

ORIGINAL RESEARCH

Comparative Evaluation of Bumblebee Bat (*Craseonycteris thonglongyai*) Droppings and Chemical Fertilizer on Mineral Content of *Ocimum gratissimum* (Basil) Leaves

David, E., Abu, M. L., Uthman, A. and Musa, A.*

Department of Biochemistry, Faculty of Natural Sciences, Ibrahim Badamasi Babangida University, Lapai, Niger State, Nigeria

*Corresponding author's email: musaamanabo@ibbu.edu.ng

ABSTRACT

Leafy greens like *Ocimum gratissimum* provide essential minerals supporting human nutrition, well-being, and immune function, though mineral concentrations are influenced by soil physicochemical properties and soil fertility. This study assessed the impact of Bumblebee Bat (*Craseonycteris thonglongyai*) droppings compared to commercial chemical fertilizer on mineral content (Fe, Zn, Mn, Cu, Co, Ni, Mg, Ca, Na, K) in *O. gratissimum* leaves. The mineral element content of *Ocimum gratissimum* was determined using a flame photometer for Na and K, and an atomic absorption spectrophotometer for Fe, Zn, Mn, Cu, Co, Ni, Mg, and Ca. The leaves of *Ocimum gratissimum* were digested with a mixture of concentrated perchloric and nitric acids, then treated with hydrochloric acid and diluted with distilled deionized water. The resulting solutions were analyzed for their mineral content. The findings revealed that the utilization of chemical fertilizer does not substantially influence the levels of Fe, Zn, Mn, Cu, Co, and K in *O. gratissimum*. Nevertheless, the concentrations of these parameters experienced a significant increase ($p < 0.05$) following the application of *C. thonglongyai* droppings in the vegetable. Similarly, while the utilization of chemical fertilizer significantly decreased ($p < 0.05$), the concentrations of Ni, Mg, and Na in *O. gratissimum*, treatment with *C. thonglongyai* droppings considerably raised ($p < 0.05$) the concentrations of these mineral elements in the vegetable. Furthermore, the outcomes also demonstrated that treatment with both fertilizers considerably enhanced the levels of Ca in *O. gratissimum*. This study revealed that *C. thonglongyai* droppings significantly enhanced all the studied minerals in *O. gratissimum* compared to chemical fertilizer, supporting the integration of organic fertilizer into sustainable agriculture that reduces chemical dependency, minimizes pollution, improves soil health, and enhances biodiversity.

Keywords: *Craseonycteris thonglongyai*, droppings, *Ocimum gratissimum*, Mineral elements, chemical fertilizer.

Introduction

Ocimum gratissimum, commonly recognized as scent leaf, is an extensively cultivated and commercially feasible perennial herbaceous plant with a remarkably fragrant odour. This plant belongs to the *Lamiaceae* family and thrives in continents such as Africa, Asia, and South America (Ugbogu et al., 2021; Akara et al., 2021). In traditional medicine, *Ocimum gratissimum* treats fungal/bacterial infections, cough, pneumonia, fever, inflammation, and anemia. It serves as a vegetable, condiment, or natural flavoring in culinary preparations of fish, meat, soups, and stews. *Ocimum gratissimum* contains substantial amounts of potassium, calcium, magnesium, and iron (Etukudo et al., 2021; Ikpeazu et al., 2019; Adebola, 2017; Kiendrebeogo et al., 2016). It also contains high levels of physio-chemicals such as; flavonoids, phenols, alkaloids, saponins, and tannins (Priscilla, 2016). Other elements found in *O. gratissimum* leaves include sodium, phosphorus, zinc, and copper. The high nutritional content of *O. gratissimum's* suggests its potential role in diet formulation diets and alleviation of malnutrition symptoms. Therefore, *O. gratissimum* serve as nutrient rich food source with its nutritional components, also supporting its medical benefits.

Like other vegetables, the nutritional content of *O. gratissimum* is influenced by soil nutrient status. One of the organic fertilizers used to amend the soil and promote plant growth and development is the droppings of *Cratogeomys thonglongyai* (Musa, 2016a). The droppings of *C. thonglongyai* contain exceptionally high amounts of nitrogen, phosphorous, and potassium, which are highly essential for optimum plant growth. Additionally, certain microbes present in *C. thonglongyai* droppings enhance soil structure thereby, facilitating nutrient absorption by plants (Musa et al., 2016a; Musa, 2016a).

Previous study has demonstrated that organic fertilizers, just like bat droppings, can promote the nutritional quality of plants by improving mineral content and minimizing harmful phytotoxins (Mlitan, 2014). Chemical fertilizers, while effective in promoting plant growth, may lead to higher nitrate accumulation, which can be hazardous to health (Pahalvi et al., 2021). The application of bumblebee bat droppings has been shown to decrease the bioaccumulation of some specific phytotoxins in plants, potentially producing a safer and more nutritious produce compared to synthetic fertilizers (Mlitan, 2014).

Organic fertilizers like vermicompost have been found to improve the mineral content and overall quality of basil leaves, suggesting that similar benefits could be expected from bat droppings (Ajayi, 2017). Using organic fertilizers such as bat droppings conforms with sustainable agricultural practices, reducing environmental impact and promoting biodiversity (Kumar et al., 2024; Mlitan, 2014). The enhanced mineral content in basil leaves fertilized with organic matter can contribute to better health outcomes for consumers, supporting the therapeutic uses of basil in traditional medicine (Olumide et al., 2019). In this context, the present study seeks to evaluate the influence of *C. thonglongyai* droppings on the bioaccumulation of trace elements such as iron, zinc, manganese, copper, cobalt, nickel, magnesium, calcium, sodium, and potassium in the leaves of *O. gratissimum*.

The use of bumblebee bat droppings, a naturally occurring organic fertilizer, to raise the mineral content of basil leaves is what makes this work novel. Using exact analytical methods like flame photometry and atomic absorption spectrophotometry, it is the first to directly compare this organic supplement to conventional chemical fertilizers. This study offers new insights into how unusual organic fertilizers can affect plant nutritional quality by focusing on the mineral nutrient profile, in contrast to many earlier studies that concentrate on antioxidants or phytotoxin levels. This approach

not only broadens our understanding of alternative nutrient management practices but also highlights a sustainable and eco-friendly fertilizer option for crop production.

Materials and Methods

The Study Area

The Pot experiment was conducted in Ibrahim Badamasi Babangida University in Lapai, Niger State, in the Department of Biochemistry. Lapai is situated at latitude 6° 49' N and longitude 6° 41' E in the Southern Guinea Savanna area of Nigeria. The rainy season lasts from April to October and has a mean annual rainfall of 1334 mm. The heaviest rainfall months are August and September, with 300–330 mm. Lapai exhibits average monthly temperatures ranging from 30–40°C (March) to 22.3°C (August).

Soil sampling and analysis

The soil samples utilized in this investigation were taken at a depth of 0–20 cm (Lawal et al., 2022) and were obtained from Ibrahim Badamasi Babangida University in Lapai. To get rid of debris, the samples were bulked, dried, powdered, and sieved through a 2 mm sieve. The physical and chemical characteristics of the soil from the location and the droppings of *C. thonglongyai* were assessed using the method of Bieganowski et al. (2016).

Source of Seeds

National Horticultural Research Institute (NIHORT) in Ibadan, Oyo State, Nigeria, provided the basil seeds.

Source of *Craseonyctris thonglongyai* droppings

The dried samples of *C. thonglongyai* droppings were collected into sterile polythene bags from a cave in Faso, Edati Local Government Area, Niger State.

Manure Treatment

In a pot experiment, 50 g of the dried powdered dung of *C. thonglongyai* per 10 kg of soil was used to cultivate *O. gratissimum* after germination. NIHORT (1983) and Musa et al. (2010) recommended that 30 mg of N, 30 mg of P₂O₅, and 30 mg of K₂O be applied to the soil while applying synthetic fertilizer. Manure and fertilizers were not applied to the plants in control pots.

Cultivation Conditions

Approximately ten seeds were planted in each pot containing ten kilograms of soil. After germination, the number of *O. gratissimum* seeds per pot was reduced to two. Three soil amendments levels were used in the Complete Randomized Design (CRD) experiment: 50 g of *C. thonglongyai* droppings every 10.00 kg of soil, synthetic fertilizer applied at the recommended dose, and control (no application). There are 90 pots altogether for the trial—10 pots for each treatment, replicated three times. The plants were irrigated in the mornings and evenings. The surrounding and inside the pot were regularly weeded to prevent competition and pest infestation. Periodically, the pots were removed to stop the plant roots from spreading beyond the container (Musa, 2016b).

Analysis of Plant Samples

After harvesting fresh *O. gratissimum* leaves at market maturity from the pots, samples were analyzed using standard methods to determine the elemental content in leaves of plants grown synthetic fertilizer, *C. thonglongyai* droppings and without application of fertilizer (control).

Mineral Element Contents

The mineral element content of the samples was assessed using a flame photometer for Na and K and an atomic absorption spectrophotometer for Fe, Mg, Mn, Zn, Cu, Ca, Co, and Ni, following the

methodology of Ezeonu et al. (2002). In this method, the leaves of the vegetable were dried in an oven at the temperature of 110⁰C for 24 hours. After drying, they were ground into powder form with mortar and pestle and exactly 0.500 g of the ground dried samples were weighed into a boiling tool and 5.0 cm³ of digestion mixture which is made up of concentrated perchloric acid and nitric acid in a ratio of 1:2 was added. The mixtures were swirled and left in a fume cupboard overnight. They were then digested at the temperature of 150 ⁰C on a hot plate for 2 hours or until frothing ceased. At the end of 2 hours, the samples were taken from the hot plate and cooled for 10 minutes after which 3.0 cm³ 6.0 M HCL was added, and the sample were further digested for another 1 ½ hour. The digestion flasks were removed from the hot plate and allowed to cool. The contents of each tube were made up to 50cm³ with distilled deionised water in volumetric flask and later transferred into sample bottles. The sample were analysed for their mineral content of interest using atomic absorption spectrophotometer and flame photometer for Na and K only.

Statistical analysis

To ascertain the differences in the amounts of mineral elements in *O. gratissimum* grown with the three treatments, Analysis of variance (ANOVA) was performed using the SAS statistical program version 9.1. The significance level was set at $P < 0.05$, and mean with statistical significant differences were separated using Duncan's Multiple Range Test (DMRT).

Results

Physical and Chemical Properties of Soil

The analysis of the soil used for the pot experiment is shown in Table I. The soil's pH of 5.7 indicates that it is strongly acidic; it has low levels of calcium and organic carbon, moderate levels of

available phosphorus and magnesium, and very high concentrations of potassium and sodium. The soil has moderate total nitrogen content (Khamis et al., 2022; Chaudhary, 2015).

Table 1: Physical and chemical properties of the soil (0 – 20 cm depth) used for the experiment

Parameters	Values
Sand (%)	83.50 ± 0.32
Clay (%)	8.89 ± 0.24
Silt (%)	6.61 ± 0.01
Textural class	Sand
Organic carbon (g kg ⁻¹)	4.51 ± 0.02
pH (H ₂ O)	5.70 ± 0.05
Total nitrogen (g kg ⁻¹)	1.68 ± 0.01
pH (CaCl ₂)	7.21 ± 0.20
Available phosphorus (mg kg ⁻¹)	15.91 ± 1.00
Na ⁺ (cmol kg ⁻¹)	3.76 ± 0.07
Mg ²⁺ (cmol kg ⁻¹)	1.30 ± 0.021
Ca ²⁺ (cmol kg ⁻¹)	3.71 ± 0.20
Acidity (cmol kg ⁻¹)	0.63 ± 0.02
EC (cmol kg ⁻¹)	8.78 ± 0.22
Base saturation (%)	94.36 ± 4.19
CEC (cmol kg ⁻¹)	9.50 ± 0.22

CEC = Cation exchange capacity, EC = Exchangeable cations. Values represent Mean ± SEM of triplicate determinations. SEM = Standard error mean.

Chemical Properties of *C. thonglongyai* Droppings

The chemical properties of *C. thonglongyai* droppings are presented in Table 2. The results showed that the available phosphorus, organic carbon, total nitrogen, potassium, sodium, and droppings are high. Whereas the concentration of magnesium in the dropping is high, the calcium content is low. The pH of the droppings, which is 7.56, indicates that it is slightly basic (Khamis et al., 2022; Chaudhary, 2015).

Table 2: Chemical properties of the *C. thonglongyai* droppings

Parameters	Values
pH (H ₂ O)	7.66 ± 0.13
Organic carbon (g kg ⁻¹)	37.21 ± 1.01
Total nitrogen (g kg ⁻¹)	7.97 ± 0.13
Available phosphorus (mg kg ⁻¹)	8758.0 ± 33.11
Na ⁺ (cmol kg ⁻¹)	3.41 ± 0.03
K ⁺ (cmol kg ⁻¹)	10.88 ± 0.60
Mg ²⁺ (cmol kg ⁻¹)	3.78 ± 0.21
Ca ²⁺ (cmol kg ⁻¹)	2.77 ± 0.23

Values represent Mean ± SEM of triplicate determinations. SEM = Standard error mean.

Mineral Element Contents

The result of the effects of *C. thonglongyai* droppings and chemical fertilizer on the concentrations of mineral elements in *O. gratissimum* is presented in Table 3. The result showed that the application of chemical fertilizer has no significant effect on the concentrations of Fe, Zn, Mn, Cu, Co, and K in the leaves of *O. gratissimum*. However, the concentrations of these elements were significantly increased ($p < 0.05$) with the application of *C. thonglongyai* droppings in the vegetable. The mean values of Fe, Zn, Mn, Cu, Co and K in the leaves of the control vegetable were 3.97 ± 0.11 , 0.27 ± 0.04 , 0.91 ± 0.27 , 0.13 ± 0.01 , 0.09 ± 0.00 and 243.33 ± 18.5 mg/kg respectively, while the corresponding values of the elements in the vegetable grown with *C. thonglongyai* droppings were 4.56 ± 0.08 , 0.49 ± 0.05 , 4.85 ± 2.09 , 0.17 ± 0.01 , 0.24 ± 0.06 and 276.67 ± 27.28 mg/kg and those grown with chemical fertilizer were 4.09 ± 0.64 , 0.28 ± 0.11 , 0.98 ± 0.27 , 0.14 ± 0.01 , 0.09 ± 0.01 and 223.33 ± 3.33 mg/kg respectively.

Similarly, while the application of chemical fertilizer significantly decreased ($p < 0.05$) the concentrations of Ni, Mg, and Na in *O. gratissimum*, grown with *C. thonglongyai* droppings significantly increased ($p < 0.05$) the concentrations of the mineral elements in the leaf of the plant.

The Ca concentration in the control group was significantly lower than the values recorded in the

vegetables grown with either fertilizer. However, the concentration of Ca in the leaves of *O. gratissimum* grown with chemical fertilizer and *C. thonglongyai* droppings did not differ significantly from each other (Table 3).

Table 3: Effect of *C. thonglongyai* droppings and Chemical fertilizer on the concentration of mineral elements in the leaf of *O. gratissimum*

Mineral elements (mg/kg)	Control	<i>C.thonglongyai</i> droppings	Chemical fertilizer
Fe	3.97 ± 0.11 ^a	4.56 ± 0.08 ^b	4.09 ± 0.64 ^a
Zn	0.27 ± 0.04 ^a	0.49 ± 0.05 ^b	0.28 ± 0.11 ^a
Mn	0.91 ± 0.27 ^a	4.85 ± 2.09 ^b	0.98 ± 0.27 ^a
Cu	0.13 ± 0.01 ^a	0.17 ± 0.01 ^b	0.14 ± 0.01 ^a
Co	0.09 ± 0.00 ^a	0.24 ± 0.06 ^b	0.09 ± 0.01 ^a
Ni	0.11 ± 0.00 ^b	0.36 ± 0.24 ^c	0.06 ± 0.02 ^a
Mg	21.68 ± 1.54 ^b	25.04 ± 2.12 ^c	17.31 ± 10.07 ^a
Ca	1.44 ± 0.05 ^a	1.80 ± 0.19 ^b	1.72 ± 0.14 ^b
Na	216.67 ± 8.82 ^b	226.67 ± 18.56 ^c	160.00 ± 20.82 ^a
K	243.33 ± 18.5 ^a	276.67 ± 27.28 ^b	223.33 ± 3.33 ^a

Mean ± Standard errors on the same row with different letters are significantly different ($p \leq 0.05$)

Discussion

The selected soil textural class for the study is sand, although it has good workability, water penetration, and aeration, it has low nutrient content and poor water retention capacity. The cation exchange capacity (CEC) is constrained despite the relatively high base saturation (Ma et al., 2024).

Low concentrations of mineral elements in the soil can be linked to the low concentrations of several critical soil nutrients in the research soil, especially the mineral elements since it is well-known that organic matter diminishes with continued land usage for crop production (Sumner and Bardhan, 2022).

According to Musa et al. (2022a), Musa et al. (2022b), and Abu et al. (2020), low concentrations of these vital nutrients for plant growth and development justify the need for adequate manure modification of the soil to improve soil nutrient content and composition. The dung utilized as a soil amendment is somewhat basic (Ndzeshala et al., 2023). Therefore, crop growers have good reason to use the dung to modify the soil to improve the nutrient composition of the soil and enhance growth, given that the high contents of nitrogen, phosphorus, and potassium in the dung of *C. thonglongyai* are among the crucial nutrients of the soils needed by plants for optimum performance (Abu et al., 2020; Musa et al., 2022a).

The significantly higher concentration of Ca in the leaves of *O. gratissimum* grown on the soil fertilized with both chemical fertilizer and *C. thonglongyai* droppings than the control is in agreement with the submission of Kansal et al. (2007); Safaa and Abd El Fattah (2007); Musa (2010); Musa et al. (2016b); Abu et al. (2020) that application of organic and inorganic fertilizers increase the mineral content in the vegetables. Safaa and Abd El Fattah (2007) further stressed that the increase in the mineral contents in vegetables following the application of fertilizers is due to the increase in the concentrations of chlorophylls. Similarly, the higher concentrations of Fe, Zn, Mn, Cu, Co, Ni, Mg, Na, and K in the leaves of *O. gratissimum* cultivated with *C. thonglongyai* droppings when compared with chemical fertilizer and control align with the submission of Abu et al. (2020), Musa et al. (2016b) and Musa (2017) that organic manures increase the concentrations of some mineral elements in vegetables. Musa et al. (2016b) stressed that organic manures trigger

some species of microorganisms in the soil, and the activities of these microbes help to stimulate nutrient absorption that improves plant growth and development.

Likewise, Alomari et al. 2024, put forward that the increase in the concentrations of Ca, Fe, and Zn in *Lactuca sativa* grown with the manure of bounce back may be ascribed to the balanced nutrient contents in the compost, which facilitates absorption of nutrients by the plant. The dung of *C. thonglongyai* used in the present study is organic manure with high concentrations of carbon, nitrogen, and essential mineral elements and also harbors some beneficial microorganisms. When applied to soil, the droppings improved the physical and chemical properties of the soil and microbial population. It therefore follows that the increase in the concentrations of Fe, Zn, Mn, Cu, Co, Ni, Mg, Na, and K in *O. gratissimum* with the application of the droppings may suggest two things: firstly, a direct nutrients interface and absorption from the soil by the plant because the droppings have high concentrations of essential nutrients including some mineral elements required by plants for normal growth and development (Abu et al., 2020).

Secondly, the high concentrations of nutrients and the activities of the microorganisms in the droppings of *C. thonglongyai* dung facilitate the breakdown of organic matter in the soil and transform them into nutrients for plants' roots. All these activities improve the cation exchange capacity (CEC) of soil and the density of soil, creating good texture and reconditioning the soil for optimal nutrient assimilation by plants (Abu et al., 2020). The decrease in the concentrations of Mg, Ni, and Na in *O. gratissimum* with the application of chemical fertilizer further strengthened the submission by several researchers that organic fertilizers are better at improving the nutrient contents in vegetables than conventional chemical fertilizers (Mukherjee et al., 2020; Musa, 2022a; Sharma and Chetani, 2017; Yu et al., 2018).

Conclusion

According to the current study, it can be concluded that adding *C. thonglongyai* droppings to the soil significantly enhances the build-up of mineral elements in *O. gratissimum* leaves when compared to chemical fertilizer (commercial reference). Consequently, it is hypothesized that growing vegetables using the droppings of *C. thonglongyai* will help reduce the prevalence of age-related and degenerative disorders brought on by the production of free radicals. With the identified enhancement of mineral elements in leafy vegetables, there could be increased interest in promoting *O. gratissimum* as a functional food. Future research could investigate its role in alleviating age-related and degenerative disorders, potentially leading to dietary recommendations or supplements aimed at improving health outcomes.

From a broader perspective, the implications of this research could extend beyond *O. gratissimum* to other crops and vegetables. Further studies could explore the effectiveness of *C. thonglongyai* droppings on a variety of plants, leading to broader adoption in different agricultural systems.

Authors Contributions

***Musa**, A. conceived the project run by **David**, E. as an MSc project, while ***Musa**, A., and **Uthman**, A. designed the experiment, whereas **Musa**, A. and **Abu**, M.L. prepared the manuscript. All authors read and approved the final manuscript.

Conflict of interest

The authors declare that there is no conflict of interest.

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