



(RESEARCH ARTICLE)



Bat guano and Nettle slurry (*Urtica dioica L.*) used as biostimulants on *Delosperma cooperi* and *Sedum rubrotinctum* plants

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Abstract

The aim of the experiment was to evaluate the use and influence of Bat guano and Nettle slurry on the growth, flowering and mineral absorption of *Delosperma cooperi* and *Sedum rubrotinctum* plants. The 3 experimental groups in cultivation were: (1) group without biostimulants (CTRL), irrigated with water and previously fertilized substrate; (2) group Bat guano (BG) and fertilized substrate; (3) group Nettle slurry (*Urtica dioica L.*) (UD) and fertilized substrate. Treatments with Nettle slurry and Bat guano resulted in a significant improvement in the growth and flowering characteristics of *Delosperma cooperi* and *Sedum rubrotinctum*. The test showed in fact a significant increase in the agronomic parameters analysed in plants treated with Bat guano and Nettle slurry. In fact, all the plants treated with (BG and UD) showed a significant increase in the number of leaves, flowers and inflorescences, in the vegetative, radical and inflorescence weight, in the flowering time. The test also resulted in an increase in the NPK mineral absorption and chlorophyll content of plants treated with bat guano and nettle slurry. Interesting aspects that make Bat guano and Nettle slurry valid alternatives to be used as plant biostimulants, when the plants are subjected to biotic and abiotic stresses, in particular transplant, water and salt stress.

Keywords: Succulent; Plant nutrition; Biofertilizer; Plant extract; Abiotic stress; Transplanting plants

1. Introduction

Delosperma is a big and varied genus, including more than 150 species, also tuberous, of variable appearance for shape, size and colour, mostly bushy and upholstered. The branches may be erect, or creeping, at times deciduous. Cultivated for the abundant and solitary flowers, of red, white, yellow colour, which are usual to open early in the morning and close towards evening. Its habitat is southern Africa, Kenya, Eritrea, Arabia and Madagascar. Minimum cultivation temperature 3-5 °C. The tuberous species: *abyssinicum*, *mahonii*, *nakurense*, *oehleri*, *steytlerae*, require a minimum of 10-12°C [1].

The *Sedum* with about 600 species is the largest genus among the Crassulaceae. These are mostly herbaceous plants, some annual and biennial. The species *dendroideum*, *frutescens*, *oxypetalum*, *torulosum* are similar to trees. The Mexican species, more delicate, bloom in spring; the European and Asian species, more rustic, bloom between August and October; they show hanging or creeping stems and fleshy leaves, of different shape, alternate or opposite, with engraved or smooth margins. Genus suitable for suspended baskets, similar to the genus *Monanthes*. It is found in temperate regions, mountains of Europe, including the Mediterranean basin, Asia, North America, Peru, Central Africa and Madagascar [1].

Bats (Mammalia: Chiroptera) are interesting and mysterious animals that perform purely nocturnal activities. During the day, bats remain in shelters. The same excrements of the bat (urine and faeces) can be collected at the bottom of

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the shelters with formation of guano. Guano may be different depending on diet (insect, beetle and moth residues, etc.), foraging biotope and other factors. Few are the data on the gastrointestinal flora of bats and guano. It is widely used as a natural fertilizer due to its high nitrogen content and its nematocidal effect [2]. Bat guano is useful for the development of microflora as it contains all the essential nutrients for their growth. Unfortunately, bat guano has been ignored for a long time and chemical fertilizers have become the nutritional source of plants. Although, bat guano has been used in several regions as organic manure for some time, the nutritional values, and the existence of macro and trace elements of bat guano are still missing [3].

Several studies have indicated the possible use of Nettle slurry (*Urtica dioica L.*) as a fertilizer in organic farming for horticultural crops [4-5]. Can produce your own slurry or buy ready-made commercial products. The chemical composition of nettle plants has been extensively studied for medical purposes [6]. Its antioxidant, therapeutic and immunological properties are important. [7-9]. Although nettle has promising applications in the food, medical and cosmetic sectors [10], few studies have focused on the agronomic use of nettle slurry as a fertilizer, the chemical composition of fermented slurry and its effect on crop yields. Some studies have been conducted on the efficacy of aficide [11] of several nettle extracts. Rosnitschek-Schimmel showed that the most important nitrogen compound in nettle plants was free amino acids, of which asparagine and arginine accounted for up to 80%. These nitrogen compounds are mainly stored in roots and rhizomes [12].

In this study, the possibility of using Bat guano and Nettle slurry as possible biostimulants for *Delosperma cooperi* and *Sedum rubrotinctum* plants, was evaluated.

2. Material and methods

2.1. Greenhouse experiment and growing conditions

The experiments, started at the beginning of March 2019, were carried out in the greenhouses of CREA-OF in Pescia (Pt), Tuscany, Italy (43°54'N 10°41'E) on plants of *Delosperma cooperi* and *Sedum rubrotinctum*. The plants were placed in pots ø 12 cm; 80 plants for thesis divided into 4 replicas of 20 plants each.

All plants were fertilized with a controlled release fertilizer (5 kg m⁻³ Osmocote Pro® 6 months with 190 g/kg N, 39 g/kg P, 83 g/kg K) mixed with the growing medium before transplanting.

The 3 experimental groups in cultivation were:

Group without biostimulants (CTRL), irrigated with water and substrate previously fertilized;

Group with Bat guano (BG) and fertilised substrate. (Kalong, 50g per 7L of substrate);

Group with Nettle slurry (UD) and fertilized substrate, (La Praglia, 1L of Nettle slurry, per 100 liters of water every 10 days, for foliar fertilizer treatments).

The plants were watered daily and grown for 6 months.

On 26 August 2019, the number of leaves, number of flowers, vegetative and root weight, number and weight of inflorescences and duration of flowering were recorded.

Ten days before the destructive analysis, the content of chlorophyll (FieldScout CM 1000 Chlorophyll Meter) and N, P, K (Kjeldal UDK 169; Jenway 630501 6300 visible spectrophotometer) was evaluated.

2.2. Statistics

The experiment was carried out in a randomized complete block design. Collected data were analysed by one-way ANOVA, using GLM univariate procedure, to assess significant ($P \leq 0.05$, 0.01 and 0.001) differences among treatments. Mean values were then separated by LSD multiple-range test ($P = 0.05$). Statistics and graphics were supported by the programs Costat (version 6.451) and Excel (Office 2010).

3. Results

3.1. Plant growth

The test showed a significant increase in the agronomic parameters analysed in plants treated with bat guano and nettle slurry. In fact, all the plants treated with (BG and UD) showed a significant increase in the number of leaves (Figure 1A-2A), flowers (Figure 1B-3A) and inflorescences (Figure 2B), in the vegetative (Figure 1C-2C-3A-3B), radical (Figure 1D-2D) and inflorescence weight (Figure 2E), in the flowering time (Figure 1E).

In *Delosperma*, the number of leaves was 263.92 (BG), 245.67 (UD) compared to 194 of the control (Figure 1A). For the number of flowers was found 63 (BG), 52.83 (UD), 40.75 in (CTRL) (Figure 1B). There was a significant increase in vegetative biomass in (BG) 109.66 g and (UD) 93.92 g, compared to 76.32 g of the control (Figure 1C). The same is true for radical biomass in (BG) 74.61g and (UD) 76.49g compared to 54.70g in the control (Figure 1D). A significant effect of the use of bat guano on the flowering time, 4.83 days (BG) compared to 3.92 days of the water-irrigated control (CTRL), was also found (Figure 1E).

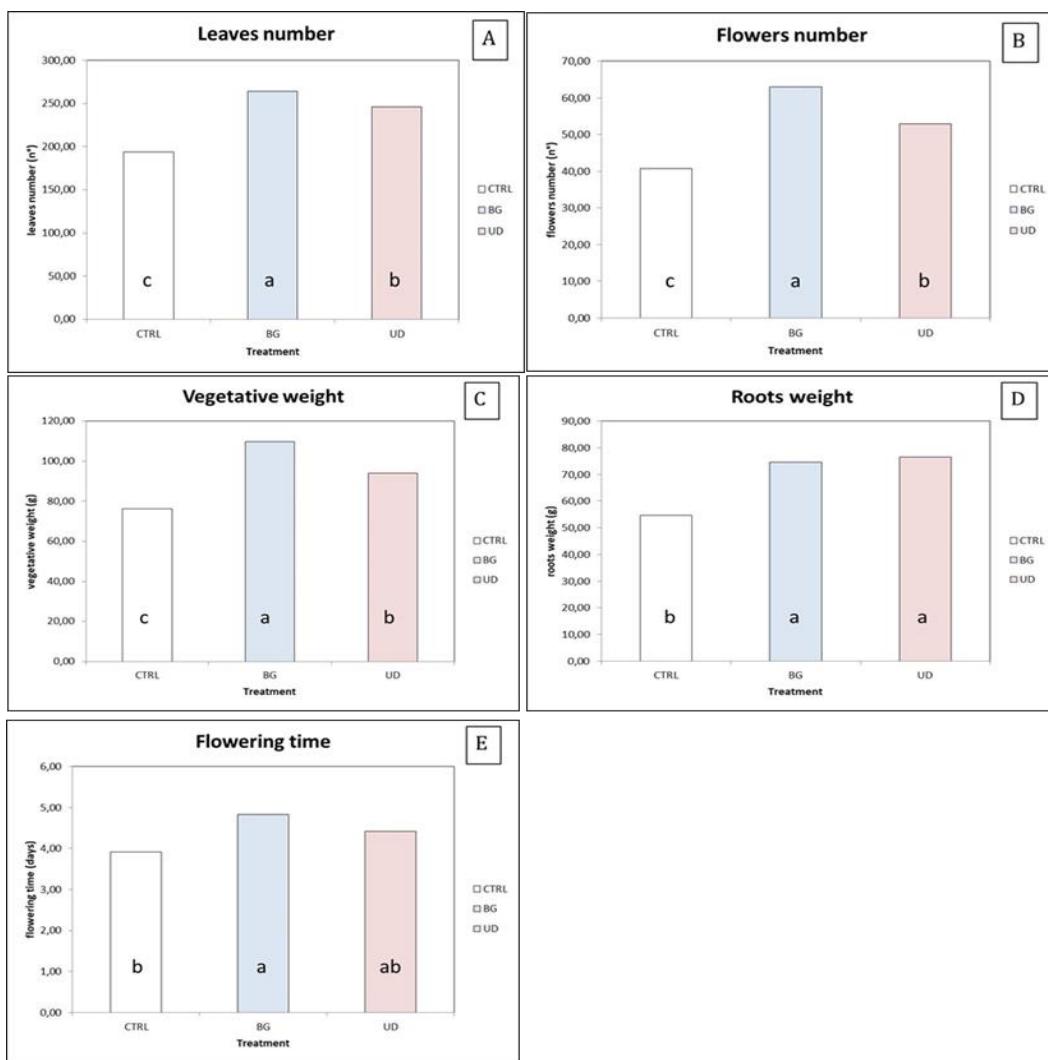


Figure 1 Effect of Bat guano and Nettle slurry (*Urtica dioica L.*) on growth and flowering improvement of *Delosperma cooperi*. Legend: (A) leaves number; (B) flowers number; (C) vegetative weight; (D) roots weight; (E) flowering time. Each value reported in the graph is the mean of three replicates ± standard deviation. Statistical analysis performed through one-way ANOVA. Different letters for the same parameter indicate significant differences according to LSD test (P = 0.05).

In *Sedum*, the number of leaves was 103.58 (BG), 105.75 (UD) compared to 86.58 of the control (Figure 2A). For the number of inflorescences was found 3.75 (BG), 3.33 (UD), 2.42 in (CTRL) (Figure 2B). Also on *Sedum* there was a

significant increase in vegetative biomass in (BG) 74.32 g and (UD) 71.13 g, compared to 65.76 g of the control (Figure 2C). Same for radical biomass in (BG) 52.89 g and (UD) 56.96 g against 46.63 g of control (Figure 2D) and weight of inflorescences 15.69 g in (BG), 14.39 g (UD) against 12.53 g of control (Figure 2E).

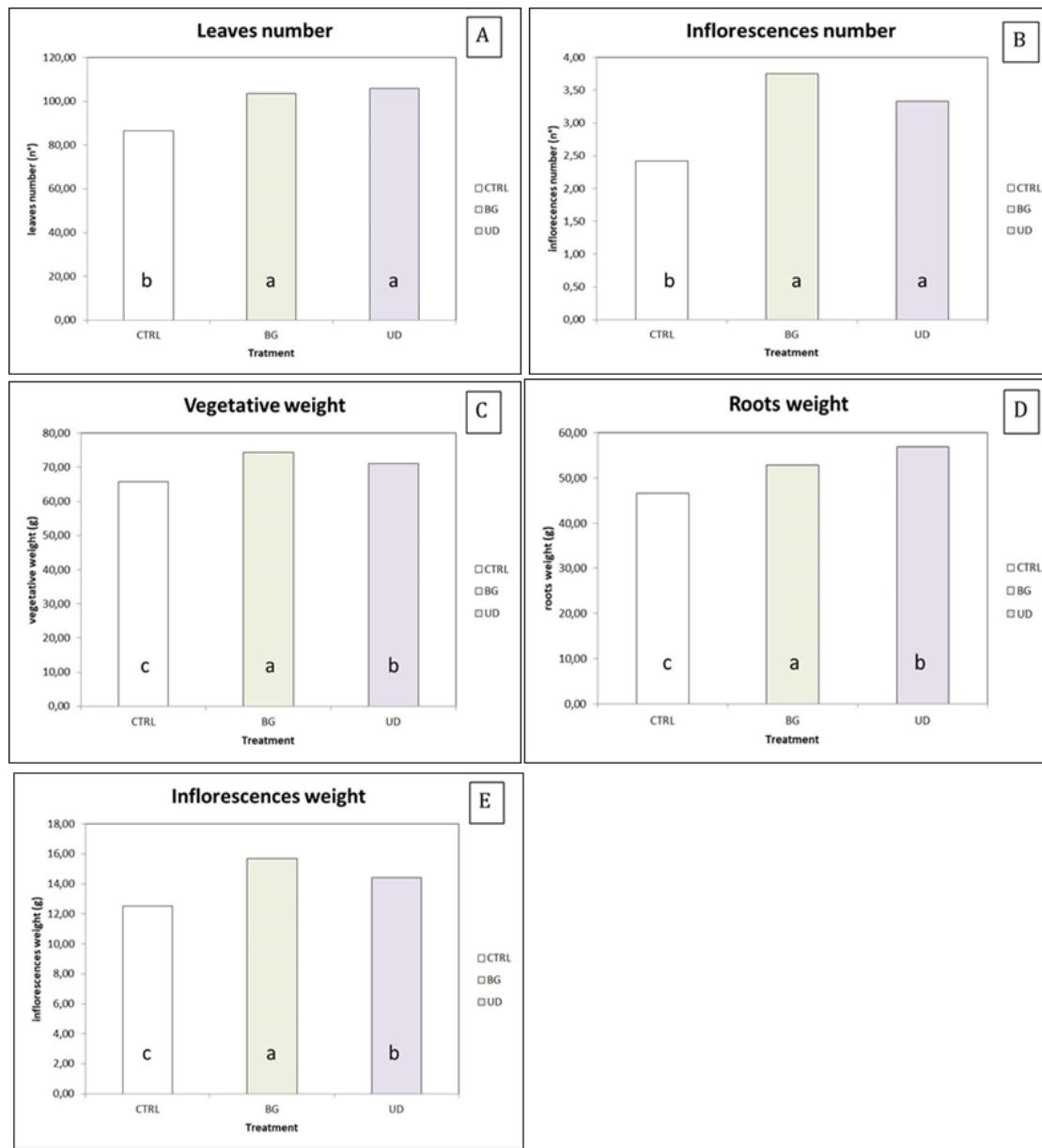


Figure 2 Effect of Bat guano and Nettle slurry (*Urtica dioica L.*) on growth and flowering improvement of *Sedum rubrotinctum*. Legend: (A) leaves number; (B) inflorescences number; (C) vegetative weight; (D) roots weight; (E) inflorescences weight. Each value reported in the graph is the mean of three replicates \pm standard deviation. Statistical analysis performed through one-way ANOVA. Different letters for the same parameter indicate significant differences according to LSD test ($P = 0.05$).

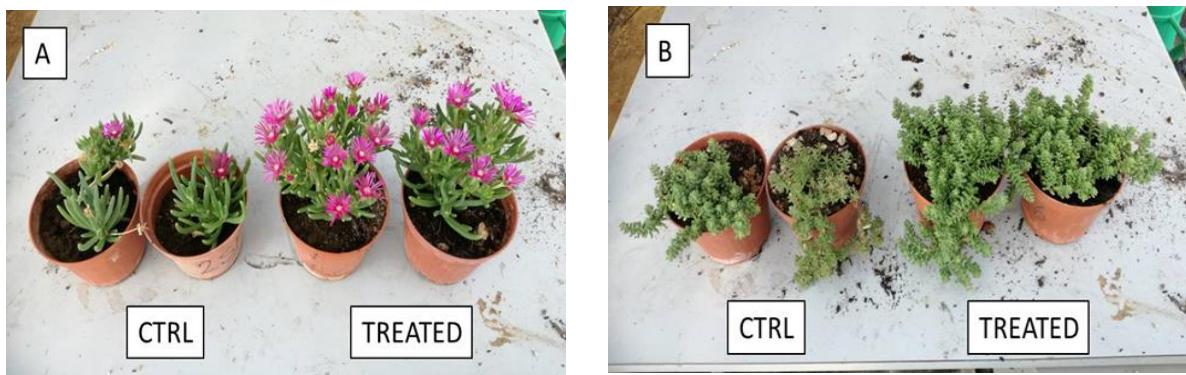


Figure 3 Effect Bat guano and Nettle slurry (*Urtica dioica L.*) on the vegetative growth and flowering of (A) *Delosperma cooperi* (B) *Sedum rubrotinctum*

Tables 1-2 show how the use of Bat guano and Nettle slurry can significantly influence the chlorophyll content and mineral absorption in plants.

In particular in *Delosperma* (Table 1), there was a significant increase in the chlorophyll content of 19.99 (BG), 19.05 (UD) compared to 16.35 (CTRL) of the control. In addition, there was also an improvement in the mineral absorption of NP in (BG) and (UD), while only in (BG) per K compared to the untreated control.

Same trend on *Sedum* (Table 2), where there was a significant increase in the chlorophyll content of 15.56 (BG), 15.30 (UD), compared to 12.32 in the control. In this case, there was an improvement in the mineral absorption of NK in (BG) and (UD), while only (BG) per P compared to the untreated control.

Table 1 Evaluation of the effect of Bat guano and Nettle slurry on the physiological and nutritional characteristics of *Delosperma cooperi*

Groups	Chlorophyll content (spad index)	N (g/Kg)	P (g/Kg)	K (g/Kg)
CTRL	16,35 c	12,42 c	1,17 c	14,21 b
BG	19,99 a	13,92 a	2,15 a	15,98 a
UD	19,05 b	13,27 b	1,60 b	14,74 b

Table 2 Evaluation of the effect of Bat guano and Nettle slurry on the physiological and nutritional characteristics of *Sedum rubrotinctum*

Groups	Chlorophyll content (spad index)	N (g/Kg)	P (g/Kg)	K (g/Kg)
CTRL	12,32 b	11,42 c	1,08 b	12,79 c
BG	15,56 a	13,42 a	1,44 a	14,92 a
UD	15,30 a	12,26 b	1,10 b	13,90 b

4. Discussion

Treatments with Nettle slurry and Bat guano resulted in a significant improvement in the growth and flowering characteristics of *Delosperma cooperi* and *Sedum rubrotinctum*. One of the main effects attributed to the use of nettle slurry on plants is precisely the stimulation due to the high concentration of nutrients (nitrogen and others). Nitrogen is a major element in plants and is assimilated in free amino acids, proteins and other nitrogenous compounds that are related to growth and development [9]. In nettle, the most important nitrogen is stored in roots and rhizomes. The use

of bat guano and nettle slurry, can stimulate the physiological and biochemical processes of plants, or act as elicitors, thus determining the production of bioactive substances. In this way, the primary and secondary metabolism can be influenced in different crops with greater resistance to various biotic and abiotic stresses [12].

Biostimulants such as Bat guano and Nettle slurry can also influence the microflora of the soil and plant growth substrates by promoting root growth and influencing nutrient availability. Bat guano contains a wide range of mineral constituents including macronutrients and micronutrients. A total of 12 elements such as aluminum, calcium, chlorine, iron, potassium, magnesium, sodium, phosphorus, sulfur, silicon and titanium were found in different species of bat guano [3]. Bat guano and Nettle slurry can positively influence vegetative and root growth, leaf development, vigour and resistance to various abiotic stresses. It can stimulate root development by increasing the ability to explore the soil and absorb nutrients. This results in a stimulation of the primary metabolism with a higher absorption of nutrients. In fact, the test showed better radical development of groups treated with Bat guano and Nettle slurry (*Urtica dioica L.*) and a significant increase in the absorption of NPK.

The diet of different species of bats can alter the nutritional profile of guano. Insect-feeding bats produce guano with a high nitrogen content. This makes it perfect for fertilization during the vegetative phase. Bats that eat fruit instead produce a guano richer in phosphorus, perfect for the flowering phase when cannabis plants need this element most. Guano is perfect as a soil improver for organic soil, added around the plant or diluted in water as a fertilizer tea. It is the backbone for any soil mix and has the particular characteristic of never burning plants, unlike most fertilizers. The benefits of guano for plant growth are numerous: It improves the consistency of the soil. If the soil is too brittle, guano can better bind its parts by increasing water retention. If it is too compact, the guano loosens its texture and helps the water to penetrate more efficiently [2]. Guano can help eliminate toxic elements from the soil, protecting the roots from a microbial point of view and at the same time fertilizing the affected plants. It favours a correct decomposition of the exhausted material. Adding guano to a compost accelerates the composting process and improves friability. It is a slow release fertilizer. Guano provides all the nutrients your plants need. It adjusts the pH by acting as a buffer on the root system. It controls nematodes and acts as a natural fungicide effective against chitin (present in insect exoskeletons). It keeps the soil crumbly and acts as a soil conditioner. Guano provides beneficial enzymes and the right microflora to maintain soil health [13]. When combined with other additives such as vermicompost and other natural fertilizers, guano helps to create blooming colonies of microorganisms in the root zones. Used as a water-diluted spray, it can protect plants from fungal attack. The long nettle macerate is used as a valuable fertilizer, thanks to the rich presence of nitrogen, and also to replenish iron and magnesium. A particularly valid use is in the cultivation in pot, since a limited soil offers less nutrients to the crops and requires more frequent fertilization [14].

Nettle slurry also has the effect of strengthening the natural defences of plants against certain pathogens, due to the salicylic acid present in the tissues of nettle: powdery mildew, peach bubble, downy mildew. It is not a resolving treatment but it helps in prevention. It is possible to use nettle slurry on the plants at the time of transplantation, wetting the roots, or as an activator of composting to be used later as part of the substrate [9].

5. Conclusion

The test showed that the use of Bat guano and Nettle slurry (*Urtica dioica L.*) can improve the growth and flowering of *Delosperma cooperi* and *Sedum rubrotinctum* plants, in particular by significantly increasing the vegetative and radical part of the plants, the number of flowers and leaves, the duration of flowering, the number and weight of inflorescences. There are also significant improvements in the physiological characteristics and mineral absorption parameters of the plants. Interesting aspects that make bat guano and nettle slurry valid alternatives to be used as plant biostimulants, when the plants are subjected to biotic and abiotic stresses, in particular transplant, water and salt stress.

Compliance with ethical standards

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Disclosure of conflict of interest

The author declares no conflict of interest.

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