Effect of vegetable protein hydrolysates on the growth of *Adromischus cooperi* and *Adromischus cristatus*

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Abstract

The aim of the experiment was to evaluate the use of vegetable protein hydrolysate on *Adromischus cooperi* and *Adromischus cristatus* to improve plant growth and quality. The two experimental groups in cultivation were: i) group without protein hydrolysates, irrigated with water and substrate previously fertilized; ii) group with protein hydrolysates, irrigated with water and substrate previously fertilized. The test showed a significant increase in agronomic parameters analysed in plants treated with hydrolysed vegetable proteins, both in *Adromischus cooperi* and *Adromischus cristatus*. In fact, all plants treated showed a significant increase in total leaf count, number of new leaves, vegetative and root weight, number of inflorescences and flowering time compared to the untreated control. The application of hydrolysed proteins in plant cultivation and in particular in succulents, allows higher quality standards of the product, higher resistance to biotic and abiotic stress, increased growth rate, very interesting aspects in the field of ornamental plants.

Keywords: Amino acids; Sustainable applications; Nutritional quality; Succulent plants, Biostimulant

1. Introduction

Protein hydrolysates are characterised by mixtures of amino acids and peptides obtained by partial, chemical, enzymatic hydrolysis of proteins of animal and vegetable origin. Normally the most used animal matrix for the production of hydrolysates is collagen, while for those of vegetable origin the biomass of legumes is used. Peptides and amino acids are attributed biostimulant activity, although the presence of carbohydrates, phytohormones, can contribute to this action [1,2]. Protein hydrolysates can directly influence the plant in terms of tolerance to abiotic stress, mineral nutrition, plant development and indirectly modify the microflora of the growing environment [3]. They can also increase nutrient availability and improve nutrient assimilation processes at the cellular level, factors related to the complexing action of amino acids and peptides [4]. There is also an increase in radical absorption which stimulates rhizogenesis and the activity of radical cell membrane transporters. Protein hydrolysates are able to improve the absorption of nitrogen, phosphorus, potassium, magnesium and iron. There is also increased crop tolerance to extreme temperatures, salinity, drought and poor light availability [5,6,7]. The use of these substances can also ensure better production quality due to increased photosynthetic activity and activation of secondary metabolism [8,9,10]. This leads to an increase in the accumulation of sugars and antioxidants in plant fruit and an increase in soluble solids. The effects of protein hydrolysates are also found on the composition and activity of rhizospheric microflora, which consequently improves soil fertility [11,12,13]. Several authors have also found that protein hydrolysates applied to leaves can activate endogenous defense mechanisms [14,15,16].

*Adromischus* includes about 30 small herbaceous species, evergreen, with habitats in Namibia and South Africa. The leaves are small, often decorative, the inflorescences have red or white pentameric flowers with summer blooming. Popular genus in cultivation, the plants prefer a well-drained soil with medium fertility and good luminosity in airy
position. Winter temperature of about 5°C., it is thoroughly watered from spring to autumn, with slight cooling in the other months; summer vegetation. Multiplication by stem and leaf cuttings. Cultivation not easy for the species: caryophyllaceus, diabolicus, fallax, humilis, nanus, phillipsiae [17].

The cultivation of these plants is not always easy, besides being slow growing, the leaves often suffer from chlorosis or water stress. The root development in pot cultivation is not optimal, this can be due to several factors such as temperatures, reduced availability of water or nutrients. These aspects also affect the development of flower inflorescences and the production of fertile seeds.

For this reason in this experiment, a vegetable protein hydrolysate was evaluated on Adromischus cooperi and Adromischus cristatus to improve plant growth and quality.

Figure 1 Detail of Adromischus cooperi (A) and Adromischus cristatus (B) in greenhouse

2. Material and methods

2.1. Greenhouse experiment and growing conditions

The experiments, started in October 2019, were conducted in the greenhouses of CREA-OF in Pescia (Pt), Tuscany, Italy (43°54′N 10°41′E) on Adromischus cooperi and Adromischus cristatus. The plants were placed in ø 12 cm pots; 60 plants per thesis, divided into 3 replicas of 20 plants each. All plants were fertilized with a controlled release fertilizer (4 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 two experimental groups in cultivation were:

- Group without protein hydrolysates (CTRL) (peat 70% + pumice 30%), irrigated with water and substrate previously fertilized;
- Group with protein hydrolysates (PRO) (peat 70% + pumice 30%), irrigated with water and substrate previously fertilized, Nutrigreen AD 2‰ (N organic: 8% p/p; N soluble organic 8% p/p; C organic: 14% p/p; total amino acids: 50% p/p; density: 1,25 g/ml; pH: 7.0 ± 0.5; electrical conductivity: 265 μS/cm)

The plants were watered 2 times a week and grown for 7 months. The plants were irrigated with drip irrigation. The irrigation was activated by a timer whose program was adjusted weekly according to climatic conditions and the fraction of leaching. On May 6, 2020, leaves number, new leaves number, the vegetative and root weight, inflorescences number and flowering time were recorded.

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 ≤ 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 agronomic parameters analysed in plants treated with hydrolysed vegetable proteins, both in *Adromischus cooperi* and *Adromischus cristatus*. In fact, all plants treated with protein (PRO) showed a significant increase in total leaf count, number of new leaves, vegetative and root weight, number of inflorescences and flowering time compared to the untreated control.

In particular in *Adromischus cooperi* (Table 1), treatment with hydrolysed vegetable protein (PRO) significantly improved leaf number, 23.80 (PRO), compared to 17.74 (CTRL). It significantly increased the number of new leaves, 5.70 (PRO) compared to 4.41 (CTRL). There was also an increase in the vegetative weight, 129.52 g (PRO) (Figure 2) compared to 118.98 g (CTRL) and 61.71 g (PRO) compared to 56.52 g (CTRL). The test also showed a significant increase in the number of inflorescences 2.83 (PRO), compared to 1.21 in untreated control and flower duration, 5.64 days in (PRO), compared to 4.59 days in (CTRL).

Also in *Adromischus cristatus* (Table 2), treatment with hydrolysed vegetable proteins (PRO) showed a significant increase in leaf number, 18.79 (PRO), compared to 15.20 in the control (CTRL). It significantly improved the number of new leaves, 5.00 (PRO) compared to 2.60 (CTRL). Also on *Acristatus* the test showed a significant increase in vegetative weight with (PRO), 119.08 g, compared to 109.72 g of (CTRL) (Figure 3) and root weight, 71.42 g (PRO), compared to 65.94 g of the control. There is also a significant increase in the number of inflorescences 3.21 (PRO), compared to 1.28 in the control and the flower duration 4.86 days (PRO) compared to 3.22 days in the untreated control.

**Table 1** Evaluation of protein hydrolysates on agronomic characters on plants of *Adromischus cooperi*

| Groups | Leaves number (n°) | New leaves number (n°) | Vegetative weight (g) | Roots weight (g) | Inflorescences number (n°) | Flowering time (days) |
|--------|--------------------|------------------------|-----------------------|-----------------|---------------------------|----------------------|
| CTRL   | 17,74 b            | 4,41 b                 | 118,98 b              | 56,52 b         | 1,21 b                    | 4,59 b               |
| PRO    | 23,80 a            | 5,70 a                 | 129,52 a              | 61,71 a         | 2,83 a                    | 5,64 a               |
| ANOVA  | ***                | **                     | ***                   | ***             | ***                       | *                    |

One-way ANOVA; n.s. – non significant; ***, *** – significant at P ≤ 0.05, 0.01 and 0.001, respectively; different letters for the same element indicate significant differences according to Tukey’s (HSD) multiple-range test (P = 0.05).

Legend: (CTRL) control; (PRO) protein hydrolysates

**Table 2** Evaluation of protein hydrolysates on agronomic characters on plants of *Adromischus cristatus*

| Groups | Leaves number (n°) | New leaves number (n°) | Vegetative weight (g) | Roots weight (g) | Inflorescences Number (n°) | Flowering time (days) |
|--------|--------------------|------------------------|-----------------------|-----------------|---------------------------|----------------------|
| CTRL   | 15,20 b            | 2,60 b                 | 109,72 b              | 65,94 b         | 1,28 b                    | 3,22 b               |
| PRO    | 18,79 a            | 5,00 a                 | 119,08 a              | 71,42 a         | 3,21 a                    | 4,86 a               |
| ANOVA  | ***                | ***                    | ***                   | ***             | ***                       | ***                  |

One-way ANOVA; n.s. – non significant; ***, *** – significant at P ≤ 0.05, 0.01 and 0.001, respectively; different letters for the same element indicate significant differences according to Tukey’s (HSD) multiple-range test (P = 0.05).

Legend: (CTRL) control; (PRO) protein hydrolysates
4. Discussion

Biostimulants are defined as substances, microorganisms or materials that can bring a benefit in plant growth. Their use on plants is growing considerably both in terms of company turnover and in terms of use on different plant species [18].

Theoretically, biostimulants result in a lower demand for nutrients to support the plant during periods of stress, while encouraging the use of more nutrients for their growth. They can help plants respond more efficiently to stress and stimulate the growth of microorganisms capable of producing useful plant molecules [19]. They are usually derived from a wide variety of biological and inorganic materials such as humic and fulvic substances, microbial fermentation products from animal or plant raw materials, macro and micro algae, industrial waste, etc. They are prepared using very different production processes [20,21,22].

They are classified according to their origin and nutritional composition and are divided into three main groups: humic substances, seaweed extracts and products containing amino acids and peptides. Protein hydrolysates, due to their composition, increase the nutrients available to plants, in particular nitrogen and iron, due to increased enzymatic activity.
They also perform chelating activities by reducing abiotic stress and may contain low molecular weight bio peptides with hormonal and defense system stimulation activity [23].

In this test, plants treated with hydrolysed vegetable proteins showed a significant increase in leaves number, vegetative and root weight, inflorescences number and flowering time in both *Adromischus cooperi* and *Adromischus cristatus*. These results may suggest that the biostimulant used contains functional compounds capable of activating certain signals in the plants that can improve the efficiency of utilization of the nutrients available for productive functions. The biostimulant used also provided amino acids and minerals that have somehow contributed to improving plant growth and quality. However, although biostimulants may contain different levels of minerals, it is necessary to compensate for the lack of all nutrients through fertilisation. The main advantage in the use of these substances is certainly to improve the absorption of minerals by both roots and leaves [24]. In numerous publications the use of these substances has led to an improvement in total root development and volume, fruit development and flowering [25,26]. These aspects are also confirmed in this experimentation conducted on succulent plants, plant species that normally resist abiotic stress better, particularly water stress, but can also benefit from the use of these products.

5. Conclusion

The test has shown how the use of hydrolysed vegetable proteins can improve the quality and development of *Adromischus cooperi* and *Adromischus cristatus*, in particular by significantly increasing the number of leaves, the number of new leaves, the vegetative and root weight, the number of inflorescences and the flowering time.

The application of hydrolysed proteins in plant cultivation and in particular in succulents allows higher quality standards of the product, higher resistance to biotic and abiotic stress, increased growth rate, very interesting aspects in the field of ornamental plants.

Compliance with ethical standards

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Disclosure of conflict of interest

The author declares no conflict of interest.

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