MULTIDISCIPLINARY SCIENCE JOURNAL

RESEARCH ARTICLE
Published Online: August 7, 2025
https://doi.org/10.31893/multiscience.2026057

Inula viscosa: A biostimulant for enhancing vegetable growth

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Abstract This study evaluated the potential of a biostimulant derived from *Inula viscosa* (fresh and dried leaves) to enhance vegetable plant growth and resistance to phytopathologies such as *Botrytis cinerea* and downy mildew. The experiments were conducted in May 2024 in CREA-OF greenhouses (Pescia, PT) on Brussels sprout and early Verona broccoli plants. Six treatment groups were tested: (i) control (water-irrigated, pre-fertilized substrate), (ii) algae, (iii) mixed microorganisms + *Inula viscosa* fresh leaves + algae, (iv) mixed microorganisms + *Inula viscosa* dried leaves + algae, (v) mixed microorganisms + herbal tea of dried *Inula viscosa* leaves + algae, and (vi) mixed microorganisms + macerated *Inula viscosa* + algae. Data were collected on November 15, 2024, including measurements of plant height, leaf number, total leaf area, root length, aerial and root biomass, and shelf life. The efficacy against *Botrytis* and downy mildew was assessed based on the number of affected plants. Treatments involving dried *Inula viscosa* leaves and herbal tea significantly improved both vegetative and root growth in both plant species. In Brussels sprouts, the INUS treatment increased vegetative growth by 46.36% and root growth by 32.46% compared to the control. In Verona broccoli, vegetative growth increased by 30.12% and root growth by 24.22% with the INUS treatment. Other treatments also enhanced plant growth, albeit to a lesser extent. Notably, the dried leaf treatment extended shelf life by 7.45% in Brussels sprouts and 6.88% in Verona broccoli. Furthermore, the INUS treatment reduced *Botrytis* incidence by 83% and downy mildew by 88.25% in Brussels sprouts. In Verona broccoli, reductions were 79.14% and 91.25%, respectively. This study highlights the potential of *Inula viscosa* as an innovative biostimulant for sustainable agriculture. Its effectiveness in promoting plant growth and controlling phytopathologies underscores its value in the development of biofertilizer products for organic farming systems.

Keywords: sustainable agriculture, organic farming, biostimulant, roots growth, rhizosphere

1. Introduction

Inula viscosa L. (also known as Dittrichia viscosa Greuter) is a perennial, bushy plant belonging to the Compositae family, which comprises over 959 genera. It is widely distributed throughout the Mediterranean region (Searchinger, 2013). Characterized by an erect, woody-based stem covered with dense foliage, *I. viscosa* typically grows to a height of 50–150 cm and emits a distinct aroma. The plant blooms in autumn, producing inflorescences composed of numerous pyramidal flower heads with golden-yellow petals and linear-lanceolate shapes. Its fruits are achenes.

Several studies have identified a wide range of bioactive compounds in various parts of *I. viscosa*, including the roots, stems, leaves, and flowers, highlighting its potential applications in the pharmaceutical, cosmetic, aromatic, and related industries (Foley et al., 2011). Traditionally, the plant has been used in folk medicine for its analgesic, anti-inflammatory, antipyretic, anthelmintic, and antifungal properties, particularly in the treatment of liver disorders (Parajuli et al., 2019; Zulfiqar et al., 2019).

While primarily considered a characteristic species of the Mediterranean region (Prisa, 2021a; Prisa, 2021b; Prisa & Gobbino, 2021), *I. viscosa* is also found in southern Europe and North Africa. Although it lacks specific habitat preferences, it is commonly observed in rural areas and along roadsides, thriving in calcareous, clayey, and often acidic soils. As a heliophilous species, it adapts well to diverse environments worldwide. Numerous research studies have highlighted the biostimulant potential of *Inula* on various vegetable and ornamental species. In particular, the application of *Inula* macerated extract has demonstrated significant improvements in both vegetative and root development of these plants (Prisa, 2021a; Prisa, 2021b). Experimental evidence suggests that the biostimulant and protective properties of *Inula* are linked to metabolites produced during maceration, which enhance the presence of beneficial soil microorganisms. These microorganisms, in turn, positively influence both vegetative and root metabolism (Prisa & Attanasio, 2022).

For many years, *I. viscosa* received limited scientific attention, but recent studies have reevaluated its ecological and agricultural significance (Mullins, 2006). The plant plays a vital role as a resource for pollinators, providing nourishment for

 Received: February 3, 2025 | Accepted: May 6, 2025

butterfly caterpillars, moths, and bees (Posmyk & Szafrańska, 2016; Kauffman et al., 2007). Its high melliferous capacity supports honey production during times when other plants are no longer in bloom, particularly from late spring to early autumn. This extended flowering period facilitates both the formation of swarms in spring and honey production in late summer and autumn (Rady et al., 2019; Prisa, 2019a).

In an effort to improve crop quality and productivity while minimizing the use of chemical fertilizers and pesticides, scientists, agronomists, and farmers are increasingly focusing on environmentally friendly solutions. Among these are biostimulants and bioprotectants derived from natural products, including secondary metabolites (Colla et al., 2017). Biostimulants help plants cope with both biotic and abiotic stresses, thereby reducing the reliance on synthetic agrochemicals (Prisa, 2020a). Numerous studies have evaluated the effects of different biostimulants on seed germination, plant growth, and overall development.

A wide range of biostimulant products is currently available on the market. For example, Auxym® (Italpollina, Rivoli Veronese, Italy), a tropical plant extract, has been shown to enhance growth and improve the mineral status of *Spinacia oleracea* (spinach) and *Solanum lycopersicum* (tomato) (Colla et al., 2017; Rouphael et al., 2018). Another commercial product, Kelpak®, derived from *Ecklonia maxima*, contains natural phytohormones such as auxins and cytokinins and has demonstrated effectiveness under various conditions (Basak, 2008). Haider et al. (2009) reported that low concentrations (0.001 mM) of calliterpenone a phyllocladane diterpenoid extracted from *Callicarpa macrophylla* Vahl stimulated the growth and branching of *Mentha arvensis* (wild mint). Similarly, aqueous garlic extracts have been found to enhance growth and hormonal activity in eggplant and snap bean (Elzaawely et al., 2018; Ali et al., 2019). Moringa leaf extracts applied to sword lily foliage improved several physiological processes, resulting in enhanced growth and extended vase life (Zulfiqar et al., 2020).

In lettuce, borage extracts increased leaf pigment content and photosynthetic activity, while reducing chlorophyll fluorescence by increasing the number of active reaction centers per cross-section (Bulgari et al., 2017). Sanchez-Gómez et al. (2016) found that vine-shoot and oak extracts significantly improved wine yield and quality by promoting the production of amino acids and volatile compounds. Additionally, moringa leaf extracts were shown to extend the shelf life and enhance the quality of avocado and citrus fruits by reducing respiration rates and limiting water loss (Adetunji et al., 2012; Tesfay & Magwaza, 2017).

2. Related Work

Research has shown that secondary metabolites from *Inula viscosa* exhibit insecticidal and antibacterial properties, while its plant extracts are effective in controlling pathogenic fungi (Prisa, 2019b; Attanasio & Prisa, 2023; Hussein et al., 2014). Several studies have highlighted the significant agro-ecological roles of *I. viscosa*, including:
i) High allelopathic activity, through the production of chemical compounds toxic to other plant species;
ii) Serving as a habitat for beneficial antagonists that combat plant pests, thereby playing a crucial role in biological control;
iii) Applications in phytoremediation, including phytoextraction (Hayat et al., 2018; Younis et al., 2018), bioaccumulation (Prisa & Attanasio, 2022), and as bioindicators (Prisa, 2023). These activities are closely associated with the plant's phenological phases.

Experimental evidence has also demonstrated that *Inula viscosa* extracts can be used to control varroa mites and *Nosema* infections in bees by incorporating the extracts into gel blocks, which induce olfactory disorientation (Culver et al., 2012; Nasir et al., 2016; Aslam et al., 2016). Research further suggests that using *Inula viscosa* as a biostimulant significantly enhances germination and growth in a wide range of plants, including *Amaranthus hypochondriacus*, *Lycopersicon esculentum*, *Daucus carota*, *Pak choi*, *Perilla frutescens*, *Petroselinum crispum*, *Plumeria frangipani*, *Spinacia oleracea*, *Lactuca sativa*, and ornamental roses (Prisa, 2019c; Prisa, 2020b). *Inula*-based biostimulants have been shown to improve water and nutrient uptake, resulting in healthier, more vigorous plants. Additionally, studies indicate that these biostimulants can protect plants from fungal pathogens such as *Pythium* spp. and *Fusarium* spp. (Prisa & Attanasio, 2023). They also enhance plant tolerance to abiotic stresses, including water and salt stress. Moreover, *Inula viscosa* exhibits allelopathic activity by releasing chemical compounds that inhibit the growth of competing plant species, potentially offering a natural method of weed control (Attanasio & Prisa, 2023).

Currently, there is limited research on the use of *Inula viscosa* as a biostimulant and plant protector in its macerated form derived from fresh and dried leaves (Prisa, 2019a; Prisa, 2019b). Therefore, further studies are warranted to develop formulations that could potentially be commercialized to enhance plant quality and defense mechanisms.

With this objective, we evaluated the potential of a biostimulant derived from *Inula viscosa* (using both fresh and dried leaves) to promote the growth of horticultural plants and improve their resistance to plant diseases, particularly *Botrytis cinerea* and downy mildew. This research aims to expand our understanding of macerate production and to explore its growth-promoting and protective effects on plants.

3. Materials and Methods

The experiments were conducted in the CREA-OF greenhouses in Pescia (PT), starting in May 2024, using Brussels sprout and early Verona broccoli plants (Figure 1).

Figure 1 Details of the growing broccoli plants and the treatment with the mix microorganisms + *Inula viscosa* + algae.

3.1. Experimental Setup

Plants were grown in pots with 30 plants per treatment, divided into three replicates of 10 plants each, transplanted in early May 2024. All plants received a slow-release fertilizer (2 kg/m³ Osmocote Pro®, 6-month release) incorporated into the growing medium at transplanting.

3.2. Treatment Groups

Six experimental groups were established:

1. Control (CTRL): Peat (70%) + pumice (20%), irrigated with water and pre-fertilized substrate.

2. Algae (AG): Peat (70%) + pumice (20%), irrigated weekly with a 1% algae solution (25 mL/plant) alongside pre-fertilized substrate.

3. Fresh leaves + microorganisms (INUF): Peat (70%) + pumice (20%), irrigated weekly with a 1% solution of algae and *Inula viscosa* fresh leaves (25 mL/plant) with microorganisms and pre-fertilized substrate.

4. Dried leaves + microorganisms (INUS): Peat (70%) + pumice (20%), irrigated weekly with a 1% solution of algae and *Inula viscosa* dried leaves (25 mL/plant) with microorganisms and pre-fertilized substrate.

5. Herbal tea of dried leaves + microorganisms (TINUS): Peat (70%) + pumice (20%), irrigated weekly with a 1% solution of algae and herbal tea from dried *Inula viscosa* leaves (25 mL/plant) with microorganisms and pre-fertilized substrate.

6. Macerated leaves + microorganisms (TINUM): Peat (70%) + pumice (20%), irrigated weekly with a 1% solution of algae and macerated *Inula viscosa* (25 mL/plant) with microorganisms and pre-fertilized substrate.

The product based on *Inula viscosa* (extract) was prepared by means of a water fermentation process. 1 kg of the plant product was mixed with 20 litres of water and fermented with a heater kept at 25°C for one week. Considering the bio-stimulating properties of the Inula-based product, it was compared with common bio-stimulants based on algae and micro-organisms used by growers and agricultural technicians, in order to verify and validate its properties.

3.3. Cultivation Practices

The plants were watered once every 3 days and grown for 8 months. They were drip-irrigated, with irrigation controlled by a timer that was adjusted weekly according to weather conditions and the leaching fraction. Climate conditions were monitored continuously (5-minute basis) during the experiment by recording radiation, relative humidity and temperature of the air through the on-site meteorological station (Dacagon Device, Pullman, WA 99163 USA). Minimum, mean and maximum daily average photosynthetic photon flux density values were 109.2, 568.3 and 750.5 mol m^{-2} s^{-1}, respectively. Mean daily global radiation averaged 21.7 MJ m^{-2} d^{-1}. Minimum, mean and maximum daily average of air temperature values were 11.6, 20.5 and 22.3 ◦C, respectively. Mean daily relative humidity of air averaged 64.5%.

3.4. Data Collection

On 15 November 2024, the following parameters were measured: plant height, number of leaves, total leaf area per plant (mm^2), primary root length (mm), biomass of the aerial and root systems, and shelf-life. Additionally, the effectiveness of plant extract, microorganism, and algae mixtures in controlling *Botrytis cinerea* and downy mildew (n° of dead plants) was evaluated based on the number of affected plants.

3.5. Statistics

The experiment followed a randomized complete block design. Data were analyzed using one-way ANOVA with the GLM univariate procedure to identify significant differences among treatments at three levels of significance ($P \leq 0.05$, 0.01, and 0.001). Mean values were compared using the LSD multiple range test ($P = 0.05$). Statistical analysis and graphical representations were performed using Costat (version 6.451) and Microsoft Excel (Office 2010). During the experiment, the assumptions of normality and homoscedasticity of the data were verified, both for the distribution and for the variance of the residuals.

4. Results and Discussion

The experiment showed that treatments based on *Inula viscosa* with dried leaves (INUS) and dried leaf tea (TINUS) significantly improved the vegetative and root growth of Brussels sprouts and broccoli grown in pots in a greenhouse (Tables 1 and 2). This was also seen in other experiments on *Amaranthus hypocondriacus* and *Petroselinum crispum*, where Inula extract improved seed germination and plant growth (Prisa, 2021; Prisa & Attanasio, 2022). Although all treatments contributed to improving plant growth, the effects of INUS and TINUS were the most pronounced. The improvements observed included an increase in plant height, number of leaves, leaf surface area, vegetative and radical biomass and root hair length (Figures 2 and 3). Increases in root and vegetative biomass have also been observed in rosa and in succulent species, although Inula has never been used in dried form in these latter cases (Prisa, 2021b). In particular, the use of dried leaves also significantly increased the shelf life of broccoli and Brussels sprouts. In addition, INUS and TINUS treatments effectively controlled plant diseases such as *Botrytis cinerea* and downy mildew, as demonstrated by a reduced number of affected seedlings. The effectiveness of Inula in controlling disease was also seen in rose plants, frangipani and pak choi (Prisa & Attanasio, 2023). In Brussels sprouts, the INUS treatment increased vegetative growth by 46.36% compared to the control and root growth by 32.46%. In Verona broccoli, on the other hand, the vegetative increase with INUS was 30.12% compared to the control and 24.22% for root growth. Other treatments also improved growth, although to a lesser extent. In particular, treatment with dried leaves prolonged the shelf life of the plants, specifically by 7.45% in INUS for Brussels sprouts and 6.88% in Verona broccoli. Furthermore, there was an 83% reduction in the incidence of botrytis (INUS vs. control) and 88.25% (INUS vs. control) for downy mildew in Brussels sprouts and 79.14% and 91.25% respectively in Verona broccoli. Although not directly evaluated, the results of this experiment confirm that Inula extracts improve plant growth and defence, probably by increasing the number and biodiversity of beneficial micro-organisms (Prisa & Attanasio, 2023; Prisa, 2019c) . One aspect to be evaluated will be to test these extracts on soil in uncontrolled conditions, to determine if Inula performs the same functions. Obviously identifying new dosages and frequencies of intervention.

Inula viscosa is commonly found in uncultivated areas such as ruins, roadsides, headlands, cliffs, and escarpments. Due to its ruggedness and adaptability, it can colonize poor, dry, and stony soils (Hemalatha et al., 2018; Sakr et al., 2018). Livestock typically avoid this plant because of its sticky leaves and strong odor. As an invasive species, *I. viscosa* often grows in degraded pastures and extensive plantations (such as vineyards, olive groves, and orchards), but it avoids arable land that is regularly cultivated. In some parts of Sardinia, *I. viscosa* has been used as a remedy for rheumatic pain (Prisa, 2020). Historically, it has been applied to treat wounds and promote hemostasis in Sicily, while fresh leaves have been used to treat excessive sweating in Tuscany. In Liguria, dried leaves are used as a tobacco substitute, and a mixture of *I. viscosa* and hay is placed in barns to deter mice.

Bees highly favor *I. viscosa* due to its abundant pollen and long blooming period. This plant produces multifloral honey in late summer and autumn, and in areas with high concentrations, it can produce monofloral honey. Several species of tephritid gall dipterans, including olive flies, attack the plant (Abdou et al., 2018; Ali et al., 2018). Olive flies serve as overwintering hosts for the polyphagous parasitoid *Eupelmus urozonus*, an effective natural antagonist of the olive fly (Hanafy et al., 2012; Shakir & Al-Rawi, 2017). Through integrated pest management, Eupelmus can be spread in uncultivated areas of olive groves, where *I. viscosa* plays a key role.

In particular, *Polistes gallicus*, a Mediterranean insect species, nests on the most developed branches of *I. viscosa* plants (El-Azim et al., 2017; Sánchez-Gómez et al., 2016). The plant's toxic costic acid has been shown to affect pests such as *Aphis nerii* and Varroa. Numerous studies have demonstrated the antifungal activity of *I. viscosa* extracts against phytopathogenic fungi, as well as its insecticidal, acaricidal, antibacterial, and cytotoxic properties (Abdou et al., 2017). Furthermore, *I. viscosa* has demonstrated biofertilizing activity in various vegetable species, particularly when combined with microorganisms and algae (Souri & Bakhtiarizade, 2019; Abbas & Akladious, 2013). The plant's biostimulant effects have also been observed in field-grown plants, where it enhances resistance to water stress and pathogenic fungi. These properties make *Inula viscosa* an effective biostimulant and a promising biocontrol agent.

Table 1 evaluation of mix microorganisms, *Inula viscosa* and algae on agronomic characters on plants of Brussels sprout.

Brussels sprout	PH (cm)	LN (n°)	TLA (mm^2)	VW (g)	RW (g)	RL (cm)	Shelf-Life (days)	BO (n° plants died)	DM (n° plants died)
CTRL	12.26 ^{f}	10.13 ^{f}	32.34 ^{f}	36.24 ^{f}	23.44 ^{f}	4.22 ^{f}	6.44 ^{f}	6.12 ^{a}	5.11 ^{a}
AG	14.34 ^{e}	12.22 ^{e}	35.31 ^{e}	37.88 ^{e}	25.32 ^{e}	5.11 ^{e}	6.96 ^{e}	5.88 ^{b}	4.12 ^{b}
INUF	17.66 ^{d}	15.33 ^{d}	36.88 ^{d}	39.22 ^{d}	27.88 ^{d}	5.66 ^{d}	7.45 ^{d}	3.99 ^{c}	2.66 ^{c}
INUS	21.66 ^{a}	18.24 ^{a}	47.46 ^{a}	47.36 ^{a}	33.46 ^{a}	6.88 ^{a}	8.45 ^{a}	1.04 ^{f}	0.60 ^{f}
TINUS	19.55 ^{b}	17.55 ^{b}	45.33 ^{b}	46.55 ^{b}	32.11 ^{b}	6.32 ^{b}	8.03 ^{b}	1.88 ^{e}	0.89 ^{e}
TINUM	18.33 ^{c}	16.21 ^{c}	41.28 ^{c}	44.28 ^{c}	30.26 ^{c}	6.06 ^{c}	7.88 ^{c}	2.33 ^{d}	1.11 ^{d}
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).
Parameters: PH = plant height (cm); LN = leaves number (cm); TLA = total leaves area (mm2); VW = vegetative weight (g); RW = roots weight (g); RL = roots length (cm); BO= plants affected or died by *Botrytis cinerea* (n°); DM= plants affected or died by Downy mildew (n°).
Treatments: CTRL=control; AG= algae; INUF= mix microorganisms + *Inula viscosa* fresh leaves+ algae; INUS= mix microorganisms + *Inula viscosa* dry leaves+ algae; TINUS= mix microorganisms + *Inula viscosa* herbal tea with dried leaves + algae; TINUM= mix microorganisms + *Inula viscosa* in maceration + algae.

Table 2 evaluation of mix microorganisms, *Inula viscosa* and algae on agronomic characters on plants of early Verona broccoli.

Verona broccoli	PH (cm)	LN (n°)	TLA (mm^2)	VW (g)	RW (g)	RL (cm)	Shelf- life (days)	BO (n° plants died)	DM (n° plants died)
CTRL	8.33 ^{f}	6.22 ^{f}	24.66 ^{f}	26.22 ^{f}	18.66 ^{f}	4.10 ^{f}	5.44 ^{f}	4.22 ^{a}	3.66 ^{a}
AG	9.11 ^{e}	6.88 ^{e}	25.24 ^{e}	26.98 ^{e}	19.21 ^{e}	4.88 ^{e}	5.92 ^{e}	4.03 ^{b}	3.11 ^{b}
INUF	10.12 ^{d}	7.12 ^{d}	26.88 ^{d}	27.33 ^{d}	20.23 ^{d}	5.11 ^{d}	6.32 ^{d}	3.77 ^{c}	2.96 ^{c}
INUS	13.21 ^{a}	9.88 ^{a}	32.32 ^{a}	31.12 ^{a}	25.22 ^{a}	7.22 ^{a}	7.88 ^{a}	0.88 ^{a}	0.32 ^{a}
TINUS	12.88 ^{b}	9.11 ^{b}	31.26 ^{b}	30.24 ^{b}	24.13 ^{b}	6.88 ^{b}	7.44 ^{b}	0.97 ^{b}	0.77 ^{b}
TINUM	10.66 ^{c}	8.66 ^{c}	30.12 ^{c}	29.66 ^{c}	23,41 ^{c}	6.34 ^{c}	7.06 ^{c}	1.45 ^{c}	1.19 ^{c}
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).
Parameters: PH = plant height (cm); LN = leaves number (cm); TLA = total leaves area (mm2); VW = vegetative weight (g); RW = roots weight (g); RL = roots length (cm); BO= plants affected or died by *Botrytis cinerea* (n°); DM= plants affected or died by Downy mildew (n°).
Treatments: CTRL=control; AG= algae; INUF= mix microorganisms + *Inula viscosa* fresh leaves+ algae; INUS= mix microorganisms + *Inula viscosa* dry leaves+ algae; TINUS= mix microorganisms + *Inula viscosa* herbal tea with dried leaves + algae; TINUM= mix microorganisms + *Inula viscosa* in maceration + algae.

Figure 2 Effect of treatment (INUS) mix microorganisms + *Inula viscosa* dry leaves+ algae on vegetative and root biomass of Verona broccoli.

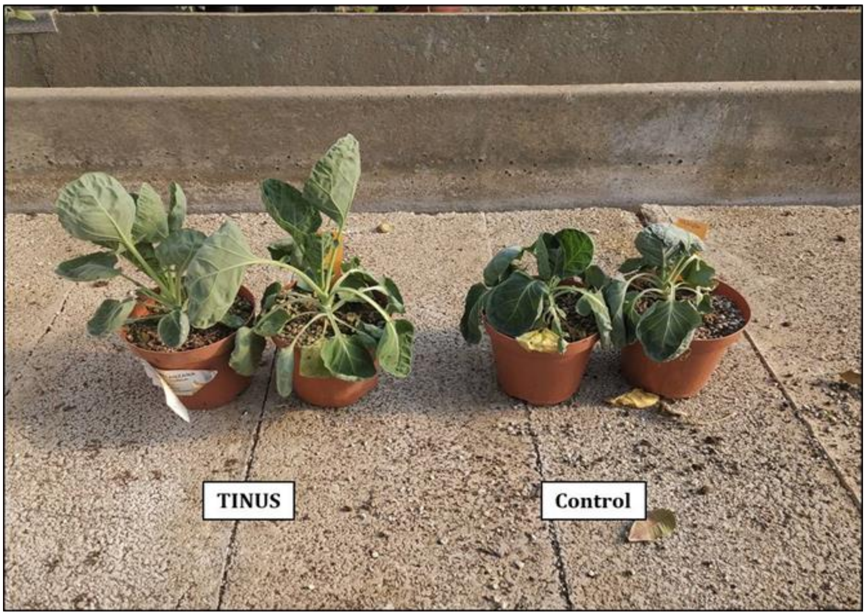

Figure 3 Effect of the treatment (TINUS) mix microorganisms + *Inula viscosa* herbal tea with dried leaves + algae on the vegetative biomass of Brussells sprout.

5. Conclusions

This study demonstrated that applying a liquid biostimulant derived from *Inula viscosa* significantly enhanced the growth, vegetative development, and root biomass of Brussels sprouts and Verona broccoli. Additionally, the treatment improved plant resistance to diseases such as *Botrytis cinerea* and downy mildew. These findings reinforce the innovative potential of *Inula viscosa*, aligning with previous studies on both vegetable and ornamental potted plant species. Given the plant's known medicinal, pollination, and biodiversity-enhancing properties, these agricultural trials are particularly valuable. They pave the way for the development of novel biofertilizer products suitable for organic and sustainable farming systems. However, further research is essential to better understand the specific effects of *Inula viscosa* across different plant species, to optimize extraction methods, and to elucidate its underlying mechanisms of action.

Acknowledgment

The research is part of the project "ZEOLINORT": Zeolitis A Chabasite Additioned to Inula Extract and Microrganisms in The Cultivation of Horticultural Plants. I would like to thank the Attanasio company for their cooperation, particularly in the person of Dr. Francesco Attanasio, who actively collaborated on the ZEOLINORT project.

Ethical considerations

Not applicable.

Conflict of Interest

The authors declare no conflicts of interest.

Funding

This research was funded by the CREA Research Centre for Vegetable and Ornamental Crops.

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