SEÇÃO 3
FISIOLOGIA VEGETAL

APPLICATION METHODS OF BIOSTIMULANT ON PRODUCTION OF LETTUCE SEEDLING

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ABSTRACT: *Biostimulants have been used as an agronomic technique to optimize the production of seedlings in various crops. A study was conducted to investigate the effect of biostimulants Fertiactyl® GZ and Fertiactyl® GR on the initial growth of lettuce seedlings (Lactuca sativa L., cv. Grand Rapids). The experiment was carried out in a nursery with 50% shading, in a randomized block design with eight replications and ten plants per plot. The treatments were: (1) without biostimulant application (control); (2) biostimulant application in the seeds (BS); biostimulant application in the seeds combined with one leaf spraying (BS+BL); (4) biostimulant application in the seeds combined with two leaf spraying (BS+2BL); (5) biostimulant application with one leaf spraying (BL); and, (6) biostimulant application with two leaf spraying (2BL). Number of leaves per plant, plant height (PH), longest root length (LRL), root volume (RV), shoot dry matter (SDM), root dry matter (RDM) and RDM:SDM ratio were evaluated thirty days after sowing. The different application methods of biostimulant did not affect most of the variables measured. Biostimulant application significantly affected only the number of leaves per plant; the Fertiactyl® GR application in the seeds combined with two leaf spraying of Fertiactyl® GZ* increased the *number of leaves compared to the control treatment.*

KEY WORD: Lactuca sativa L., Fertiactyl®, seed treatment, leaf application, plant nutrition.

MÉTODOS DE APLICAÇÃO DE BIOESTIMULANTE NA PRODUÇÃO DE MUDAS DE ALFACE

RESUMO: *A aplicação de bioestimulantes pode melhorar a qualidade das mudas das hortaliças produzidas. Conduziu-se um experimento com o objetivo avaliar o efeito dos bioestimulantes Fertiactyl® GZ e Fertiactyl® GR no crescimento inicial das mudas de alface crespa (Lactuca sativa L., cv. Grand Rapids). O experimento foi realizado em viveiro com 50% de sombreamento, em um delineamento de bloco ao acaso com oito repetições e dez plantas por parcela. Os tratamentos foram: (1) cultivo da alface sem aplicação de bioestimulante (controle); (2) aplicação de bioestimulante nas sementes; (3) aplicação de bioestimulante nas sementes combinado com uma aplicação foliar; (4) aplicação de bioestimulante nas sementes combinado com duas aplicações foliares; (5) aplicação de bioestimulante com uma aplicação foliar; e, (6) aplicação de bioestimulante com duas aplicações foliares. Aos 30 dias após a semeadura foram avaliados o número de folhas por planta, altura da planta, comprimento da maior raiz, volume radicular, matéria seca das raízes e da parte áerea e relação raiz:parte aérea. Os diferentes modos de aplicação de bioestimulante não afetou significativamente a maioria das variáveis avaliadas. A aplicação de bioestimulante influenciou apenas o número de folhas por planta, de modo que a* *aplicação de Fertiactyl® GR nas sementes associado a duas pulverizações foliares de Fertiactyl® GZ aumentou o número de folhas em comparação ao tratamento controle.*

PALAVRAS-CHAVE: Lactuca sativa L., Fertiactyl®, tratamento de sementes, aplicação foliar, nutrição de planta.

INTRODUCTION

Lettuce (*Lactuca sativa* L.) is an annual specie from the Asteraceae family. The lettuce is considered as the most important vegetable in the group of leafy vegetables. This specie is almost exclusively used as a fresh vegetable in salads, but some forms are also cooked (Filgueira, 2008). Lettuce is produced commercially in many States of Brazil and is also widely grown as a vegetable in home gardens. This vegetable has been prominent among the leafy vegetables by their higher content of protein, carbohydrates, fiber, vitamin-A, vitamin-C and minerals like calcium, phosphorus, iron, potassium, manganese and sodium (Filgueira, 2008). The cultivation of lettuce can vary according to the producer and their technologies that can be applied in field and protected cultivations in the soil, substrates or hydroponic.

The use of quality seedlings is critical to providing good growth and development of plants in the field. Seedlings of better quality can be obtained with the adoption of new production techniques, which are at the same time, accessible to economic conditions of the producers (Minami, 1995). Any failure in the production process of seedlings will reflect negatively on their quality. Thus the importance of research that aims to increase plant initial growth rate.

In recent years, it has been verified, in the trade of agricultural products, the launching a series of nutritional products containing nutrients, amino acids, vitamins, growth regulators, humic and fulvic acids which are recommended to producers with the purpose to improve the quality and increase the production of cultivated species. These products can be used in various stages of plant development, including the initial phase of seedling (Bezerra et al. 2007).

Biostimulants are biologically active substances that enhance metabolisms and promote plant growth when applied in small quantities. Biostimulants include hormones, enzymes, proteins, amino acids, vitamins, microelements and other compounds. Most biostimulants are synthetic agents, whereas plant and alga extracts are also very popular (Dobromilska et al., 2009; Kunicki et al., 2010). Biostimulants can be successfully used in vegetable production to improve plant tolerance to stress factors, plant health, productivity and yielding at different growth stages. It is an environmentally sustainable method of improving plant development that reduces fertilizer and pesticide consumption. The results of biostimulant application are dependent on plant species, cultivar type, environmental factors, rate and time of application (Kunicki et al., 2010). Vernieri et al. (2002) found a positive effect of biostimulant on the growth and quality of lettuce and tomato seedlings during the cultivation period in the nursery. Bezerra et al. (2007) treated lettuce with Fertiactyl® GZ, a leaf-applied biostimulant, found a positive effect on the quality of seedlings. Amanda et al. (2009) suggested that applications of biostimulants might be used for stimulating nutrient use efficiency and improving the quality of baby leaf vegetables. Biostimulants can be soil-, seed- or leaf-applied, depending on their composition and expected results. Grabowska and Kunicki (2009) treated broccoli with Aminoplant®, a leaf-applied biostimulant, in the transplant stage and soil-applied Goëmar Goteo® before head formation with a positive effect on the crop yield.

The aim of this study was to investigate the effect of application methods of biostimulants Fertiactyl® GZ and Fertiactyl® GR on the initial growth of lettuce seedlings (*Lactuca sativa* L., cv. Grand Rapids).

MATERIAL AND METHODS

The experiment was carried out in a nursery with 50% shading, localized in the Department of Horticulture of Faculdades Integradas de Ourinhos – FIO, in Ourinhos, São Paulo, Brazil (24º55'20" S, 49º54'24" W, and 480 m a.s.l.), during the months of July and August 2013. The regional climate is relatively warm and dry. The 30-year mean annual temperature is 22.1 °C with a July minimum of 13.7 °C and a January maximum of 29.8 °C, and mean annual precipitation of 1,350 mm.

Seeds of curly lettuce (*Lactuca sativa* L., cv. Grand Rapids) were sown in 168-cell expanded polystyrene trays filled with commercial substrate composed of coconut fiber (Bioplant®). Three seeds were sown per cell, and nine days after sowing, they were thinned to one plant per cell.

Treatments consisted of six application methods of biostimulant: (1) without biostimulant application (control – water); (2) biostimulant application in the seeds (BS); biostimulant application in the seeds combined with one leaf spraying (BS+BL); (4) biostimulant application in the seeds combined with two leaf spraying (BS+2BL); (5) biostimulant application with one leaf spraying (BL); and, (6) biostimulant application with two leaf spraying (2BL). Biostimulants used were Fertiactyl® GR in the seed treatment and Fertiactyl® GZ in the leaf spraying. Fertiactyl® composition includes 13% of total nitrogen, 5% of potassium oxides, humic and fulvic acids, amino acids, glycine-betaine and zeatin. The leaf applications were performed at 12 and 20 days after sowing. The rate applied (2% of the spray volume) was defined according to the product recommendation to the lettuce crop. Leaf applications were performed with a CO_2 pressurized sprayer with 0.8 MPa working pressure capacities, equipped with flat fan nozzle, adjusted to apply 250 L ha^{-1} broth.

Lettuce seedlings were evaluated 28 days after sowing. Seedlings in all treatments were removed from the trays and washed with water to remove substrate adhered to the roots. Posteriorly, the seedlings were separated into roots and leaves. The root and shoot lengths were measured (cm $plant^{-1}$) using meter scale. Root volume (cm^3 $plant^{-1}$) was determined by water displacement using a calibrated cylinder. The number of leaves per plant was also measured. The plant material from shoot and root were dried for three days at 65±2 ºC, and then weighed.

The experiment was arranged in a randomized block design with eight replications (ten plants per trays represented one replicate). Original data were analyzed by ANOVA, and means of treatment were compared by the Tukey test at the 0.05 level of confidence. All analyses were performed using Sisvar 5.1 software for Windows (Statistical Analysis Software, UFLA, Lavras, MG, BRA).

RESULTS AND DISCUSSION

The different application methods of biostimulants (Fertiactyl® GR e Fertiactyl® GZ) did not affect plant height (PH), longest root length (LRL), root volume (RV), shoot dry matter (SDM), root dry matter (RDM) and RDM:SDM ratio (Table 1). Costa et al. (2008) evaluated the effect of different biostimulants concentrations on the production of watermelon seedlings, obtained seedlings of higher quality with Fertiactyl® GZ and Rutter® AA concentrations ranging from 0.26 to 0.52%. In the present study, same using a higher concentration of biostimulant (i.e., 2% of Fertiactyl®), it was not possible to detect improvement in the lettuce quality.

Silva et al. (2000) evaluating the biostimulant application on the root growth of ryegrass found that the use of Fertiactyl® GZ resulted in an increase of 100% in root length. Rhauthan & Schinitzer (1981) found that the product application containing humic and fulvic acids in its composition increased in 125% the root dry matter of barley. In lettuce (cultivar Babá-de-Verão) the use of humic substances in nutrient solution favored increments of 227% on shoot dry matter and 240% on root dry matter (Silva & Jablonski, 1995). In the present study the biostimulant application not favored the growth and development of lettuce seedlings (Table 1). The results presented here can be explained due to very low temperatures observed during the experiment (data not shown). Low temperatures can slow the physiological metabolism of the plants, causing the biostimulant has not effective action on plant development.

Table 1. Effect of different application modes of biostimulant on the number of leaves per plant (NLP), plant height (PH), longest root length (LRL), root volume (RV), root dry matter (RDM), shoot dry matter (SDM) and RDM:SDM ratio of curly lettuce seedlings (*Lactuca sativa* L., cv. Grand Rapids)

Application methods of biostimulant[(1)]	NLP	PH	LRL	RV	RDM	SDM	RDM:SDM ratio
	units	cm	cm	cm^3 $plant^{-1}$	----- mg $plant^{-1}$	------	
Control	5.39 b	5.56 a	10.42 a	5.31 a	52.5 a	21.5 a	2.44
BS	5.66 ab	5.62 a	10.89 a	6.00 a	53.5 a	24.2 a	2.21
BS + BL	5.65 ab	5.80 a	10.77 a	5.56 a	53.5 a	24.4 a	2.19
BS + 2BL	5.89 a	6.32 a	10.48 a	5.94 a	63.8 a	23.9 a	2.67
BL	5.61 ab	5.74 a	10.23 a	5.88 a	54.1 a	22.9 a	2.36
2BL	5.64 ab	6.05 a	10.56 a	5.69 a	60.9 a	22.9 a	2.66
C.V. (%)	4.2	9.2	8.2	16.6	16.1	17.2	16.6

(1) BS – biostimulant application in the seeds. BL – biostimulant application by leaf spraying. Mean values represented by the different letters, in each column show significant differences (Tukey test, $p < 0.05$). C.V.: coefficient of variation.

Biostimulant application significantly affected only the number of leaves per plant (Table 1). The Fertiactyl® GR application in the seeds combined with two leaf spraying of Fertiactyl® GZ increased the number of leaves compared to the control treatment. Bezerra et al. (2007) studied the effect of different biostimulants concentrations on the production of lettuce seedlings, and found higher number of leaves per plant with the Fertiacty® GZ use at the concentration of 0.75%.

CONCLUSSION

The different application methods of biostimulant (Fertiactyl® GR e Fertiactyl® GZ) did not affect plant height, longest root length, root volume, shoot and root dry matter, and root:shoot ratio of lettuce seedlings.

Biostimulant application affected the number of leaves per plant; the Fertiactyl® GR application in the seeds combined with two leaf spraying of Fertiactyl® GZ increased the number of leaves compared to the control treatment.

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