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### RESPONSE OF GARLIC CROP (*Allium sativum* L.) TO THE APPLICATION OF TWO BIOPRODUCTS IN THE SOIL AND CLIMATE CONDITION OF THE EAST CENTRAL PART OF LAS TUNAS PROVINCE, CUBA

Respuesta del cultivo del ajo (*Allium sativum* L.) a la aplicación de dos bioproductos en las condiciones edafoclimáticas del centro este de la provincia Las Tunas, Cuba

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ABSTRACT. Indiscriminate use of chemical fertilizers in the cultivation of garlic (Allium sativum L.) makes his productions unsustainable especially from the ecological point of view, hence the importance of implementing viable alternatives in their nutrition. An experimental plot was set up in areas of the farm "Los Pérez" Credit and Service Strengthened Cooperative (CCSS) "Niceto Pérez García" Las Tunas municipality, between 2011 and 2014. A design of random blocks with four treatments and six replicates was used. The treatments consisted of single and combined use of EcoMic® and Fitomas E® and a control without application. The effect of these treatments in some morphological parameters and performance (pseudostem diameter, mass of bulbs, bulbs diameter, number of cloves by bulb, mass and width of the cloves and determined performance in t ha-1). The data were subjected to analysis of variance dual classification and comparison of means. The application of all the alternatives showed positive results in the studied parameters compared to a control without fertilization at showing the feasibility of its use in the conditions of climate and soil in the center of Las Tunas province and contributes to the sustainability of these agroecosystems.

*Key words*: biofertilizers, vegetable crops, arbuscular mycorrhiza, sustainability

**RESUMEN.** Un uso indiscriminado de fertilizantes químicos en el cultivo del ajo (Allium sativum L.) hace que sus producciones sean insostenibles, sobre todo desde el punto de vista ecológico, de ahí la importancia de aplicar alternativas que contribuyan a disminuir su aplicación. Para ello se estableció una parcela experimental en áreas de la finca "Los Pérez" de la Cooperativa de Créditos y Servicios Fortalecida (CCSF) "Niceto Pérez García", del municipio Las Tunas, entre los años 2011 y 2014. Se utilizó un diseño de bloques al azar con cuatro tratamientos y seis réplicas. Los tratamientos utilizados consistieron en el uso individual y combinado de EcoMic® y Fitomas E® y un testigo sin aplicación. Se determinó el efecto de estos tratamientos en algunas variables morfológicas y de rendimiento (diámetro del pseudotallo, masa de los bulbos, diámetro de los bulbos, número de diente por bulbos, masa de los dientes, ancho de los dientes y rendimiento en t ha-1). Los datos se sometieron a análisis de varianza de clasificación doble y comparación de medias. La aplicación de todas las alternativas mostraron resultados positivos en los parámetros estudiados respecto a un testigo sin aplicación, por lo que muestra la efectividad de su utilización en las condiciones de clima y suelo en la zona centro de la provincia Las Tunas y contribuye a la sostenibilidad de esos agroecosistemas.

Palabras clave: biofertilizantes, hortaliza, micorrizas arbusculares, sostenibilidad

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#### INTRODUCTION

Agroecological initiatives aim to transform production systems agribusiness to an alternative paradigm that promotes local agriculture and domestic food production by farmers and rural and urban families from innovation, local resources and solar energy (1, 2). Among the most valuable items you can use organic farming is the use of bio-fertilizers and plant growth stimulants, in order to achieve an ecologically sustainable agricultural development (3, 4).

Among biofertilizers, it can cite the EcoMic® made from arbuscular mycorrhizae fungi (AMF) (5), which has been used successfully in a wide variety of horticultural, medicinal and woody plants (6-8). While also successfully, used, among plant growth stimulants, Fitomás E® (8, 9).

Among the crops which were made applications, in many cases of excessive forms of chemicals for pest management and fertilization, which is the garlic (*Allium sativum* L), common situation in Cuba, according to Cuban Association of Agricultural and Forestry Technicians (10). This crop is among the plant species consumed since ancient times by man and it is used for culinary and medicinal purposes (11).

In Cuba, even though fertilization of this crop is carried out, usually with mineral fertilizers, their historical yields are low (12), compared with those obtained in other countries reporting more than 10 t ha<sup>-1</sup> (13, 14). Fertilizer use in the production of garlic in Las Tunas, with doses as technical instructions (10), has allowed only to be obtained average yields of 2,2 t ha<sup>-1</sup> in the last five years, well below the needs of the population.

Therefore, in areas located in the center of the province dedicated to the cultivation of garlic, problems related to their production, leading to yields are low, although they will apply high volumes of inputs (mineral fertilizers and pesticides persist), which makes put in doubt the sustainability of these productions. For this, to evaluate the effect of applying EcoMic® and FitoMas E® and the combination of these was raised as an objective, in the agronomic performance of garlic, cultivar "Criollo" at the "Los Pérez" of Las Tunas municipality.

#### MATERIALS AND METHODS

The research was conducted at the farm "Los Pérez" belonging to the CCSF "Niceto Pérez García" community of San José, Las Tunas municipality, in a gray ochric Brown soil (15). The experiments were conducted between the months of December 2011-March 2012 and November 2013 to March 2014.

In both cases, the data of average temperature, rainfall and relative humidity, were taken from rum-records off Provincial Meteorological Center in Las Tunas (Table I).

An experimental design of randomized blocks with four treatments and six replicates was used. The treatments were:

T1- control without application

T2 - Application of EcoMic®

T3- Application FitoMas E®

T4-Application of FitoMas E® + EcoMic®

In both experiments were defined as study variables, the mass of the bulbs (g); the number of cloves per bulbs (u); the diameter of the bulbs (cm); clove mass (g); clove width (cm); pseudostem diameter (cm) at the time of harvest and yield (t ha<sup>-1</sup>).

Samples were taken at the end of the crop cycle, for which an area of 10,80 m<sup>2</sup> took 40 plants per plot were sampled randomly. The outer grooves were discarded to avoid the edge effect.

Months	Campaigns		Camp	aigns	Campaigns	
	2011-2012	2013-2014	2011-2012	2013-2014	2011-2012	2013-2014
	Mean temp	erature (°C)	Precipitati	ons (mm)	Relative mean	humidity (%)
November	-	25,6	-	30	-	78
December	23,8	25,0	29,1	16,5	79	72
January	23,2	24,1	0,0	59,9	75	73
February	24,1	24,8	15,4	17,8	73	75
March	24,6	24,8	3,4	39	68	69

 Table I. Behavior of the main climatic variables during the experimental periods

Propagules used from the previous harvest in the same productive unit were selected and they were selected so that was homogeneous in their size-no. Six rows per plot were planted. The planting distance used was  $0,50 \times 0,10 \text{ m}(5)$ . Each plot measured nine meters long and three meters wide with an area of 27 m<sup>2</sup>. To separate plots a distance of one meter left, before planting irrigation was made.

EcoMic® strain *Glomus Cubense* from the INCA was used at a concentration of 20 spores g<sup>-1</sup>, a rate of one kg per 10 kg of seed. This product is applied as seed coating mixture. The mixture prepared 24 hours before the time of planting and had an inoculum/water ratio of 2:1. The seeds after treated began to dry in the shade until sowing (16).

FitoMas E® aspersion was performed fortnightly from 30 days after planting. Three applications were made at 2,0 L ha<sup>-1</sup> (17). For this purpose, a backpack Matabi 16 liter capacity was used, with a nozzle FloodJet.

The plant breeding work, except those related to mineral fertilization were performed according to the instructional coach garlic cultivation (10). The harvest took place, when the false stalk bent and lost the stiffness (18).

An analytic scale, model Sartorius BP 310 S, with a precision 0,001 g was used to determine the mass of the bulbs and cloves. A vernier caliper was used in determining the diameter of the pseudostem and the bulb and the width and length of the clove.

For the calculation of the yield the mass of 40 heads of garlic selected per plot was calculated and with the area that they occupied the calculations were estimated to one hectare.

The data were subjected to analysis of double classification variance and means were compared using Duncan (19) to 0,05 % significance with the package-test statistic is InfoStat 2013 version (20).

#### **RESULTS AND DISCUSSION**

Pseudostem larger diameters in the 2011-2012 season (Table II) were obtained when four and two treatments were applied that do not differ from each other and in turn the latter does not differ from the results obtained with the implementation of FitoMas E® of independent way. The witness submitted a diameter significantly less than other treatments pseudostem.

Table II.	EcoMic® and FitoMas E® influence in
	the garlic pseudostem diameter, cultivar
	"Criollo", at the end of the crop cycle
	during the 2011-2012 and 2013-2014
	campaigns

	Treatments	Diameter of the pseudostem (cm)			
		Campaign 2011-2012	Campaign 2013-2014		
T1	Control without application	0,79 c	0,74 c		
Т2	EcoMic <sup>®</sup>	0,84 ab	0,79 ab		
Т3	FitoMas E®	0,81 b	0,76 b		
T4	FitoMas E <sup>®</sup> + EcoMic <sup>®</sup>	0,87 a	0,82 a		
SE $\overline{X}$		0,13	0,1611		
CV %		10,98	4,33		

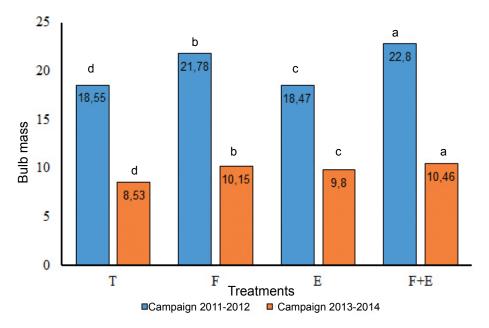
Means with different letters in the same column indicate significant differences for Duncan (P <0.05) (19)

In the 2013-2014 campaign, the trend pseudostem diameter was similar to those obtained in the prior-year, although in the latter the results were slightly higher.

Other researchers found no significant differences when analyzed pseudostem diameter garlic at 90 days after planting and using combinations FitoMas E® (1 L ha<sup>-1</sup>) with different doses of nitrogen fertilization on soil conditions and climate of the Guantanamo province (21).

The soil conditions of the experimental area, especially near neutral pH, potentiated growth, development and positive action EcoMic®. Moreover, it must bear in mind that the FitoMas E® exerts very significant physiological and metabolic effects on the biological cycle of the crop, since it is able to stimulate cell division, cell elongation and crop nutrition, which favors turn plant growth and fruit production (13). To all these effects must be added the beneficial effect of arbuscular mycorrhizal fungi (AMF) in making nutrients and water from the soil, which explains why in this and other variables analyzed, the combined use of FitoMas E® and EcoMic®, was the best treatment.

The mass bulb analysis during campaigns 2011-2012 and 2013-2014 (Figure 1) showed that treatment comprises combining EcoMic® FitoMasE® + (T4) was statistically higher than the other treatments, followed by EcoMic® application (T2) which in turn only differed from the results held with the application of FitoMas E® (T3). The lower mass of the bulbs was obtained in unfertilized treatment (T1).



Means with different letters in each column indicate significant differences for Duncan (P < 0.05) (19)

EE:  $\overline{X}$  Campaign 2011-2012: 0.13; 2013-2014 Campaign: 0.5026 T- Control without application; E-EcoMic<sup>®</sup>; F- FitoMas E<sup>®</sup>; F+E- FitoMas E<sup>®</sup>+EcoMic<sup>®</sup>

# Figure 1. Influence of EcoMic<sup>®</sup> and FitoMas E<sup>®</sup> on the mass of garlic bulbs "Criollo" during the campaigns 2011-2012 and 2013-2014

Several researchers found significant differences in the mass of the bulbs in different from this research conditions using among treatments, alternatives including bio-fertilizers and plant growth stimulants combined, alone or with mineral fertilizers (21, 22).

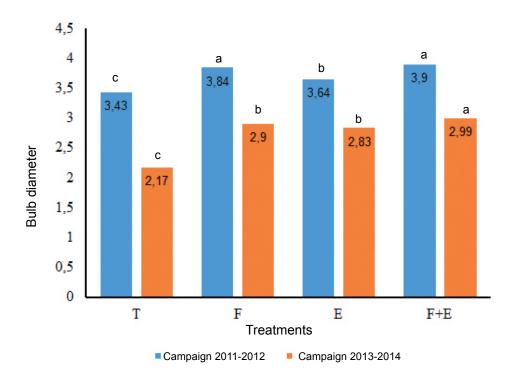
Increases in the masses of the bulbs by single or combined application of EcoMic® and FitoMas E® is corroborated by the issues raised by other researchers, who consider the AMF as a diverse and active biological community, essential to increase the sustainability of agro-ecosystems, representing the symbiosis of greater relevance in agro-ecological systems. This association allows a greater absorption of nutrients especially diffusion limited, such as P, Zn and Cu (23).

Meanwhile, Fitomás E® exerts its positive effect since it acts as transcription factor extracellular (messenger RNA stimulation) on protein synthesis and acting as ripeners as sucrose transporters through cell membranes (17). These effects favor the bulbs grow more.

The diameter of the bulb in the 2011-2012 season (Figure 2) it was observed that treatments FitoMas composed E® + EcoMic® and applying EcoMic® showed results that did not differ statistically higher and treatments 3 and 1, which were smaller in diameter. In the 2013-2014 campaign, it agreed that the combination of FitoMas E<sup>®</sup> + EcoMic<sup>®</sup> presented significantly higher than other treatments results. Applying E<sup>®</sup> FitoMas EcoMic<sup>®</sup> and independently, showed intermediate results, though they differed among themselves and with the other treatments. The treatment results obtained statistically lower was the control without application.

The effect of using two plant growth stimulants (Enerplant® and FitoMas E®), on the diameter of the bulbs of garlic cultivar Criollo was also evaluated by other authors independently and combined with different doses of N (21). Moreover, this effect was also evaluated in the cultivation Onion (*A. cepa*) with different doses of FitoMas E® (23). These authors found that as the dose is increased diameters larger bulbs are obtained.

These results affirm that the use of bio-fertilizers, coupled with the plant growth stimulants, are basic pillars for driving, with biopesticides microbial and botanical origin and application of organic fertilizers to sustainable management of the different systems agricultural production (24). In this regard, it is noted that the beneficial effect of FitoMas E® could be related to the presence in its chemistry promoting substances plant growth such as amino acids, proteins, peptides, carbohydrates and macronutrients (N, P composition, K, Ca) (17).



Means with different letters in each column indicate significant differences for Duncan (P < 0.05) (19)

SE: Campaign 2011-2012: 0.03; 2013-2014 Campaign: 0.1788

T- Control without application; E-EcoMic<sup>®</sup>; F- FitoMas E<sup>®</sup>; F+E- FitoMas E<sup>®</sup>+EcoMic<sup>®</sup>

# Figure 2. Influence of EcoMic<sup>®</sup> and FitoMas E<sup>®</sup> in the diameter of garlic bulbs "Criollo" cultivar during the campaigns 2011-2012 and 2013-2014

The diameter of the bulbs is an indicator that directly influences the quality of the product at the time of marketing in the market, as consumers prefer bulbs with predetermined quality standards, at the time of his election dimensions for both uses as food or propagation material (25, 26). Large garlic heads with few cloves are preferred, but also take into account the health status of the product (27).

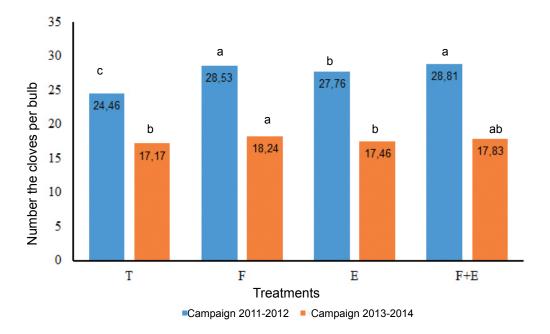
In the 2011-2012 campaign, when the combination of FitoMas E® + EcoMic® (T4) and FitoMas E® independently (T2) is applied, a number of cloves bulbs significantly higher than other treatments was obtained (Figure 3). The treatment scored lower for this variable was treatment without application (T1), whereas when EcoMic® was used independently, presented interim results between the latter and treatments 2 and 4.

In the 2013-2014 campaign, when FitoMas E® applied independently (T2) and the combination of FitoMas E® + EcoMic® (T4) an amount of cloves was obtained by superior to other treatments bulbs. In addition, this last treatment did not differ treatments 1 and 3, which were those with lower amounts of cloves bulbs.

Other researchers had lower amounts of cloves per bulbs different from this research (21, 27) conditions.

As seen in the treatments where EcoMic® used, individually and combined with FitoMas E®, the results were higher. This may be due to the positive effect of the application of EcoMic® in agricultural systems. Furthermore, FitoMas E® applied to foliage, it is rapidly absorbed and translocated without any additional expenditure of energy, which rapidly influences vegetative tissue elongation and promotes plant growth (17).

In the 2011-2012 season, the combination of FitoMas E® + EcoMic® presented statistically higher than the other treatments, relative to the mass of the cloves (Table III), followed by treatment consisting EcoMic® 2 results. Treatments 1 and 3 were statistically inferior to others, with respect to this variable and statistically similar.



Means with different letters in each column indicate significant differences for Duncan (P < 0.05) (19)

SE x : Campaign 2011-2012: 0.25; 2013-2014 Campaign: 0.1054

T- Control without inoculation ; E-EcoMic<sup>®</sup>; F- FitoMas E<sup>®</sup>; F+E- FitoMas E<sup>®</sup>+EcoMic<sup>®</sup>

# Figure 3.Influence of EcoMic<sup>®</sup> and FitoMas E<sup>®</sup> in the number of cloves per bulb of the cultivar "Criollo" during the campaigns 2011-2012 and 2013-2014

#### Table III. Influence of EcoMic<sup>®</sup> and FitoMas E<sup>®</sup> in the mass of garlic cloves cultivate "Criollo" during the campaigns 2011-2012 and 2013-2014

	Treatments	Mass of the cloves (g)		
		Campaign 2011-2012	Campaign 2013-2014	
T1	Control without application	0,59 c	0,48 d	
T2	EcoMic <sup>®</sup>	0,67 b	0,53 b	
Т3	FitoMas E <sup>®</sup>	0,60 c	0,50 c	
T4	FitoMas E® + EcoMic <sup>®</sup>	0,73 a	0,58 a	
SE	$\overline{X}$	0,13	0,0894	
ĈV	%	10,98	10,25	

Means with different letters in each column indicate significant differences for Duncan (P <0.05) (19)

In the 2013-2014 campaign, when analyzing this variable, the greater clove mass treatment was obtained, when FitoMas E® + EcoMic®, which differ statistically from the other treatments, are combined. The application of EcoMic® and FitoMas E® independently, although intermediate results obtained differ significantly among themselves and with other treatments. Treatment that related to the mass of clove statistically lower results with the control without application was obtained.

In analyzing the effect of different treatments on the clove width (Table IV) in 2011-2012 campaign, it was found that the highest values were obtained in the combined application of FitoMas E® + EcoMic® and EcoMic®, deferring significantly from other treatments. Unfertilized treatment and application of FitoMas E® were those who got the lower results on these variables, there was no significant difference among them.

In the 2013-2014 season it was observed that the wider was also obtained using FitoMas E® + EcoMic®, which differed significantly from other treatments, while applying EcoMic® not differ from the width obtained using FitoMas E®. The latter treatments not differ statistically from the results obtained with the control without application, which was the lower clove widths presented.

Generally the treatments were better yields were 4 (FitoMas E® + EcoMic®) and 2 (EcoMic®) significantly higher than those obtained with the use of FitoMas E®, who presented yields lower than the first, but significantly higher than those obtained with unfertilized treatment, which was the lower yield presented (Table V).

#### Table IV. Influence of EcoMic® and FitoMas E® in width clove garlic cultivar "Criollo" during the 2011-2012 and 2013-2014 campaigns

	Treatments	Mass of the cloves (g)		
		Campaign 2011-2012	Campaign 2013-2014	
T1	Control without application	0,57 b	0,59 c	
T2	EcoMic®	0,65 a	0,62 b	
Т3	FitoMas E®	0,60 b	0,61 bc	
T4	FitoMas E <sup>®</sup> +EcoMic <sup>®</sup>	0,68 a	0,66 a	
SE	$\overline{X}$	0,13	0,0632	
CV	%	10,98	14,76	

Means with different letters in the same column indicate significant differences for Duncan (P <0.05) (19).

The application of FitoMas  $\mathbb{E}$ ® + EcoMic® was an increase of more than 24 % yield relative to the control without application, while the application of EcoMic®, the increase was more than 20 % and the use of FitoMas E® was lower.

The highest yield in the 2013-2014 season (Table VI) was obtained with the combined use of FitoMas E® + EcoMic®, followed by applications and FitoMas E® and EcoMic® independently. The results of these three treatments were not statistically different from each other. Treatment of lower yield it was the control without application, which was statistically lower than other treatments.

As it can see the FitoMas E<sup>®</sup> + EcoMic<sup>®</sup> application also produced an increase of more than 22 % yield relative to the control without application, while the EcoMic<sup>®</sup> the application increase was over 18 % and E<sup>®</sup> FitoMas using more than 14 %.

During the 2011-2012 season, yields were higher than the average reported in our country, which is 2 t ha<sup>-1</sup> (12,13); however, the campaign 2013-2014 behaved closest to these values. This marked difference between the two campaigns, regarding the performance and other variables analyzed, it is due to weather conditions during the 2011-2012 season were more favorable for crop growth and development, compared with 2013-2014.

Generally, climatic conditions (Table III) were not ideal for growing garlic. Although temperatures behaved acceptable levels for their development (23,2 to 24,6 °C), they were not at optimum ranges for growth bulb (below 21 °C) (5).

On the other hand, the damages of *Thrips tabaci* Lin. influenced these yields since the incidence of this pest, can affect yields when infective levels are high (28). In this research during the campaign 2013-2014, infective rates up to 25 % were detected.

It should be noted that among the effects of plant growth stimulants on plants, reducing the negative consequences of adverse conditions associated, among other causes, the effects of pest infestations states (29).

All treatments had yields significantly higher than the control, which shows the positive effect of using EcoMic® and FitoMas E® in growth, development and garlic crop yields. It was also found that mycorrhizal plants grow better than the non-mycorrhizal, as they improve nutrition through the hyphae, who explore a larger volume of soil that plants do not mycorrhizal (30).

Table V. Influence of	EcoMic® y	FitoMas	E <sup>®</sup> in the	yield (	t ha-1) (	of garlic,	Criollo	cultivar"in the
campaign2011	-2012							

	Treatments	Yields (t ha <sup>-1</sup> )	Increase respect to control (t ha <sup>-1</sup> )	Increase respect to control (%)
T1	Control without application	3,65 c	0	0
T2	EcoMic®	4,46 a	0,81	22,19
Т3	FitoMas E®	3,86 b	0,21	5,75
T4	FitoMas E <sup>®</sup> + EcoMic <sup>®</sup>	4,54 a	0,89	24,38
SE $\overline{X}$		0,2994		
CV%		14,92		

Meanings with different letters in the same column indicate significant differences for Duncan(P < 0,05) (19)

	Treatments	Yields (t ha <sup>-1</sup> )	Increase respect to control (t ha <sup>-1</sup> )	Increase respect to control (%)
T1	Control without application	1,71 d	0	0
T2	EcoMic®	2,03 b	0,32	18,71
Т3	FitoMas E®	1,96 c	0,25	14,61
T4	FitoMas E <sup>®</sup> + EcoMic <sup>®</sup>	2,10 a	0,39	22,80
SE	$\overline{X}$	0,0026		
CV <sup>0</sup>	6	2,54		

# Table VI. Influence of EcoMic<sup>®</sup> and FitoMas E<sup>®</sup> in the yield (t ha-1) of the culture of garlic "Criollo" in the campaign 2013-2014

Means with different letters in each column indicate significant differences for Duncan (P < 0.05) (19)

Other authors reported yields higher than those obtained in this research up to 6,41 t ha<sup>-1</sup>, with the combined treatment of FitoMas E® + 300 kg N ha<sup>-1</sup> fertilizer, while the application form FitoMas E® independent obtained 2,34 t ha<sup>-1</sup> (21).

In the south of Las Tunas yield higher than 5 t ha<sup>-1</sup> were obtained using different plant growth stimulants (16) and in Sancti Spiritus other authors came to 6,81 t ha<sup>-1</sup>, but with *Azospirillum brasilense* applying two different cultivars to that used in this research (31). Much higher yields were also reported in other parts of the world but with the application of inorganic fertilizers (32).

The results obtained in yields, it was found that the use of EcoMic® and FitoMas E® implied an increase in garlic crop yield, in correspondence with the points made by several authors (17, 23, 24, 33-35).

This statement coincides with what enunciated by other authors who argue that the use of FitoMas E® translates into considerable savings currency, due to imports of chemical fertilizer to help minimize the toxic burden Agroecosystem, while the population healthier vegetables are offered(21).

### CONCLUSIONS

The use of FitoMas E® + EcoMic® and the EcoMic® of independent way were the treatments that best results obtained in most of the variables studied with higher yields (4,54 and 4,46 t ha<sup>-1</sup> in the campaign 2011-2012 and 2,10 and in the 2,03 t ha<sup>-1</sup> in 2013-2014) to FitoMas E® application and the control without application.

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