruiting rate (FrD) was calculated using the following formula: $FrD = \{[(FrH - FrG) / FrH] * 100\}$, where FrH is the fruiting rate in treatments 11 or 12 and FrG the fruiting rate in treatments 9 or 10. The combined effects of *C. rufipes* and compost on the mean number of seeds per pod and the percentage of normal seeds were evaluated using the same method as mentioned above for the fruiting rate.

2.11. Data analysis

Data were treated using descriptive statistics (calculation of 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 relationship between two variables, chi-square ($\chi 2$) for the comparison of percentages, ANOVA (F) for the comparison of means of more than two samples and Microsoft Excel 2010 for plothing of graphics and histograms.

3. Results and discussion

3.1. Reproduction mode of *Phaseolus vulgaris*

The podding indexes were 0.88, 0.27, 0.85 and 0.26 in treatments 1, 2, 3 and 4, respectively. Allogamy rate (TC) was 69.32% and 69.41%, while autogamy rate (TA) was 30.68% and 30.59% respectively in 2028 and 2021. For the two cumulative years, TC was 69.37%, while TA was 30.63%. Consequently, the pink variety of *P. vulgaris* has a mixed allogamous-autogamous reproduction mode, with the predominance of allogamy over autogamy.

3.2. Influence of compost on number of nodules, nodule dry weight and biomass of *Phaseolus vulgaris*

Plants applied with compost at sowing produced a significantly greater number of nodules, nodule dry weight and plant biomass compared to plants applied with NPK and untreated plants in 2018 and 2021 (table 1).

Table 1 Variation of number of nodules, nodule dry weight and biomass of *Phaseolus vulgaris* affected by the application of compost at Dang in 2018 and 2021

Years	Subplots	Number of nodules per plant	Weight of dry nodules (g/plant)	Plant biomass (g)
2018	T_C	$(32.68 \pm 0.91)^a$	$(2.38 \pm 0.05)^a$	$(69.43 \pm 2.36)^a$
	T_{NPK}	$(10.37 \pm 0.91)^b$	$(0.64 \pm 0.05)^b$	$(46.69 \pm 2.36)^b$
	T_0	$(4.29 \pm 0.91)^c$	$(0.18 \pm 0.05)^c$	$(23.52 \pm 2.36)^c$
2021	T_C	$(41.46 \pm 1.09)^a$	$(2.89 \pm 0.04)^a$	$(71.82 \pm 2.24)^a$
	T_{NPK}	$(13.41 \pm 1.09)^b$	$(0.82 \pm 0.04)^b$	$(45.42 \pm 2.24)^b$
	T_0	$(5.33 \pm 1.09)^c$	$(0.21 \pm 0.04)^c$	$(24.35 \pm 1.12)^c$

T_C: subplot with compost; T_{NPK}: subplot with fertilizer-NPK; T_0: untreated subplot. In each column, means followed by the same letter are not significantly different at 5% level.

Similar findings were obtained from fields trials on *Glycine max* in 2018 and 2019 at Dang [12]. Compost was reported to improve plant growth and the diffusion of nutrients to plants through microbiological processes [28].

3.3. Place of *Chalicodoma rufipes* in the flower entomofauna of *Phaseolus vulgaris*

In the experimental field, out of the 392 and 390 visits of 11 and 11 insect species recorded on *P. vulgaris* flowers in 2018 and 2021 respectively, *C. rufipes* ranked second accounting for 21.86% and 20.23% of all visits (table 2). The first place was occupied by *Megachile cincta* in the first (22.64%) and the second (22.3%) seasons. Thus *C. rufipes* was one of the main floral visitor of *P. vulgaris* during the observation period. At Dang Kingha *et al.* [4] found *C. rufipes* to occupy the second rank with 15.82% and 14.65% amongst the 177 visits of 15 insect species in 2009 and 157 visits of 16 insects species in 2010, respectively recorded on *P. vulgaris* (black seed outlets) flowers.

The significant difference between the yearly percentage visits of *C. rufipes* could be ascribed to the presence of six nests of *C. rufipes* near the experimental plot in 2018 compared to that of two nests in 2021.

3.4. Activity of *Chalicodoma rufipes* on *Phaseolus vulgaris* flowers

On *P. vulgaris* flowers, *C. rufipes* were seen intensively collecting nectar on plants from compost, control and chemical NPK-fertilizer subplots (Figure 1). No pollen harvest was observed.

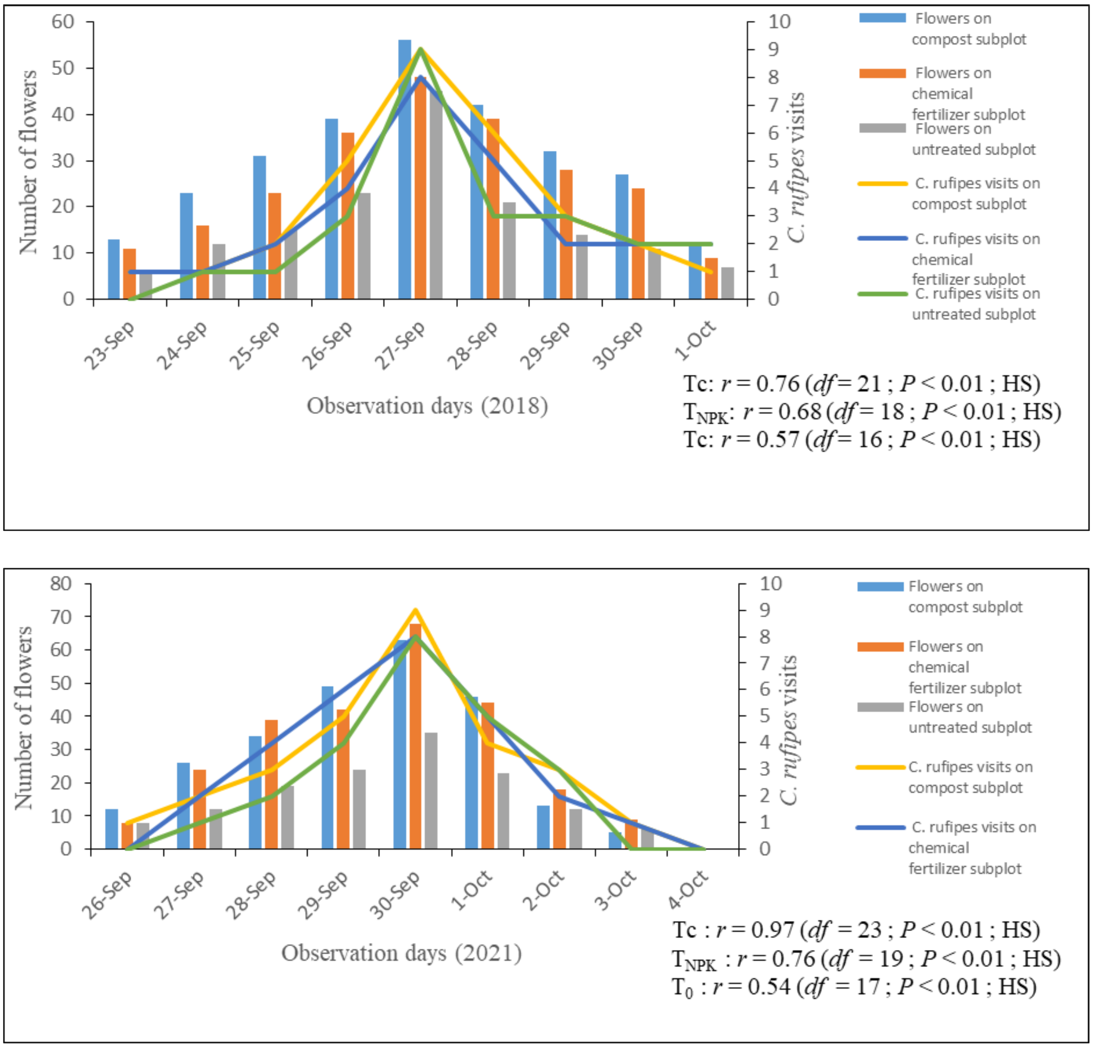
Figure 1 *Chalicodoma rufipes* collecting nectar in a flower of *Phaseolus vulgaris* at Dang in 2021

In 2018 as well as in 2021 and in each treatment (T_0, T_C and T_{NPK}), *C. rufipes* were numerous to visit *Phaseolus vulgaris* plants when the number of opened flowers was high. Furthermore, we found a positive and highly significant correlation between the number of *C. rufipes* visits and the number of *P. vulgaris* opened flowers in 2018 and 2021 (Figure 2).

Table 2 Diversity of insects visiting *Phaseolus vulgaris* flowers as influenced by compost and chemical fertilizer at Dang in 2018 and 2021, number and percentage of insect visits

Insects			2018					2021					2018/2021	
			Subplots			Total		Subplots			Total		Total	
Orders	**Family**	**Genus and species**	T_C	T_{NPK}	T_0	n_1	P_1 (%)	T_C	T_{NPK}	T_0	n_2	P_2 (%)	n_t	P_t (%)
Hymenoptera	Apidae	*Amegilla* sp. 1 (ne)	-	-	-	-	-	12	10	8	30	7.69	30	3.84
		Apis mellifera (ne and po)	24	24	15	63	16.07	27	23	23	73	18.72	136	17.43
		Ceratina sp. 1 (ne and po)	10	8	8	26	6.63	7	8	10	25	6.41	51	6.54
		Xylocopa inconstans (ne)	6	5	5	16	4.08	-	-	-	-	-	16	2.05
		Xylocopa olivacea (ne)	17	13	9	39	9.95	7	6	5	18	4.62	57	7.31
	Halictidae	*Crossisaspidia chandleri* (ne)	5	-	-	5	1.28	6	3	7	16	4.1	16	2.05
		Tyreus sp. (ne)	-	-	-	-	-	4	4	3	11	2.82	11	1.41
	Megachilidae	*Chalicodoma rufipes* (ne)	31	28	25	84	21.43	28	28	23	79	20.26	163	20.9
		Megachile cincta (ne)	36	31	22	89	22.7	31	29	27	87	22.31	179	22.95
		Megachile sp. 6 (ne)	4	2	2	8	2.04	8	8	6	22	5.64	30	3.85
	Vespidae	*Belonogaster juncea* (ne)	3	5	3	11	2.81	-	-	-	-	-	11	1.41
Total Hymenoptera			136	116	89	341	86.99	130	119	112	361	92.56	700	89.74
Lepidoptera	Hesperiidae	(1 sp.) (ne)	3	5	-	8	2.04	3	3	5	11	2.82	19	2.44
	Pieridae	*Eurema* sp. 1 (ne)	18	15	10	43	10.97	6	6	6	18	4.62	61	7.82
Total Lepidoptera			21	20	10	51	13.01	9	9	11	29	7.44	80	10.26
Total		13 espèces	157	136	99	392	100	139	128	123	390	100	780	100

T_C: subplot with compost; T_{NPK}: subplot with fertilizer-NPK; T_0: untreated subplot; *n1, n2, nt*: number of visits in 17, 18 and 35 days, respectively; sp.: Undetermined species; *P1, P2, Pt*: percentages of visits; *P1* = (*n1*/392)*100; *P2* = (*n2*/390)*100; *Pt* = (*n2*/780)*100; ne: collection of nectar; po: collection of pollen.



T_C: subplot with compost; T_{NPK}: NPK-fertilizer subplot; T_0: untreated subplot; *r:* correlation coefficient; *df*: degrees of freedom; *P:* level of significance; HS: highly significant.

Figure 2 Variations of the number of *Phaseolus vulgaris* opened flowers and the number of *Chalicodoma rufipes* visits on these organs according to the observation days in 2018 and 2021 at Dang

Chalicodoma rufipes was active on *Phaseolus vulgaris* flowers from 6.00 am to 5.00 pm in 2018 and in 2021, with a peak of visits between 10.00 am and 1.00 pm (Table 3). This daily period probably corresponds to that of the highest availability of nectar in flowers of this Fabaceae. Indeed, the daily period activity of many flowering insects on a given plant species depends on the availability of pollen [29] or nectar [30] in flowers. However, the decreased activity after 4.00 pm to 5.00 pm could be related to decreased temperature in the experimental field. Although, foragers preferred warm or sunny days for good floral activity [30], the enhanced temperature positively influenced the insect activity on foraged flowers.

Chalicodoma rufipes activity on *Phaseolus vulgaris* flowers was reported to be conditioned by some climatic factors [31]. In 2018, the correlation between the number of insect visits and the temperature was positive and significant on untreated ($r = 0.67$; $P < 0.05$), compost applied ($r = 0.78$; $P < 0.05$), and fertilizer-NPK applied ($r = 0.81$; $P < 0.05$) plants. The correlation between the relative humidity and the number of *C. rufipes* visits was not significant on untreated ($r = -0.33$; $P > 0.05$), compost applied ($r = -0.41$; $P > 0.05$), and chemical fertilizer applied ($r = -0.48$; $P > 0.05$) subplots. Equally, in 2021 the correlation between the number of insect visits and the temperature was positive and significant on untreated ($r = 0.89$; $P < 0.05$), compost applied ($r = 0.93$; $P < 0.05$), and fertilizer-NPK applied ($r = 0.82$; $P < 0.05$) plants. The correlation between the relative humidity and the number of *C. rufipes* visits was not significant on untreated ($r = -0.29$; $P > 0.05$), compost applied ($r = -0.35$; $P > 0.05$), and chemical fertilizer applied ($r = -0.28$; $P > 0.05$) subplots.

Table 3 Frequency of *Chalicodoma rufipes* visits on *Phaseolus vulgaris* flowers based on daily observation periods at Dang in 2018 and 2021

years	Daily periods (hours)												A
	6-7		8-9		10-11		12-13		14-15		16-17		
	n	*P* (%)	*N*	*P* (%)	*N*	*P* (%)	*n*	*P* (%)	*n*	*P* (%)	*N*	*P* (%)	
2018	1	1.19	7	8.33	33	39.29	35	41.6	7	8.33	1	1.19	84
2021	0	0	9	11.39	34	43.04	22	27.85	10	12.66	4	5.06	79
Total	1	0.61	16	9.82	67	41.1	57	34.97	17	10.43	5	3.07	163

n: number of visits in 11 et 9 observation days; ***A:*** total number of visits; ***P:*** percentage of visits; ***P*** = (*n* / *A*) * 100.

In 2018 and 2021, the highest mean number of *C. rufipes* simultaneously in activity was 1 per flower in each treatment. The mean number of *C. rufipes* in activity per 1000 flowers was 27, 18 and 14 in T_C, T_{NPK} and T_0, respectively in 2018 and 20, 16 and 14 in T_C, T_{NPK} and T_0, respectively in 2021 (table 4).

In 2018, a *C. rufipes* visit duration varied from 2 to 23 sec in subplots with compost, from 2 to 22 sec in NPK-fertilizer subplot, then from 2 to 21 sec in control subplots as shown in Table 5. In 2021 this bee visit duration varied from 2 to 22 sec in subplots with compost, from 2 to 23 sec NPK-fertilizer subplot, then from 2 to 21 sec in control subplots.

Table 4 Number of *Chalicodoma rufipes* individuals simultaneously in activity per 1000 *Phaseolus vulgaris* flowers (C_{1000}) in each subplot in 2018 and 2021 at Dang

Treatments	Years	C_{1000}					
		N	*m*	*Sd*	*mini*	*maxi*	Comparison of means
T_C	2018	64	27	11.17	10	62	*F* = 62.14 (*df1* = 5; *df2* = 232; *P* < 0.001; HS)
	2021	110	20	7.37	11	40	
	$T_{2018/2021}$	174	23.5	9.27	10.5	51	
T_{NPK}	2018	64	18	6.65	10	35	
	2021	110	16	4.33	10	27	
	$T_{2018/2021}$	174	17	5.49	10	31	
T_0	2018	64	14	4.83	10	37	
	2021	110	14	3.84	9	23	
	$T_{2018/2021}$	174	14	4.34	9.5	30	

T_C: subplot with compost; T_{NPK}: NPK-fertilizer subplot; T_0: untreated subplot; *m:* mean; *sd:* standard deviation; *n:* sample size; *maxi:* maximum; *mini:* minimum; *F*: ANNOVA test; *df1* and *df2:* degrees of freedom; *P:* level of significance; HS: highly significant.

Table 5 Duration of a flower visit by *Chalicodoma rufipes* in each *Phaseolus vulgaris* subplots in 2018 and 2021 at Dang

Treatments	Years	Duration of a flower visit (sec)					
		n	***m***	***sd***	***mini***	***maxi***	**Comparison of means**
T_C	2018	115	6.09	4.6	2	23	*F* = 54.68 (*df1* = 5; *df2* = 336; *P* < 0.001; HS)
	2021	171	6.13	3.28	2	22	
	$T_{2018/2021}$	286	6.11	3.94	2	22.5	
T_{NPK}	2018	115	5.57	3.56	2	21	
	2021	171	5.62	3.32	2	23	
	$T_{2018/2021}$	286	5.6	3.44	2	22	
T_0	2018	115	5.9	3.74	2	21	
	2021	171	5.82	3.36	2	21	
	$T_{2018/2021}$	286	5.86	3.55	2	21	

T_C: subplot with compost; T_{NPK}: NPK-fertilizer subplot; T_0: untreated subplot; ***m:*** mean; ***sd:*** standard deviation; ***n:*** sample size; ***maxi:*** maximum; ***mini:*** minimum; *F*: ANNOVA; ***df1*** and ***df2:*** degrees of freedom; ***P:*** level of significance; **HS:** highly significant.

Table 6 presents the foraging speed of *C. rufipes* in each *P. vulgaris* subplots. In 2018, a *C. rufipes* individual visited between 3.83 and 30 flowers/min in subplots with compost, 2.9 and 21 flowers/min in subplots with NPK-fertilizer, and 3.78 and 21 flowers/min in control subplots. In 2021 this bee visited between 3.67 and 21.43 flowers/min in subplots with compost, 2.94 and 11.54 flowers/min in NPK-fertilizer subplot then 3.87 and 13.55 flowers/min in control subplots.

During the observation period, flowers of many other plant species growing in the study station were visited by *C. rufipes* individuals, for nectar (ne) and pollen (po) collection. Among these plants were: *Vigna unguiculata* (Fabaceae; ne and po), *Phaseolus coccineus* (Fabaceae; ne), *Cajanus cajan* (Fabaceae; ne), *Senna mimosoides* (Fabaceae; po) and *Gossypium hirsutum* (Malvaceae; ne and po). During the whole observation period, *C. rufipes* foraging on *P. vulgaris* were not observed moving to neighboring plant species and vice versa.

Table 6 Foraging speed of *Chalicodoma rufipes* in each of *Phaseolus vulgaris* subplots in 2018 and 2021 at Dang

Treatments	Years	Foraging speed (flowers/min)					
		n	***m***	***sd***	***mini***	***maxi***	**Comparison of means**
T_C	2018	39	11.44	4.99	3.83	30	*F* = 54.68 (*df1* = 5; *df2* = 336; *P* < 0.001; HS)
	2021	36	9.81	3.92	3.67	21.43	
	$T_{2018/2021}$	75	10.63	4.46	3.75	25.72	
T_{NPK}	2018	39	8.22	3.42	2.9	21	
	2021	36	7.36	2.20	2.94	11.54	
	$T_{2018/2021}$	75	5.6	3.44	2.92	16.27	
T_0	2018	39	9.22	4.07	3.78	21	
	2021	36	7.95	2.09	3.87	13.55	
	$T_{2018/2021}$	75	7.54	3.08	3.83	17.28	

T_C: subplot with compost; T_{NPK}: NPK-fertilizer subplot; T_0: untreated subplot; *m:* mean; *sd:* standard deviation; *n:* sample size; *maxi:* maximum; *mini:* minimum; *F:* ANNOVA; *df1* and *df2:* degrees of freedom; *P:* level of significance; HS: highly significant.

3.5. Impact of Flowering Insects Including *Chalicodoma rufipes* on *Phaseolus vulgaris* production

Table 7 shows the data on the production parameters of *P. vulgaris* for each treatment applied in untreated subplots.

During nectar or pollen harvest on *P. vulgaris*, foraging insects always shook flowers and regularly contacted anthers and stigma, increasing self - pollination and/or cross - pollination possibilities of this plant species. It appears from this table 7 that:

- In 2018, the fruiting rate were 88.33 %, 43.33 %, 82.61 % and 66.67 % in treatments 1, 2, 7 and 9, respectively. While in 2021, it was 85.83 %, 45.83 %, 78.05 % and 65.08 % in treatments 3, 4, 8 and 10, respectively. The differences between these eight percentages were highly significant (χ 2 = 38.75; df = 7; P < 0.001). The two - by - two comparisons showed that the difference observed is significant between treatments 1 and 2 (χ 2 = 11.68; df = 1; P < 0.01) and highly significant between treatments 3 and 4 (χ 2 = 25.54; df = 1; P < 0.001). Consequently, in 2018 and 2021, the fruiting rate of unprotected flowers (treatments 1 and 3) was higher than that of protected flowers (treatments 2 and 4).
- In 2018, the mean numbers of seeds per pod were 7.82, 5.64, 8.42, and 6.7 in treatments 1, 2, 7 and 9, respectively. Whereas in 2021, it was 7.92, 5.98, 8.45 and 6.25 in treatments 3, 4, 8 and 10, respectively. The differences between these eight means are highly significant (F = 43.15; df1 = 143; df2 = 8; P < 0.001). The two - to - two comparisons showed that the difference observed is significant between treatments 1 and 2 (t = 23.54; df = 59; P < 0.01) and significant (t = 18.65; df = 74; P < 0.01) between treatments 3 and 4. Consequently, in 2018 and 2021, the mean number of seeds per pod of unprotected flowers was higher than that of protected flowers.
- In 2018, the percentages of normal seeds were 90.79 %, 67.37 %, 91.09 % and 63.43 % in treatments 1, 2, 7 and 9, respectively, compared 91.92 %, 77.26 %, 92.02 % and 76 % in treatments 3, 4, 8 and 10, respectively. The differences between these eight percentages are generally highly significant (χ 2 = 84.32; df = 7; P < 0.001). Paired wise comparisons showed that the difference observed was highly significant between treatments 1 and 2 (χ 2 = 35.11; df = 1; P < 0.001) as well as between treatments 3 and 4 (χ 2 = 62.24; df = 1; P < 0.001). Hence, in 2018 and 2021, the percentage of normal seeds of unprotected flowers was higher than that of protected flowers.

Table 7 Production parameters of *Phaseolus vulgaris* as influenced by insects Including *Chalicodoma rufipes* in untreated subplots in 2018 and 2021 at Dang

Years	Treatments	NF	NFP	FrR (%)	Seeds/pod		TNS	NNS	% NS
					m	*Sd*			
2018	1 (Uf)	120	106	88.33	7.82	1.02	391	355	90.79
	3 (Pf)	120	52	43.33	5.64	0.92	282	190	67.37
	7 (Fpvx)	92	76	82.61	8.42	0.78	337	307	91.09
	9 (Fpwv)	69	46	66.67	6.7	0.82	268	170	63.43
2021	2 (Uf)	120	103	85.83	7.92	0.83	396	364	91.92
	4 (Pf)	120	55	45.83	5.98	0.98	299	231	77.26
	8 (Fpvx)	82	73	89.02	8.45	0.75	338	311	92.02
	10 (Fpwv)	63	41	65.08	6.25	0.74	250	190	76

NF: number of flowers; NFP: number of formed pods; FrR: fruiting rate; TNS: total number of seeds; NNS: number of normal seeds; %NS: percentage of normal seeds; *m:* mean; *sd:* standard deviation; Uf: unprotected flowers; Pf: flowers bagged; Fpvx: flowers protected using gauze bag nets then uncovered, visited once by *C. rufipes* and rebagged; Fpwv: flowers protected using gauze bag nets, uncovered, then rebagged without the visit of insects or any other organism.

In 2018, the numeric contribution of the anthophilous insects including *C. rufipes* in the fruiting rate, the mean numbers of seeds per pod and the percentages of normal seeds of *P. vulgaris* were 33.33 %, 16.57 % and 28.88 % respectively. In 2021, the corresponding figures were 31.17 %, 21.83 % and 17.09 % respectively. For the two cumulated years, the numeric contributions of flowering insects were 32.25 %, 19.2 % and 22.98 % for the fruiting rate, the mean numbers of seeds per pod and the percentages of normal seeds, respectively.

3.6. Pollination efficiency of *Chalicodoma rufipes* on *Phaseolus vulgaris*

During the collection of nectar on each *P. vulgaris* flower individuals of *C. rufipes*, always come into contact with stigma and anthers. During nectar collection in *Phaseolus vulgaris* flowers, *C. rufipes* were always found to shake flowers. This movement could facilitate the liberation of pollen by anthers, for the optimal occupation of the stigma [32]. The percentage visits during which individual bees came into contact with anthers and stigma was 100% of 864 nectar harvest observations. Thus, *C. rufipes* highly increased the pollination potentials of *P. vulgaris* flowers.

In 2018, the fruiting rate due to *C. rufipes* was 19.3 %. The difference was highly significant between treatments 7 and 9 ($\chi 2$ = 59.95; df = 1; P < 0.001). While in 2021, it was instead 26.89 %. The difference was highly significant between treatments 8 and 10 ($\chi 2$ = 64.63; df = 1; P < 0.001). For the two cumulated years, the fruiting rate due to *C. rufipes* was 23.1 %.

During the same year, the mean number of seeds per pod due to *C. rufipes* was 20.43 %. The difference was highly significant between treatments 7 and 9 (t = 4.56; P < 0.01), compared to 26.04 % in 2021. The difference was highly significant between treatments 8 and 10 (t = 6.86; P < 0.01). For the two cumulated years, the mean number of seeds per pod due to *C. rufipes* was 23.24 %.

In 2018, the percentage of normal seeds due to *C. rufipes* was 30.37 %. The difference was highly significant between treatments 7 and 9 ($\chi 2$ = 64.59; df = 1; P < 0.001). In 2021, the percentage of normal seeds due to *C. rufipes* was 17.41 %. The difference was highly significant between treatments 8 and 10 ($\chi 2$ = 59.61; df = 1; P < 0.001). For the two cumulate years the the percentage of normal seeds due to *C. rufipes* was 23.89 % in subplots.

3.7. Impact of compost on production of *Phaseolus vulgaris*

Table 8 shows the datas on the production parameters of *P. vulgaris* as influenced by compost.

The fruiting rate due to compost was 40.23% in 2018 and 37.5% in 2021. The difference observed between the fruiting rate from treatments 2 and 5 ($\chi 2$ = 3.43; df = 1; P < 0.001) on one hand treatments 4 and 6 ($\chi 2$ = 1.27; df = 1; P < 0.001) on the other hand were highly significant. For the two cumulated years the fruiting rate due to compost was 38.87%.

Table 8 Production parameters of *Phaseolus vulgaris* as influenced by compost in 2018 and 2021 at Dang

Years	Treatments	NF	NFP	FrR (%)	Seeds/pod		TNS	NNS	% NS
					M	***Sd***			
2018	5 (Pfc)	120	87	72,5	6,44	1,16	322	270	83,85
	3 (Pf)	120	52	43,33	5,64	0,92	282	190	67,37
2021	6 (Pfc)	120	88	73,33	7,02	1,15	351	312	88,89
	4 (Pf)	120	55	45,83	5,98	0,98	299	231	77,26

NF: number of flowers; NFP: number of formed pods; FrR: fruiting rate; TNS: total number of seeds; NNS: number of normal seeds; %NS: percentage of normal seeds; *m:* mean; *sd:* standard deviation; Pf: flowers bagged; Pfc: flowers bagged on compost subplots.

In 2018, the mean number of seeds per pod due to compost was 12.42% against 14.81% in 2021. The difference observed between the fruiting rate from treatments 2 and 5 were highly significant (t = 141.26; df = 352; P < 0.001). The difference observed between the fruiting rate from treatments 4 and 6 were highly significant (t = 125.49; df = 284; P < 0.001). For the two cumulated years the mean number of seeds per pod due to compost was 13.62%.

In 2018, the percentage of the normal seeds attributed to compost was 19.65% compared to 13.08% in 2021. The difference observed between the fruiting rate from treatments 2 and 5 were highly significant ($\chi 2$ = 1.32; df = 1; P < 0.001). The difference observed between the percentage of the normal seeds from treatments 4 and 6 were highly significant ($\chi 2$ = 1.86; df = 1; P < 0.001). For the two cumulated years, the percentage of the normal seeds accounting for compost was 16.37%. Consequently, in 2018 and 2021, the fruiting rate, the mean number of seeds per pod and the percentage of normal seeds from bagged flowers on plant applied with compost (treatment 5 and 6) was higher than that of bagged flowers on plant untreated (treatment 2 and 4).

The positive and significant contribution of compost in pods and seed production of *P. vulgaris* could be justified by its richness in nutrients such as phosphorus, nitrogen and potassium [12]. In fact, nitrogen is considered as the most important macro-element, responsible for longitudinal growth of branches and fruit production [33].

3.8. Cumulative impact of compost and *Chalicodoma rufipes* on pollination and production of *Phaseolus vulgaris*

Table 9 shows the datas on the production parameters of *P. vulgaris* as influenced by *Chalicodoma rufipes* and compost.

Table 9 Production parameters of *Phaseolus vulgaris* as influenced by *Chalicodoma rufipes* and compost in 2018 and 2021 at Dang

Years	Treatments	NF	NFP	FrR (%)	Seeds/pod		TNS	NNS	% NS
					M	*Sd*			
2018	9 (Fpwv)	69	31	44.93	6.59	0.87	369	243	65.85
	11 (Fpvxc)	126	121	96.03	8.23	0.85	955	946	99.05
2021	10 (Fpwv)	63	32	50.79	6.19	0.77	347	222	63.97
	12 (Fpvxc)	122	119	97.54	8.29	0.8	962	954	99.17

NF: number of flowers; NFP: number of formed pods; FrR: fruiting rate; TNS: total number of seeds; NNS: number of normal seeds; %NS: percentage of normal seeds; *m:* mean; *sd:* standard deviation; Fpwv: flowers protected using gauze bag nets, uncovered, then rebagged without the visit of insects or any other organism; Fpvxc: flowers protected using gauze bag nets then uncovered, visited once by *C. rufipes* and rebagged on compost subplots.

The fruiting rate due to cumulative impact of compost and *Chalicodoma rufipes* was 20.46% in 2018, whereas in 2021, it was 29.45% in 2021. The difference observed between the fruiting rate from treatments 9 and 11 were highly significant ($\chi 2$ = 0.87; *df* = 1; *P* < 0.001). The difference observed between the fruiting rate from treatments 10 and 12 were highly significant (χ *2* = 0.49; *df* = 1; *P* < 0.001). For the two cumulated years, the fruiting rate due to combined effect of compost and *Chalicodoma rufipes* was 24.96%.

In 2018, the mean number of seeds per pod accounting for combined effect of compost and *Chalicodoma rufipes* was 18.96%, and 19.06% in 2021. The difference observed between the fruiting rate from treatments 9 and 11 were highly significant (*t* = 178.32; *df* = 153; *P* < 0.001). The difference observed between the fruiting rate from treatments 10 and 12 were highly significant (*t* = 143.74; *df* = 178; *P* < 0.001). For the two cumulated years, the mean number of seeds per pod attributed to combined effect of compost and *Chalicodoma rufipes* was 19.01%.

In 2018, the percentage of normal seeds due to cumulative impact of compost and *Chalicodoma rufipes* was 19.81%, and 22.69% in 2021. The difference observed between the fruiting rate from treatments 9 and 11 were highly significant ($\chi 2$ = 1.31; *df* = 1; *P* < 0.001). The difference observed between the percentage of the normal seeds from treatments 10 and 12 were highly significant ($\chi 2$ = 0.93; *df* = 1; *P* < 0.001). For the two cumulated years the percentage of the normal seeds accounting for to combined effect of compost and *Chalicodoma rufipes* was 21.25%. Consequently, in 2018 and 2021, the fruiting rate, the mean number of seeds per pod and the percentage of normal seeds from flowers exclusively visited once by *C. rufipes* on plant applied compost (treatment 11 and 12) was higher than that of flowers protected, uncovered and rebagged without the visit of insects or any other organism on plant untreated (treatment 9 and 10).

Both *Chalicodoma rufipes* and compost highly improved the seed and pod production of *P. vulgaris*. *Chalicodoma rufipes* increased pollination possibilities [34]. The synergistic effects of *C. rufipes* and compost has largely increased the *P. vulgaris* production.

4. Conclusion

This study has revealed that *P. vulgaris* pink variety has a mixed reproduction regime allogamous-autogamous, with the predominance of allogamy. *P. vulgaris* is an important nectariferous plant for *C. rufipes*. The comparison of pod and seed sets of flowers visited once by *C. rufipes* with that of flowers protected and destinated to be uncovered then rebagged without the visit of insects or any other organism underscores the value of this bee in increasing pods and seed production as well as seed quality. When plants were amended with compost, *C. rufipes* had more flowers to visit on *P. vulgaris*. Compost and *C. rufipes* offered a cumulative impact which improved *P. vulgaris* production. Application of compost and installation of *C. rufipes* nests close to *P. vulgaris* pink variety fields is recommended to farmers for the increasing pod and seed production of this valuable crop.

Compliance with ethical standards

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Disclosure of conflict of interest

Me the corresponding author and the other authors who are my supervisors, declare that there is no conflict of interest between us.

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