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THE EFFECT OF QUIK-LINK, TRAINER AND SLAVOL BIOSTIMULATORS ON VEGETATIVE AND GENERATIVE DEVELOPMENT OF *CHRYSANTHEMUM* X *MORIFOLIUM* 'ZEMBLA WHITE' AND 'CODE GREEN'

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Abstract
Open field stock of *Chrysanthemum* x *morifolium* 'Zembla White' and 'Code Green' were divided into four groups according to the untreated control, and use of three biostimulator (Quik-Link, Trainer, Slavol). The use of Slavol gave the best results for 'Zembla White' variety, both in terms of plant and leaf sizes, as well as the flower number and diameter. For the 'Code Green', this biostimulator had positive effect on the development of green parts, but not on the number of flowers. The flowering features also differed due to the different habit of the varieties: 'Zembla White' usually developed more and larger flowers than the 'Code Green' that has smaller flowers due to its varietal characteristics. However, the former cultivar continued to develop the most and largest flowers under the influence of Slavol, while this product had not effective for 'Code Green'.

1. Introduction

Chrysanthemum species are perennial plants with a well-developed root system, string stems, pubescent leaves in various (entire or lobed) shapes, soft or leathery textures and lighter or darker green, even greyish tones. Their main ornamental value is the extremely diverse color, form and size of the compound flower head typical of the *Asteraceae* family, the inflorescence (hereinafter: flower) can be partially or completely double. The multitude of varieties is the result of crossing East Asian species for example *C.* x *morifolium*, *C. indicum, C. lavandulifolium, C. nankingense, C. chanetii* and *C. zawadskii* [6], thus, cultivated varieties are complex hybrids with different interspecific backgrounds [1, 12]. These short-day plants introduced into the European cultivation in the 18th century, firstly in the Netherlands, then in France and England. The breeding started at the beginning of the 19th century, and the arrival of new stocks from Japan in 1860 became a landmark in Western breeding works that developed and enlarged the assortment. The main goals were to create new flower colours and shapes, furthermore, to shorten the growing period and achieve earliness, as well as to increase durability and stress tolerance [13, 7].

In the international trade of cut flowers, chrysanthemum is second after rose, enjoying a major share of the ornamental plant market worldwide. One of the main reasons for the popularity is the particularly long vase life and durability of the cut flowers, which is one of the most important quality factors for consumers [11]. It is also one of the most commonly used traditional herb in East Asian regions, due to the anti-inflammatory, antipyretic and antihypertensive effects. Flowers are usually eaten as salad, and dried flower petals are used to make dietary tea for health maintenance and as an integral component of traditional Chinese medicine [16, 3].

Biostimulants are natural substances, alternative bio-fertilizers that have a beneficial effect on plant growth and development, resistance, yield and quality. Their physiological effects depend on

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their composition, as they contain various organic and mineral compounds that plants can use as metabolic products, growth regulators and nutrients [10]. The following three products were used in our experiment.

The natural microbiological fertilizer **Slavol** is a certified, universal product that combines nitrogen fixing and phosphorus mineralizing bacteria (*Azotobacter chroococcum, A. vinelandi, Derxia sp., Bacillus megaterium, B. lichenformis* and *B. subtilis*). Slavol is known for its positive effects (promoting vegetative growth and root development) on different plant production such as rosemary [4], strawberry and cauliflower [8, 9], bedding and bulbous ornamental plants [14,15]. Recommended dosage: 10-20 ml/l at 10 days interval during vegetative period [17].

The product **Trainer** is a plant-derived protein hydrolysate (mixtures of poly- and oligopeptides, amino acids, nitrogene) is advisable to spray on the foliage or to water close to the root system. It can be used to improve the nutrient uptake, stimulate growth, and increase the tolerance of plants againts abiotic stresses [2]. It is recommended to apply it as a foliar fertilizer for ornamental plant cultivation at a concentration of 2-3 mlha or 1-3 ml/l, repeated in a 10-12 day cycle [19].

Quik-Link is a biopolymer-based biostimulant, contains lateral root promoting peptides and other biopolymers such as lignosulphonates that have a high biological activity on plants. It also contains microelements like iron (1%), manganese (0.7%), zinc (0.3%), copper (0.1%) and molybdenum (0.02%). Recommended dose in horticultures: 3-5 l/ha, applied after planting. Quik-Link is allowed in organic agriculture according to the Council Regulation (EC) No. 834/2007 of 28 June 2007 [5, 18].

2. Material and method

The experiment was conducted in Subotica. The soil of the plot (where the plants grown) was analyzed and the following parameters were obtained:

- pH: 7,95 (alkaline) $CaCO_3$(%): 27,65 (high)
- Humus content (%): 4,27 (optimal) Total N (%): 0,25 (optimal)
- P_2O_5 (mg per 100 g soil): 45,2 (high) K_2O (mg per 100 g soil): 19,14 (optimal)

We used two varieties in the experiment, and 120 seedlings of each were planted. ‘Zembla White’ (Figure 1., left side) has decorative type, larger (11 cm), white flowers [20]. ‘Code Green’ (Figure 1., right side) is suitable for spray cut flower production with small (3.5 cm), green, pompon type flowers [21].

Figure 1: Flowers of ’Zembla White’ (left) and ’Code Green’ (right) (Photos: Horváth)

Rootless cuttings ordered from the Dutch company Straathof Plant B.V. were placed in 104 cell plug trays filled with Klassman TS1 peat mixture at the beginning of June, 2023. During the rooting period (2 weeks), the plants were covered with green Raschel netting and, in addition to watering twice a day (Figure 2).

Figure 2: Freshly planted cuttings (left), 2 weeks later, rooted ones on the right (Photos: Horváth)

The rooting percentage was 100%, and all rooted cuttings were planted out into the open field at the beginning of July 2023 (Figure 3). We made the beds wider than usual to avoid the biostimulant spreading of the neighboring groups, so the row spacing was 50 cm, the plant spacing was 30 cm. Irrigation was done with a drip system. Plant protection treatment was carried out three times during the entire vegetation. Firstly in August (against spider mites), and in the next two months, against aphids (with a combination of a fungicide as a preventive treatment). We used flonicamid and acetamiprid for aphids, abamectin for mites, and a product with fludioxonil + cyprodinil to prevent fungal growth. To prevent the plants falling, we used plastic carnation support net with 10x10 cm mesh. We did not pinch back the shoot tips, and plants produced flowers (in the first week of November) without any short-day treatment.

We established four groups, planting 30 plants of both *Chrysanthemum* variety in each group, for a total stock of 240 plants. The concentration and way of use were determined by the producer of the products. The groups were as follows:

1. group – Quik-Link (3 litre per ha), we applied it once by irrigation, on July 8th
2. group – Trainer (2.5 ml per 1 litre), the first treatment was also done on July 8th (by spraying onto the foliage), then repeated every 10-12 days until the appearance of flower buds, a total of 5 times
3. group – Slavol (10 ml per 1 litre), given the same method and at the same times as group 2
4. group – control (untreated)

Figure 3: The four groups of the stock, 3 weeks after planting (Photo: Horváth)

On 10th November 2023, we examined the following characteristics of the plants:

- plant height and plant diameter at the widest point (hereinafter: plant diameter, cm)
- the length and width of the largest leaf (hereinafter: leaf width, cm)
- flower number, root number (pcs)
- diameter of the largest flower (hereinafter: flower diameter, cm)

- length of the longest root (hereinafter: root length, cm)

All data were evaluated with the use of SPSS 23.0 software (IBM Corp., USA), significance differences between groups was determined by one-way analysis of variance (ANOVA). In the presence of significances, means were compared using Tukey-Kramer test at probability levels of $p \leq 0.05$, $p \leq 0.1$.

3. Results

Plant height, plant diameter

The highest group with average 116.73 cm was obtained in the untreated 'Code Green' stock. In the treated groups, the use of Slavol led to the tallest plants in both varieties ('Zembla White': 115.23 cm, 'Code Green': 116.13 cm), while the lowest values were also shown in both varieties by those treated with the Quik-Link, and this group of 'Code Green' (with an average of 92.75 cm only), differed significantly from the other groups. In terms of plant diameter, the Slavol-treated 'Zembla White' group had considerably the highest value (45.5 cm) compared to all groups (except the control part of the same variety with average 44.2 cm). The smallest plant diameters (27.96 and 37.93 cm) were obtained in the stocks of 'Code Green' treated by Quik-Link and 'Zembla White' grown with the use of Trainer, as shown on Figure 4 (vertical lines represent the standard deviations, and differing letters indicate significant differences between groups, as in the cases of all the other graphs).

Leaf length, leaf width

Regarding the length of the leaves the Slavol-treated 'Zembla White' group had the highest average (13 cm), and only this value exceeded the 12.7 cm length of the control. In the case of the 'Code Green' variety, the use of Slavol also resulted the highest leaf length (11.4 cm). The Trainer treatment led to the development of the shortest leaves in both varieties ('Zembla White': 11.23 cm, 'Code Green': 9.85 cm), the difference in the latter average was considerable compared to most of the other groups. In the case of leaf width, Slavol and Quick Link usually resulted significantly narrower leaves than the control and Trainer groups. The 'Zembla White' stock treated by Slavol had the widest (7.54 cm), while the 'Code Green' groups treated with Quik-Link and Slavol had the narrowest leaves, both with an average of 6.43 cm. (Figure 5).

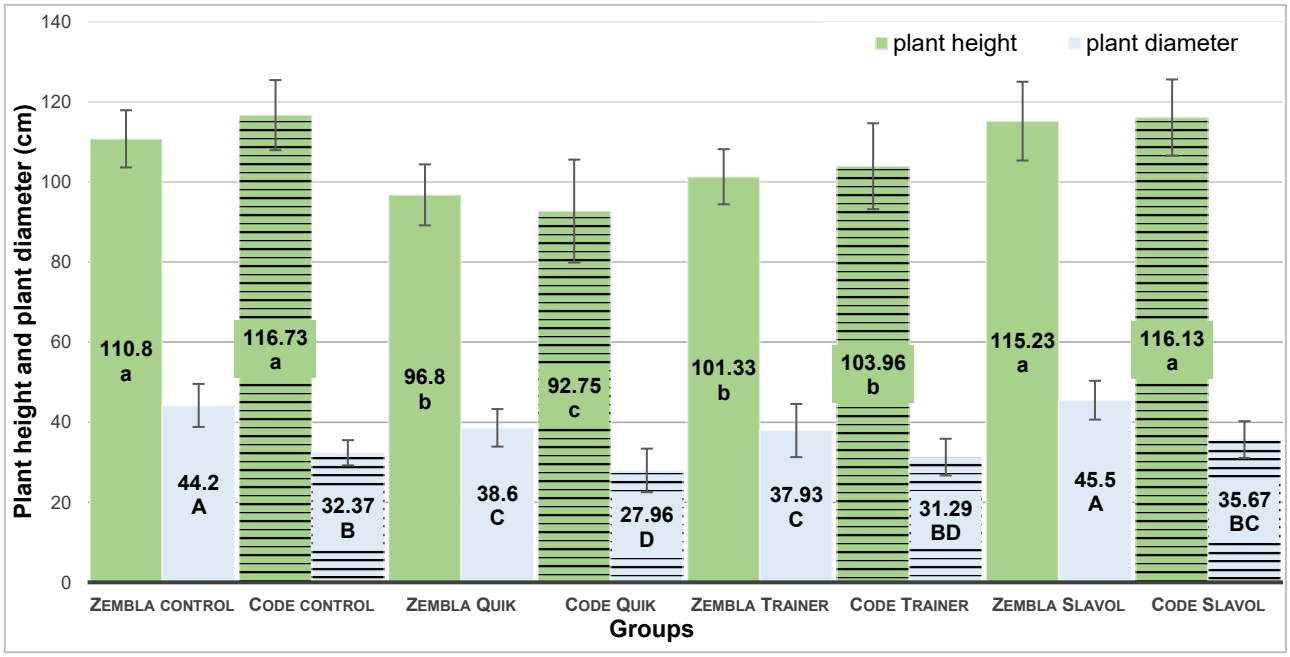

Figure 4: Plant height and plant diameter of Chrysanthemum x morifolium 'Zemba White' and 'Code Green' treated by Quik-Link, Trainer and Slavol biostimulators

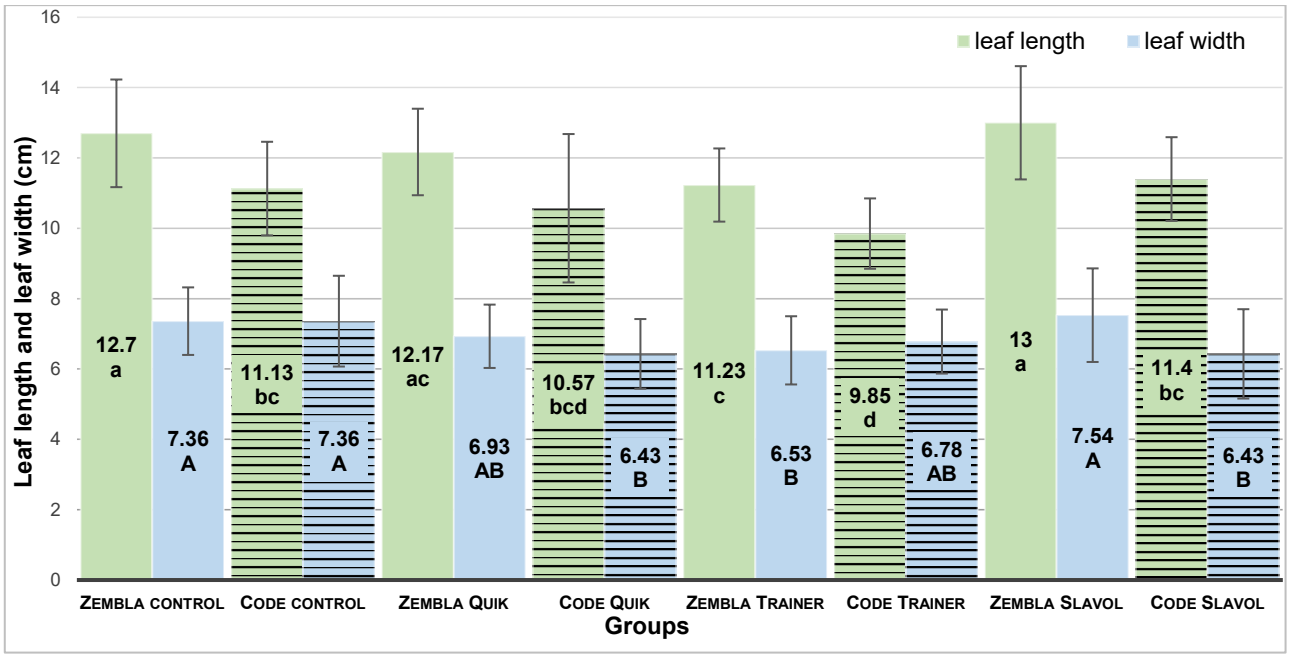

Figure 5: Leaf length and leaf width of Chrysanthemum x morifolium 'Zemba White' and 'Code Green' treated by Quik-Link, Trainer and Slavol biostimulators

Flower number, flower diameter

In the case of 'Zembla White', all biostimulants effected significantly more flowers compared to the control group, as well as Slavol resulted the most flowers (31.27 pcs) and Trainer the fewest ones (29.6 pcs). 'Code Green' developed the most flowers (22 pcs) when the latter product was applied, while the other biostimulants had no significant effect on this cultivar (and we did not experience any significant difference compared to the control, respectively). In terms of flower diameters, the Slavol-treated group of 'Zembla White' had the largest average of 10.03 cm, and this was the only group among all that produced larger flowers compared to the control. In the case of 'Code Green', the stock which treated with Trainer had the highest value (4.89 cm), while Quik-Link resulted the smallest diameter (4.69 cm), however, compared to the control, neither biostimulator affected significantly the development of flower size in this variety (Figure 6).

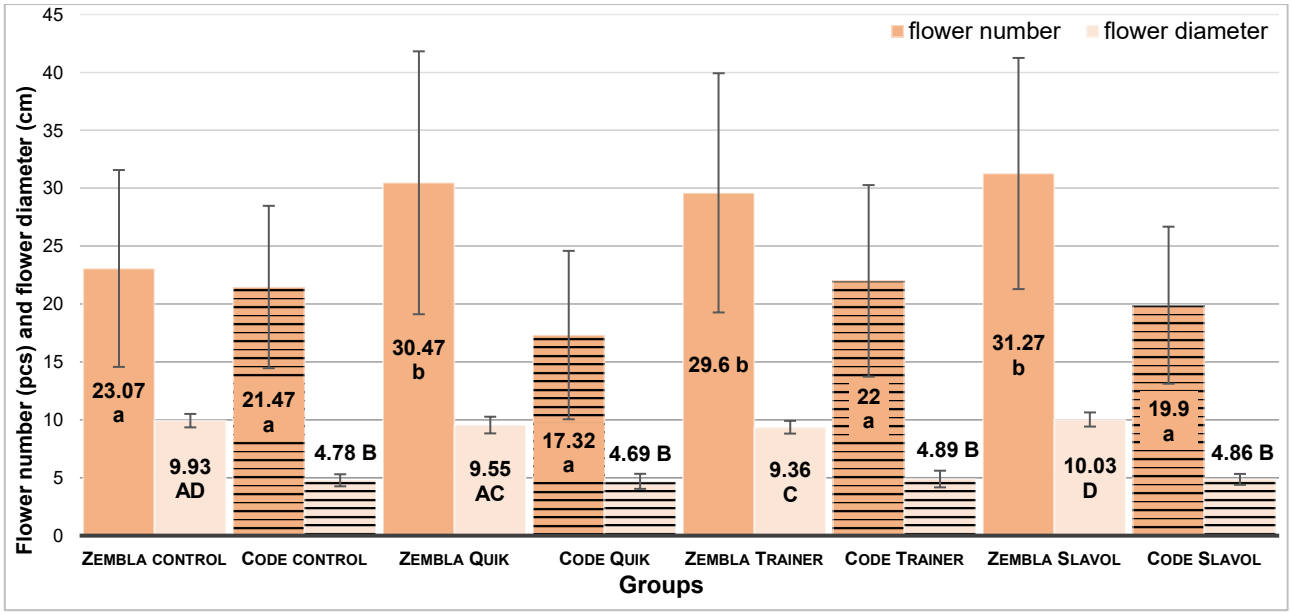

Figure 6: Flower number and flower diameter of Chrysanthemum x morifolium 'Zemba White' and 'Code Green' treated by Quik-Link, Trainer and Slavol biostimulators

Root number, root length

In the case of the root number, the groups treated by Trainer produced the most ones, with average of 6.66 pcs of ‘Zembla White’ and 4.53 of ‘Code Green’ (only in the former value the difference was significant compared to the control). The group of the latter variety grown with the use of Slavol produced the fewest (3.73 pcs) roots, while the control group of ‘Zembla White’ had the lowest average (5.3 pcs). As a result of Quik Link, the root length of ‘Zembla White’ was significantly longer (18.9 cm) compared to all other groups of this variety. The use of Quik-Link also effected the longest roots of 10.75 cm in the ‘Code Green’, but there were no remarkable differences comparing values of the stocks of this variety treated by the other biostimulators (Figure 7).

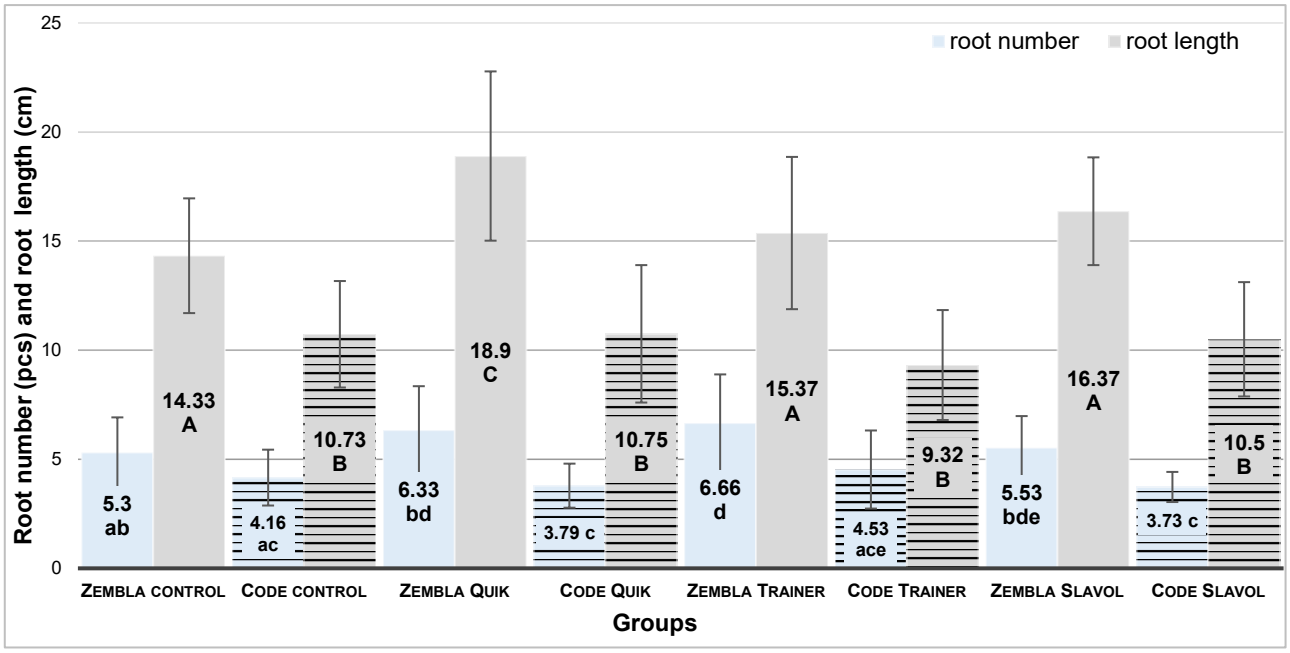

Figure 7: Root number and root length of Chrysanthemum x morifolium 'Zemba White' and 'Code Green' treated by Quik-Link, Trainer and Slavol biostimulators

4. Conclusions

Among the applied biostimulators in this trial, Slavol had the most positive effect on the vegetative parts of *Chrysanthemum*s, similarly as this microbiological product also resulted higher *Rosmarinus officinalis* plants with greater fresh and dry weight [4], and larger leaf area of strawberry [8], longer and wider leaf of cauliflower [9]. In bedding annual (*Antirrhinum majus, Alyssum maritima syn. Lobularia M., Salvia splendens)* cultivation, the highest plants were grown when Slavol was added to the water during irrigation, and the application of this liquid fertilizer also increased *Lilium* and *Gladiolus* plants’ height, especially when the latter plants’ bulbs previously soaked in 150 ml/l Slavol for 24 hours before planting [14]. The robustful ‘Zembla White’ produced higher averages of both plant height and diameter, as well as leaf size. The lowest values were observed in the groups treated with Quik-Link and Trainer for both varieties.

In the case of ‘Zembla White’, the use of Slavol led to the most and largest flowers; otherwise, in this variety, all three biostimulators significantly increased flower size compared to the untreated stock. Somewhat similar, for *Gladiolus*, also Slavol (used as 1 hour soaking treatment of bulbs) increased the lenght of the flowers and the flower number in the inflorescence, and the highest dosage (600 ml/l) resulted significantly higher values than the control group [15]. On the contrary, the variety ‘Code Green’ responded less well to the treatments, and neither the number of flowers nor the average flower diameters differed significantly compared to the control. Therefore, the effect of the products on flowering parameters may depend on the characteristics of the variety or the species.

All biostimulants had positive effect on the number and length of roots of ‘Zembla White’, the difference in length compared to the control was significant in the case of Quik Link and there was also a significant difference in root number when Trainer was applied. For melon, the total root length and root dry biomass of Quik-Link treated stocks were significantly higher in comparison with untreaed

plants [5]. In another study, Slavol resulted shorter but denser root system with considerably higher fresh and dry root weight of *Rosmarinus officinalis* [4]. Thus, the variety factor is also an important aspect here, as the rooting characteristics of 'Code Green' usually did not differ considerably compared to the untreated group.

Overall, Slavol proved to be the most effective for the *Chrysanthemum*s, especially in the case of 'Zemba White'; presumably the more favorable soil life provided by the beneficial bacteria promoted the plants' nutrient uptake and development better than if the necessary macro- and microelements had been provided with products without these microorganisms, as in the case of Trainer and Quik-Link. However, it may be worthwhile to combine these biostimulators or include additional *Chrysanthemum* varieties in such experiments, furthermore, we could also try other preparations that contain plant growth promoting rhizobacteria (PGPR). The use of products containing ingredients that may delay flowering (for example 5% nitrogen in Trainer) is only recommended when the plants are young, before they enter the generative phase.

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