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Original article

Effect of QuitoMax® bioproduct on papaya cultivation (*Carica papaya* L.) in the nursery phase

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ABSTRACT

The present work was carried out in the central area of the National Institute of Agricultural Sciences (INCA) under covering conditions, with the objective of studying two forms of application of the QuitoMax® bioproduct, in the growth and development of papaya plants (*Carica papaya* L .) var. Maradol Roja in nursery conditions. Two factors were considered in the study, Factor A: dose of QuitoMax® applied by imbibition of seeds, with three levels (0, 0.1 and 0.2 g L^{-1} of the product) and Factor B: dose of QuitoMax® applied with foliar sprayer, with three levels (0, 0.1 and 0.2 g L^{-1} of the product), for a total of nine treatments. The plants grew in a substrate composed of soil and cow manure in the ratio 1:1 (v:v). The nursery was conducted for 60 days, with manual watering three times a week. Every 10 days the height and the number of leaves emitted were evaluated and at 60 days the aerial and root dry mass was quantified. The effect of the product was observed from 30 dag, with a stimulation of the height and the number of leaves, regardless of the dose or application time. The plants reached a maximum height between 7.70 and 7.90 cm, 10 leaves and 6.67 g of dry mass of the aerial part and 8.3 g of roots. The application of 0.1 g L^{-1} of QuitoMax® is recommended by imbibing the seed and foliar spraying at 20 dag.

Key words: manure, seedling, Chitosan

INTRODUCTION

The *Carica papaya* L. species, of the *Caricaceae* family, is native to the American tropics and one of the most cultivated fruit trees in tropical and subtropical regions of the world for fresh consumption and for its various uses in industry (1). The 70 % of the world production of this crop in recent years comes from India, Brazil, Indonesia, Nigeria and Mexico (2).

Sexual propagation or by seeds constitutes, at present, the practical and commercial means used in the propagation of papaya. To maintain the genetic purity of the crop or material used, seeds should be used from self-fertilized hermaphrodite plants, open pollinated hermaphrodite plants or from female plants fertilized by hermaphrodites (3). The establishment and management of the plant in the nursery is the first stage of the crop production process and it is essential to produce healthy and vigorous plants (4).

On the other hand, the search for new alternatives that help to reduce agricultural production costs while taking care of the environment, makes it necessary to study the possibility of using the potential that bioproducts have for plants.

In this context, phytostimulants, regardless of their nutrient content, may contain substances, compounds or microorganisms, whose functional use, when applied to plants or the rhizosphere, implies the improvement of crop development, vigor, yield and quality, by stimulating natural processes that benefit growth and responses to biotic and abiotic stress (5).

In particular, chitosan has been widely used for its biological potentialities, mainly because it has antimicrobial activity, induces defensive responses and tolerance to abiotic stress, in addition to promoting the growth and development of various species (6).

QuitoMax® liquid biostimulant, based on chitosan, works as an activator of innate resistance and physiological conditions of plants. Through preventive applications, it protects crops against potential pathogens and positively influences plant growth (7).

The present work was carried out with the objective of studying two forms of biostimulant QuitoMax® application in the growth and development of papaya plants, a study where no publication is reported in this regard under these conditions.

MATERIALS AND METHODS

An experiment was carried out with the technology of tubes or pressed specks, during the period December/2017-February/2018 in the central area of the National Institute of Agricultural Sciences (INCA) under covering conditions, using Zarán Negro Protection Mesh, which solar incidence decreases between 30–35 % (3).

Polyethylene boxes with 96 alveoli and tubes 0.140 m deep by 0.035 m in diameter were used. The boxes were placed in aerial beds (Tables) at 0.70 m from the ground for plant establishment.

The preparation of the substrate and the filling of the boxes were carried out manually, the sowing was carried out by placing a seedling per socket.

Beef manure was used as an organic source to form the substrate, from dairy 45 of the East Havana Genetic Company. The leached Red Ferrallitic soil (8) came from the areas of the Department of Agricultural Services (DSA) of INCA, taken at a depth of 0.00-0.20 m. The mixture was carried out using a soil-substrate ratio of 1:1 (v: v).

Table 1 shows the main chemical properties of the soil and the organic fertilizer used in the substrate for the production of papaya seedlings, as well as the properties resulting from the mixture of both sources.

Table 1. Main chemical properties of soil, cow manure and the substrate prepared with the mixture of both in a 1:1 ratio (v: v) and used in the production of papaya seedlings

Source	pH	MO	P	Ca	Mg	K
	Unidades	(g kg^{-1})	(mg kg^{-1})		(g kg^{-1})	
Manure	6.4	326.7	91	5.50	2.12	0.69
					cmol$_c$ kg^{-1}	
Soil	6.5	28.1	246.30	9.00	2.00	0.28
Substratum	6.7	103.5	189.10	22.50	9.00	1.07

Methods: potentiometric pH (H_2O) with soil: solution ratio of 1: 2.5 (9); OM (Organic matter) Walkley Black (10); P assimilable by extraction with H_2SO_4 0.1 N with soil: solution ratio 1:25 (11); exchangeable cations (cmolc kg^{-1}) by extraction with NH_4Ac 1 mol L^{-1} at pH 7 and determination by complexometry (Ca^{2+} and Mg^{2+}) and flame photometry (K^+) (12)

Seeds of a hermaphroditic plant of *Carica papaya* L. of the Maradol Roja variety were selected and subjected to the pre-germination process. Once pregerminated, they were sown in the tubes described and the cultural attentions recommended by the Technical Instructions for papaya cultivation were carried out (4).

The QuitoMax® bioproduct was used, which is obtained from the lobster exoskeleton, has a degree of deacetylation of 88 %, molecular mass of 1.35x10^{-5}, obtained by the National Group of Bioactive Products of the National Institute of Agricultural Sciences (7). Three doses of bioproduct 0 were used; 0.1 and 0.2 g L^{-1}; both in seed imbibition and in the foliar application, the latter being carried out 20 days after germination (dag).

The application of the bioproduct to the seeds was carried out by imbibition for half an hour, using one liter of water plus 25 or 50 mL of the product, depending on the treatment. Subsequently, seeds were placed in a pre-germination chamber. The foliar spraying was carried out using the same doses, by means of a manual sprayer, in the early hours of the morning.

Growth and development variables, height, number of leaves, dry mass of the aerial part and roots were evaluated at 60 dag, which coincided with the moment of culminating the nursery stage.

The dry mass was determined by drying in an oven with forced air circulation ten plants extracted for each treatment, at 70 ºC, until reaching constant mass values.

The foliar concentration of N, P, K was determined as a percentage of the dry mass of the aerial part of the ten plants evaluated by treatment, by means of humid digestion with H_2SO_4 + Se and colorimetric determination with the Nessler reagent for N, colorimetric determination by the blue color method with sulfomolybdic for P and flame photometry for K.

The extraction of N, P and K was calculated from the data of the dry mass of the aerial part and the concentration of each element (% N, P, K), by the following formula:

Extraction de N,P,K (mg plant-1) = [Dry mass (g) x concentration of the element in each organ (%)]*10

The experiment was developed with a completely randomized design and 10 repetitions. Two factors were considered in the study, Factor A: dose of QuitoMax® applied by seed imbibition, with three levels (0, 0.1 and 0.2 g L^{-1} of the product) and Factor B: dose of QuitoMax® applied per foliar sprayer, with three levels (0, 0.1 and 0.2 g L^{-1} of the product), for nine treatments. The data obtained were processed using the statistical package STATGRAPICS® Centurión XV [(13)].

The normality of the data and the homogeneity of the variance were checked. Subsequently, an analysis of variance was carried out in correspondence with the experimental design used.

In the cases in which significant differences were found between the treatments, the means were compared by Duncan's Multiple Range test ($p < 0.05$). In the case of germination percentage of seeds, the confidence interval of the means was calculated.

RESULTS AND DISCUSSION

Table 2 shows the effect of treatments under study on the height and number of papaya leaves, evaluated 60 days after germination. In the analysis of variance carried out, an interaction was found between the factors under study, the doses and time of QuitoMax® application in the two variables evaluated.

Table 2. Effect of three doses of QuitoMax® and two application times on the height and number of papaya leaves under nursery conditions

Treatments	Height (cm)	Number of leaves (u)
0 g L^{-1} seed–0 g L^{-1} foliar	6.85 b	7.80 c
0 g L^{-1} seed –0.2 g L^{-1} foliar	7.43 ab	9.60 a
0 g L^{-1} seed –0.1 g L^{-1} foliar	7.48 a	9.10 ab
0.2 g L^{-1} seed – 0 g L^{-1} foliar	7.77 a	8.90 abc
0.2 g L^{-1} seed –0.2 g L^{-1} foliar	7.23 ab	8.90 abc
0.2 g L^{-1} seed –0.1 g L^{-1} foliar	7.38 ab	8.30 bc
0.1 g L^{-1} seed –0 g L^{-1} foliar	7.45 ab	8.90 abc
0.1 g L^{-1} seed –0.2 g L^{-1} foliar	7.31 ab	9.70 a
0.1 g L^{-1} seed –0.1 g L^{-1} foliar	7.67 a	8.30 bc
Se $\bar{x}$	0.31*	0.37

*Means with different letters in the same column differ from each other, according to Duncan's test (P <0.05)

In the evaluation of the height of plants, which is the end of the stay of plants in the nursery was found that treatments with application of the bioproduct at doses of 0.1 g L^{-1} foliar; 0.2 g L^{-1} in the seed and 0.1 g L^{-1} in seed and foliar, exceeded the behavior of the absolute control. It shows that chitosan can stimulate plant height also in papaya cultivation. Regarding the number of leaves, the treatments that were superior to the absolute control were 0.1 and 0.2 g L^{-1} foliar and 0.1 g L^{-1} in seed and 0.2 g L^{-1} foliar.

In relation to the effect of QuitoMax® on plant height, 15 days after the germination of *Solanum lycopersicum* L. grown in a traditional seedbed on beds, a stimulation of the height of the plants was detected, with the imbibition doses of the seeds 0.1; 0.5 and 1.0 g L^{-1}, the highest dose being the one with the best performance [(14)]. The height of plants is the first visible indicator that indicates the transplantation time and although in this work a decrease in the time of stay in papaya plants in the nursery was not detected, there is no doubt that this decrease would imply economic savings in the production of postures [(14)].

In addition, a positive effect has also been found when soaking the seeds of tomato variety "Carcaman", in two concentrations, a result that agrees with those obtained in this work [(15)].

The response shown by the plants treated with QuitoMax®, in relation to the growth variables, agree with the results reported with chitosan use in HA 3819 hybrid tomato seedlings, produced in cultivation houses in the Business Base Unit of Nickel in Moa [(16)].

Regarding the evaluation of leaf number, in tomato (*Solanum lycopersicum* L.) seedlings, the influence of different doses of QuitoMax® has been found on leaf number of seedlings, the greatest effect being found with the dose of 1.0 g L^{-1}, even from 10 days after germination [(14)].

In soybean cultivation, increases in stem length, leaf number and general plant development have been reported when chitosan was applied by foliar spraying at doses of 200 mg ha^{-1} at the beginning of flowering [(17)]. These results are in relation to the increases in jasmonic acid and abscisic acid found in the tissues of plants treated with chitosan [(18)].

Table 3 shows the different doses effect and QuitoMax® application times on dry mass of papaya plants, 60 days after germination. Interaction between factors under study was found in both variables.

It can be observed that, both in the dry weight of the aerial part, and of the root, the highest values were found in both cases in treatment where the product was only applied at a dose of 0.1 g L^{-1} foliar. In the case of the aerial part, this treatment did not show differences with the treatment of only 0.2 g L^{-1} of foliar spray.

Table 3. Effect of treatments under study on the dry mass of papaya at 60 days after germination

Treatments	Dry mass aerial part (g)	Root dry mass (g)
0 g L^{-1} seed–0 g L^{-1} foliar	0.5 c	.31 bc
0 g L^{-1} seed –0.2 g L^{-1} foliar	0.66 ab	0.37 b
0 g L^{-1} seed –0.1 g L^{-1} foliar	0.77 a	0.45 a
0.2 g L^{-1} seed –0 g L^{-1} foliar	0.47 c	0.25 c
0.2 g L^{-1} seed –0.2 g L^{-1} foliar	0.58 bc	0.32 bc
0.2 g L^{-1} seed –0.1 g L^{-1} foliar	0.54 bc	0.36 b
0.1 g L^{-1} seed –0 g L^{-1} foliar	0.54 bc	0.31 bc
0.1 g L^{-1} seed –0.2 g L^{-1} foliar	0.58 bc	0.3 bc
0.1 g L^{-1} seed –0.1 g L^{-1} foliar	0.48 c	0.26 c
Se $\bar{x}$	0.05*	0.03*

*Means with different letters in the same column differ from each other, according to Duncan's test (P <0.05)

Similar results were found in tomato and pepper seedlings, with significant increases in the dry mass of aerial and root part of seedlings, which is a desirable result in the agricultural context, because it increases the quality of the plants suitable for transplantation [(14)].

It is suggested that one of the most interesting properties that chitosan has is that of a growth biostimulant; as for example, during the development of inductive response in the root system growth of plants, in root growth rate, in the advance of flowering, in the increase of crop yields; among others, although to date no clear mechanisms have been proposed to explain this effect [(19)].

Table 4 shows the total extraction of N, P and K carried out by papaya plants at 60 days after germination. For the three variables evaluated, interaction was obtained between the factors under study: dose and time of application of the QuitoMax® bioproduct.

Table 4. Effect of the treatments under study on the total extraction of nutrients (N, P and K) from papaya at 60 days after germination

Treatments	Extraction N	Extraction P	Extraction K
		(mg per plant)	
0 g L^{-1} seed– 0 g L^{-1}foliar	14.70 bc	4.00 b	21.00 bc
0 g L^{-1} seed –0.2 g L^{-1} foliar	21.70 ab	4.30 ab	24.70 ab
0 g L^{-1} seed –0.1 g L^{-1} foliar	29.30 a	6.30 a	32.00 a
0.2 g L^{-1} seed –0 g L^{-1} foliar	15.00 bc	4.00 b	21.30 bc
0.2 g L^{-1} seed –0.2 g L^{-1} foliar	18.30 bc	5.00 ab	25.00 ab
0.2 g L^{-1} seed –0.1 g L^{-1} foliar	9.70 c	5.00 ab	25.00 ab
0.1 g L^{-1} seed –0 g L^{-1} foliar	17.00 bc	3.00 b	13.70 c
0.1 g L^{-1} seed –0.2 g L^{-1} foliar	16.30 bc	5.00 ab	20.00 bc
0.1 g L^{-1} seed –0.1 g L^{-1} foliar	17.00 bc	4.30 ab	21.30 bc
Se $\bar{x}$	3.20*	0.70*	2.90*

*Means with different letters in the same column differ from each other, according to Duncan's test (P <0.05)

For the three elements evaluated, the highest value of variables was found in the treatment that only applied 0.1 g L^{-1} of the product by foliar spraying. In the case of the extraction of N, without differences with the treatment of 0.2 g L^{-1} of foliar spray; for the extraction of P, there were no differences between the treatment with 0.1 g L^{-1} foliar and 0.2 g L^{-1} foliar; 0.2 g L^{-1} in seed and foliar; 0.2 g L^{-1} seed – 0.1 g L^{-1} foliar and 0.1 g L^{-1} in seed with 0.1 and 0.2 g L^{-1} foliar. In the case of K extraction, the 0.1 g L^{-1} foliar treatment did not show differences with the 0.2 g L^{-1} foliar treatments; 0.2 g L^{-1} seed, 0.2 g L^{-1} foliar and 0.2 g L^{-1} seed – 0.1 g L^{-1} foliar.

This result is in correspondence with that obtained with the dry mass, since the absorption of nutrients is a variable directly proportional to plant dry weight. A similar result was found in peppers, reporting foliar concentration values in leaves of plants treated with the bioproduct in the optimal range for cultivation, raised by the literature [15].

In soybean cultivation and in the ornamental plant *Eustoma grandiflorum*, an effect of the application of chitosan has been found, by imbibition of the seed and by foliar spraying on the concentrations of nutrients in leaves [17,20–22].

The biostimulant effect shown in this research could be attributed to its chemical composition formed by linear polymers with structural units of 2-amino-2-deoxy-D-glucopyranose connected to each other by glucosidic bonds 1,4 [23].

On the other hand, results similar to those found in this work have been published when applying chitosan in a cucumber (*Cucumis sativus*) plantation by increasing its growth and development [24]. In this same sense, other authors report increases in tomato yield [14], as well as similar results in the height of crops such as tomato and rice (*Oryza sativa*) [25,26].

Taking into consideration the results it is shown that it is feasible, from the agronomic point of view, the application of the QuitoMax® bioproduct for the production of papaya seedlings by the tube method. It is advantageous for the producer, by obtaining larger and more vigorous plants, which is a positive aspect when carrying out the definitive plant transplant to the field.

CONCLUSIONS

- Under the conditions in which the research was carried out, the QuitoMax® bioproduct did not have a marked influence on the germination of papaya seeds.
- The QuitoMax® bioproduct had a positive impact on plant height, the number of leaves and the dry mass of the aerial part and roots, 60 days after germination, applying the dose of 0.1 g L^{-1} by seed imbibition before sowing and 0.1 g L^{-1} by foliar spraying 20 days after germination.

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