Filtrate seaweed extract as biostimulant in nursery organic horticulture

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Abstract

Many naturally-derived products are used as growth promoters or biostimulants on vegetables, even if their mechanisms of action are not completely clarified. Seaweeds and seaweed products are admissible as organic fertilizers in the Annex I of the Reg. (EC) No. 889/2008, although their biostimulant properties are not recognized in organic farming.
After a previous bioassay on maize germination (definition of optimal dose), a filtrate seaweed extract (FSE) was applied at different doses to a greenhouse organic lettuce, for assessing its biostimulant effect on vegetable growth and nutrient uptake.
At the highest dilution, FSE increased of lettuce shoot dry weight and related nutrient uptake, particularly in relation to P and K, confirming that FSE biostimulant activity acted without any nutritional effect. Thus, the FSE appeared to be a good opportunity for promoting the early-stage vegetable development for organic nursery production.

Introduction

Many naturally-derived materials are recognized as biostimulant for vegetables growth, although their mechanisms of action are not still completely clarified. In the Annex I of the Reg. (EC) No. 889/2008, the "seaweeds and seaweed products" are inserted as admissible organic fertilizers. However, some natural hormones contained in seaweeds increase plant root development (Jameson, 1993). *Ascophyllum nodosum* extracts were able to improve seedlings emergence and vigor in many crops (Demir et al., 2006), being their plant-growth promoter activity yet recognized by reference bioassay on "model" crops (Rayorathet al., 2008).
Since the biostimulant activity should be expressed at low rate of application, in the present research we applied two diluted doses of filtrate seaweed extract to a greenhouse organic lettuce, to evaluate the biostimulant effect on vegetable growth and nutrient uptake.

Material and methods

A fluid filtrate seaweed extract (FSE) from *Ascophyllum nodosum* L., produced after micro-grinding, flocculation and filtration in aqueous solution, was considered as parent materials for following dilutions. Elemental content was: 1,7% C_{org}, 0.05% N, 0.08% CaO, 0.34% SO_3, 0.40% Na_2O, 0.10% MgO, 0.8 mg/100g Fe, 3.5 mg/kg Zn, 20.0 mg/100g Cu.
Preliminary bioassay - A previous screening on *Zea mais* L. (cv. Suarta, class 300) seeds was performed by applying decreasing dilutions of FSE (1/600, 1/500, 1/400, 1/300 and 1/200 v/v) for identifying the optimal rates to put in evidence the biostimulant activity. The bioassay was performed in a growth chamber (12h/12h light/dark cycle, 28°C, 80% humidity), by adding 50 mL of each FSE solution to little pots(∅=6 cm) filled with quartz sand, where 2 maize seeds/pot were placed. Test was realized in three replicates. At third true leaf (early-stage maize cropping cycle), following parameters were measured: root length (cm), root and shoot fresh/dry weight (g), total dry biomass (g). Results were evaluated by one-way analysis of variance (F test) at $P \leq 0.05$ and the means of treatments were compared by Duncan test at $P \leq 0.05$.
Greenhouse trial - After definition of the best FSE rates, that means 1/500 and 1/250 v/v dilutions, a greenhouse trial was conducted in containers. Plantlets of about 6 cm high of *Lactuca sativa* L., var. Romana, were transplanted in 1,5 L plastic pots containing a high-fertility soil, whose main characteristics are the following: silty-loamy soil, 7.6 pH, 29.5 meq/100g CEC, 1.8 % organic matter, 0.12 % N_{tot}, 8.7 C/N.
Plantlets were sub-irrigated through saturation of pot bottom zone by saucers filled with distilled water (150 $mLH_2O \times pot^{-1}$). The fluid FSE was added at the two (1/500 and 1/250 v/v) aqueous dilutions, directly to the irrigation water, once a week for the first 4 weeks. Drip irrigation was guaranteed every 3 days. Each treatment was performed in 6 replicates, taking treatment without FSE addition as control. After 90 days from transplantation, plantlets were evaluated through the following measurements: root fresh/dry weight, shoot

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fresh/dry weight and dry matter, number of leaves, LAI, specific leaf fresh/dry weight, total dry biomass. Leaf macro and micro elements (P, K, Ca, Mg, Fe, Na, Mn, Cu, Zn, B) content and total chlorophyll were determined by simultaneous plasma emission spectrophotometer (ICP-OES) on dry matter, incinerated at 400°C for 24 hours. Results were evaluated by one-way analysis of variance (F test) at P ≤0.05 and the means of treatments were compared by the Duncan test at the P ≤0.05. Macro and micronutrients contents in vegetables were then evaluated by vector analysis which consists in the simultaneous comparison of plant yield (i.e. biomass dry weight) and nutrients content, by an integrated bi-dimensional graphic format (De Lucia et al., 2013).

Results

Preliminary bioassay – Results obtained in preliminary test on maize seeds are showed in Figure 1. At the 1/300 and 1/500 v/v dilutions, FSE showed the highest growth-promoter attitude: actually, even if the maize root elongation was not influenced, both the shoot dry weight and the total dry biomass of seedlings were significantly increased, confirming the expected biostimulant effect.

Greenhouse trial – In Table 1, lettuce yield parameters are reported. After treatments with 1/250 and 1/500 v/v of FSE, significant increases of lettuce shoot dry weight, shoot dry matter, specific leaf dry weight, root dry weight and total dry biomass were recorded, even if the 1/250 v/v FSE rate determined a significant decrease in shoot fresh weight. Lettuce LAI values were lower in all treated lettuces with respect to the untreated ones.

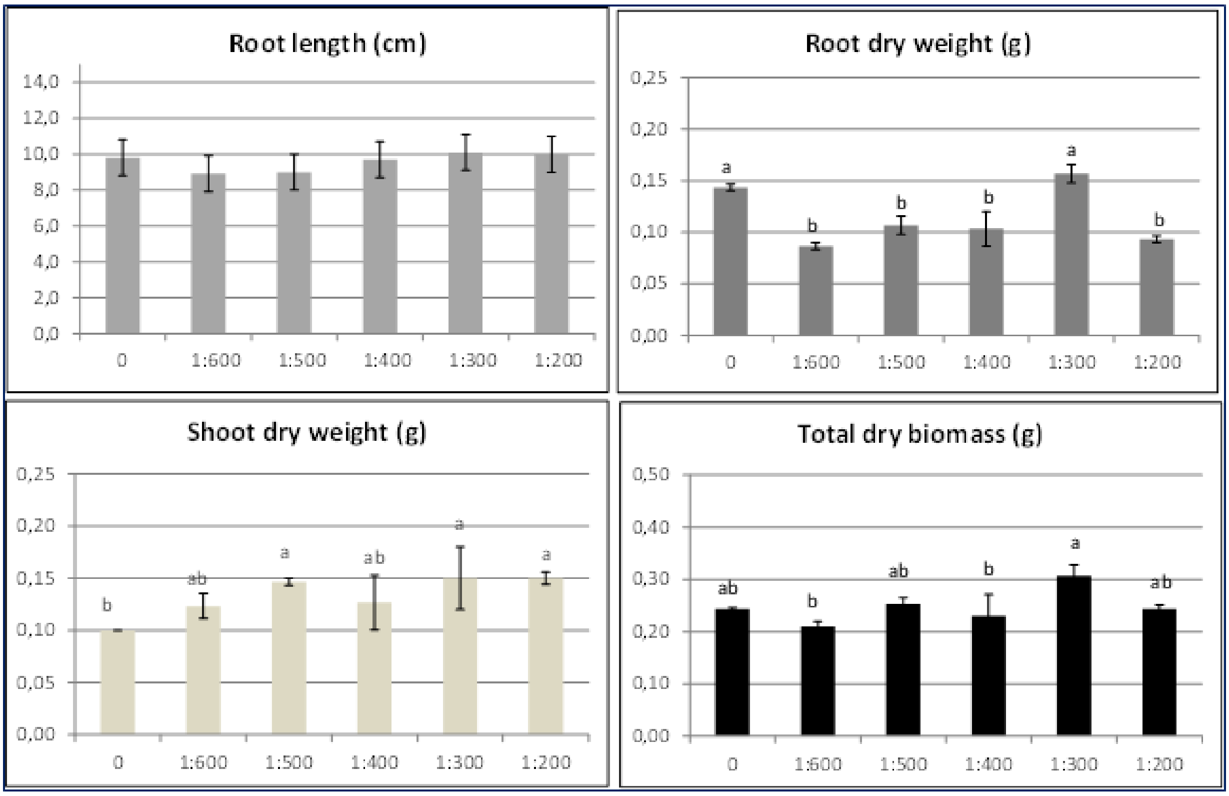

Figure 1. Statistically different seedlings parameters of maize after FSE addition (0 = untreated control; 1/600, 1/500, 1/400, 1/300 and 1/200 v/v dilutions). Different letters means significant differences at 50% (P<0.05).

Table 1. Lettuce yield parameters

Dose	Shoot fresh weight (g)	Shoot dry weight (g)	Shoot dry matter (g)	Leaf area – LAI (cm^2)	Specific leaf fresh weight (g/cm^2)	Specific leaf dry weight (g/cm^2)	Number of leaves	Root dry weight (g)	Total dry biomass (g)
Control (C)	66.14 a	4.13 b	6.26 b	1596.8 a	41.53	2.61 b	24	1.28 b	5.41 b
Low dose (L, 1/500 v/v)	63.48 a	4.58 a	7.23 a	1571.4 a	40.39	2.91 a	25	1.49 a	6.06 a
High dose (H, 1/250 v/v)	57.86 b	4.37 ab	7.57 a	1480.0 b	39.09	2.95 a	24	1.46 a	5.83 a
Significance	**	*	**	*	NS	**	NS	**	*

* significant at P<0.05; ** significant at P<0.01; *** significant at P<0.005

For evaluating lettuce nutrient use efficiency, changes in shoot dry weight and leaf nutrients content were plotted, normalizing them with respect to the same crop parameters obtained in the untreated lettuce, as percentage respect to these control values (intersection point between X/Y axes, Figure 2).
This vector analysis applied to the greenhouse lettuce showed that the low FSE rate (1/500 v/v dilution) gave the highest crop yield (+11% of shoot dry weight), with a corresponding decrease in vegetable nutrient content, being vectors of P, K, Mg, Fe and Mn below the isolines theoretical curves. Results indicate an improved nutrient use efficiency of lettuce when FSE was added at lower dose respect to the untreated control. Otherwise, the higher 1/250 v/v dose increased lettuce shoot dry weight at minor extent (+6%), and seemed to positively influence P and K nutrient use efficiency.

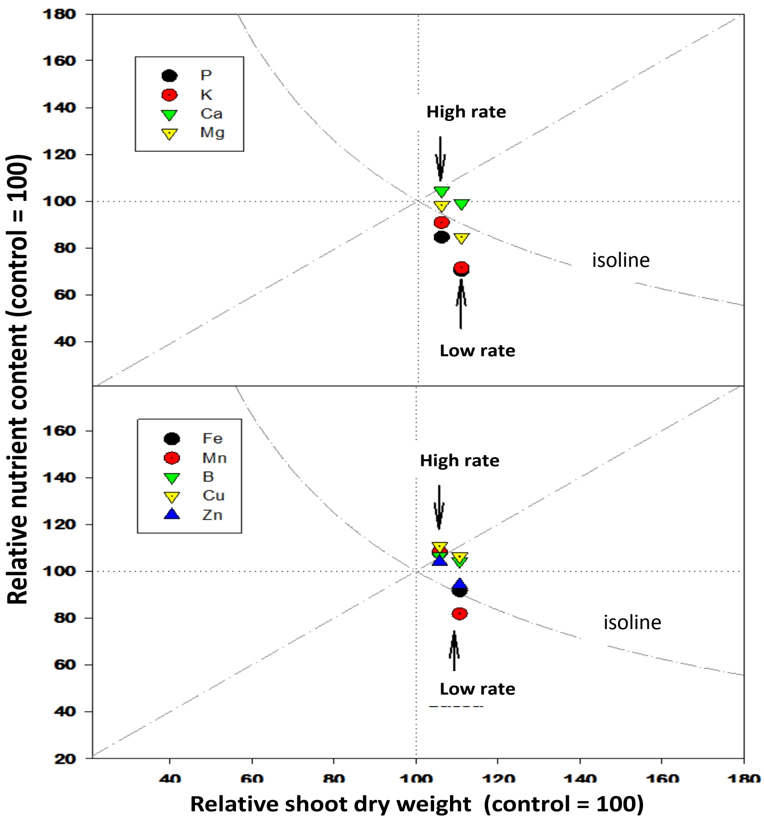

Figure 2. Vector analysis of P, K, Ca, Mg, Fe, Mn, B, Cu Zn shoot content vs. shoot dry weight in lettuce after FSE addition (High rate = 1/250 v/v; Low rate = 1/500 v/v).

Discussion

This research showed that the filtrate seaweed extract from *Ascophyllum nodosum* L*.,* in particular at the lowest rate, was able to promote the development of lettuce root apparatus and, as a consequence, the final lettuce yield. The biostimulant effect was expressed not only by the increase of lettuce LAI, but also promoting a highest crop quality, that means more compact vegetable product, more resistant to dryness on the basis of the net increase of shoot dry matter. The improved nutrient use efficiency of the lettuce at the highest dilution, particularly in relation to P and K, confirmed that the biostimulant activity of the formulate has acted without any nutritional effect: then, the FSE appears to be a good opportunity for promoting the early-stage development for organic vegetable nursery production, also taking into account its admissibility as organic fertilizers in the Annex I of the Reg. (EC) No. 889/2008.

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