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Original article

Stimulating effect of thermotherapy on soybean (*Glycine max* L.) seeds for use in summer sowing

Efecto estimulante de la termoterapia sobre semillas de soya (*Glycine max* L.) para su uso en siembras de verano

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ABSTRACT: Thermotherapy has been proved to be an effective and easy to apply method to achieve a stimulation in germination and development of seedlings, in results of laboratory evaluations, based on dry heat to soybean seeds for the germination stimulation of these seeds and their vigor. The best exposure time to the treatment that enhanced germination and the characters associated to this, in nine Cuban soybean cultivars was determined. It was proved that the most effective thermotherapeutic treatment for the studied variables was the one resulting from five hours of exposure of the seeds to 50 °C. In this work, the field results are presented with such treatment of five hours of exposure to 50 °C in soybean seeds prior to sowing, under summer sowing of 2020, to evaluate the effect on morphological characters, yield components and real yield in seven *Cuban* soybean cultivars (*Glicyne max* L.). The trial of the present research was carried out in agricultural areas of the Agricultural Services Department of INCA, in the period from July 13 to November 5, 2020. The thermotherapeutic seed treatments of 50 °C during 5 hours define significant productive increases to any cultivar in summer sowing, at the time of harvesting executed to each cultivar according to its productive cycle.

Key words: exposure, germination, seedlings.

RESUMEN: La termoterapia se ha comprobado que es un método eficaz y de fácil aplicación para lograr la estimulación en la germinación y el desarrollo de las plántulas; en resultados de la aplicación de calor seco a las semillas de soya, en evaluaciones de laboratorio, se determinó el mejor tratamiento que favoreciera la germinación y los caracteres asociados a esta, en nueve cultivares cubanos de soya. Se comprobó que el tratamiento de termoterapia más efectivo para las variables estudiadas fue cinco horas de exposición de las semillas a 50 °C. En este trabajo se presentan los resultados en campo con semillas de soya a las que se aplicó dicho tratamiento de cinco horas de exposición a 50°C previo a la siembra, en el verano de 2020*,* para evaluar el efecto sobre caracteres morfológicos, componentes del rendimiento y el rendimiento real en siete cultivares de soya (*Glicyne max* L.) cubanos. El ensayo de la presente investigación se realizó en áreas agrícolas del departamento de Servicios agrícolas del INCA, en el período comprendido del 13 de julio al 5 de noviembre de 2020. Los tratamientos a la semilla de 50 °C durante 5 horas definen aumentos significativos productivos a cualquier cultivar en siembras de verano, en el momento de la cosecha ejecutada a cada cultivar según su ciclo productivo.

Palabras clave: exposición, germinación, plántulas.

INTRODUCTION

Soybean (*Glycine max* L.) is the most important oilseed worldwide, being one of the ten most promoted crops due to its great diversity of uses (1). Its production, at the international level, has shown a profound growth since 1960, which places it in the first position within the ranking of most demanded agricultural products in the world market, in the region of America and the Caribbean. By the year 2027, in which agricultural land will grow by 11 million ha, soybean will represent more than 62 % of the expansion of the region area, according to Agricultural Outlook 2018 to 2027 report (2).

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Soybeans have gained great importance in recent times within Cuban imports, due to their highly nutritious characteristics and multiple uses. As of 2010, soybean imports range from US$250 to US$400 million (3). In the Cuban industry, the following derivatives are obtained: edible oil, flour for human and animal consumption, concentrate for livestock, texturized, lecithin for the pharmaceutical industry, for meat and dairy foods, among others.

In the world, great importance is given to the quality of the seed to be used in the sowing of this oilseed. To achieve a higher yield per area, it is essential, in addition to adequate cultural techniques, the use of high quality seeds, expressed by the genetic, physical, physiological and sanitary attributes that are responsible for influencing the capacity to originate high productivity plants (2). Several factors are responsible for the loss of seed quality, mainly biotic and physiological factors (5).

The elimination or reduction of microorganisms in seeds has been efficiently achieved by chemical treatments; however, the search for alternative methods to stimulate seed development has attracted worldwide attention, since they cause less damage to the environment and human health under agroecological production, mainly those based on plant extracts, essential oils, biological control and physical treatment (3).

With the developing request and concern for more advantageous nourishments, beginning from family cultivating, without chemicals and produced in frameworks that cause less harm to the environment, thermotherapy is an elective in the treatment of natural seeds. This method comprises of uncovering the fabric to be treated to the activity of warm (damp or dry), in combination with the treatment period (temperature/time binomial), permitting the control of phytopathogenic life forms and redressing physiological variables. The rule of the treatment is based on the distinction of deadly warm focuses, that is, the temperature of water or discuss must be higher than the deadly temperature for pathogens, without harming the seeds (7).

On the other hand, thermotherapy has been proven an effective and easy to apply method to stimulate germination and seedling development. In the Dept. of Genetics and Improvement of the National Institute of Agricultural Sciences, Mayabeque, Cuba, research is being carried out with thermotherapy to obtain more vigorous seeds, in results of laboratory evaluations (8,9), based on dry heat to soybean seeds. For the stimulation of the germination of these seeds and their vigor, the best time of exposure to the treatment that favored germination and the characters associated to this was determined. It was found that the most effective thermotherapy treatment for the variables studied was the one resulting from five hours of exposure of the seeds to 50 °C, which increased the speed and germination power of the seeds, the length of the radicles and the dry mass of the radicles with respect to the control.

This work presents the results of field sowing of soybean seeds with five hours of exposure to 50 ºC, prior to summer sowing in 2020, to evaluate the effect of this treatment on morphological characters, yield components and actual yield in seven Cuban soybean cultivars (*Glicyne max* L.).

MATERIALS AND METHODS

The experimental trial of the present research was carried out in areas of the Department of Agricultural Services of the National Institute of Agricultural Sciences (INCA). In the summer period, from July 13 to November 5, 2020.

The best result of the previous laboratory evaluations (8) was used as treatment with different thermotherapy treatments based on dry heat to soybean seeds and exposure times for the germination stimulation of these seeds and their vigor. The most effective treatment was five hours of exposure of the seeds at 50 °C, prior to sowing, which increased the speed, germination power of these and the vigor of seedling development with respect to the control.

In experimental areas in the field, seeds of soybean cultivars that had undergone 50 ºC dry heat treatments for five hours were sown. The planting material used for the trials during summer planting consisted of original certified seeds of six INCA-registered cultivars (INCASoy 1, 2, 24, 27, 35 and 36), and the genetically modified cultivar CIGB CC6 (from INCASoy-27), recently registered in the National Variety Registry by CIGB and INCA collaboration. The seeds of the seven soybean cultivars used in the experiments presented values of 12 to 13 % moisture determined with the H-5 FARMPRO moisture meter at the time of sowing.

To carry out the experiment, seeds of the seven soybean cultivars were taken at random with two replicates and were divided into two lots. One of these lots of seeds of the seven soybean cultivars was placed in paper envelopes identified by cultivar and were exposed to 50 °C for five hours in the Boxun forced air circulation oven. After treatment, seeds were left to rest for 24 hours at room temperature (±25 °C) and the experiment was sown in the field with the treated and untreated seeds.

Before sowing, a mixed biofertilizer was applied to the seeds, covering them with biofertilizers based on Azofert® 200 g per 50 kg of seed and Ecomic® equivalent to 8 % of seed weighet (10).

The experimental design used was a completely randomized block design with 14 treatments (7 cultivars x heat treatment and a control of each untreated cultivar) and 3 replicates. The blocks consisted of seven plots of 10 furrows of 5 meters (m) in length separated by 0.75 m and a separation of one meter between blocks. The plots consisted of five furrows with treated seeds and five furrows with untreated seeds of each of the seven cultivars under study.

Planting was done in a red Ferrallitic soil (11) with good physical properties and good drainage. Soil preparation consisted of plowing, a disc harrow pass, a cross, another disc harrow mulching and finally furrowing at 0.75 m distance between furrows.

Seeds of the seven oilseed cultivars were sown at a rate of 23 seeds per linear meter (305.900 plants ha^{-1}) (12), who recommend sowing between 20 and 25 plants per linear meter for a population of 300,000 to 400.000 plants ha_{-1} in the spring-summer seasons in tropical countries.

The experimental area was maintained at all times with a low threshold of weeds, and control was carried out manually with hoe during the critical periods of the crop, which were: the first 15 days after planting (das), before flowering at around 30 das, and during the grain filling phase. At 20 days, a small amount of mechanical tillage was done with the tractor and a cultivator. Irrigation was carried out throughout the production cycle depending on the crop requirements; water requirements were linked to the rainfall present in the summer season of 2020 (Figure 1). The records of the climatic conditions during the field evaluation were collected from Tapaste meteorological station, San José de las Lajas, Mayabeque, located less than 400 m away. No phytosanitary treatment was applied because the crop was maintained with low incidence of phytophagous insects and diseases.

To carry out the pertinent evaluations at each moment, 10 points of one linear meter per cultivar were marked in the three central furrows of the subplots, both for the plants emerged from treated seeds and the plants emerged from control seeds (3, 4 and 3 points per furrow, respectively). For each point, 10 plants were evaluated, which were identified with a plastic sheet during the completely productive cycle and the number of plants in these marked linear meters was counted.

Variables related to plant emergence were evaluated as the main criterion for the evaluation of the effect of the seed treatment. At 10 DAS, the number of plants per linear meter was counted. Plant height and number of branches were evaluated at the flowering stage (R1) (50 % of the plants have at least one pod) using a tape graduated in millimeters for the first. The height of the first pod (cutting height) was evaluated with the same tape (mm) at the pod formation stage (R3) (50 % of the plants have at least one pod). At the physiological maturity stage (R8), yield components of ten plants were evaluated at each point in a linear meter, determining number of pods per plant, number of grains per pod and mass of grains (gr).

At harvest time, plants from each experimental area were harvested from treated and untreated plants, which were threshed and weighed differentially.

Ten replicates per cultivar were sampled and for each replicate 10 linear meters were evaluated (total area evaluated 100 linear m x cultivar) for both seed-emerged treated and control plants.

All evaluated variables were subjected to factorial analysis (7 cultivars x treatment five hours of exposure to 50 °C and a control of each untreated cultivar) and the means of the interactions and treatments were compared by Tukey's test ($P \leq 0.05$) using the IBM SPSS Version 22 statistical package.

RESULTS AND DISCUSSION

Table 1 shows the results of the factorial analysis of the plants emerged per linear meter of the cultivars under study. As can be seen, the treatment factor shows highly significant differences, the other factors and their interactions do not show significant differences with respect to the variable in question.

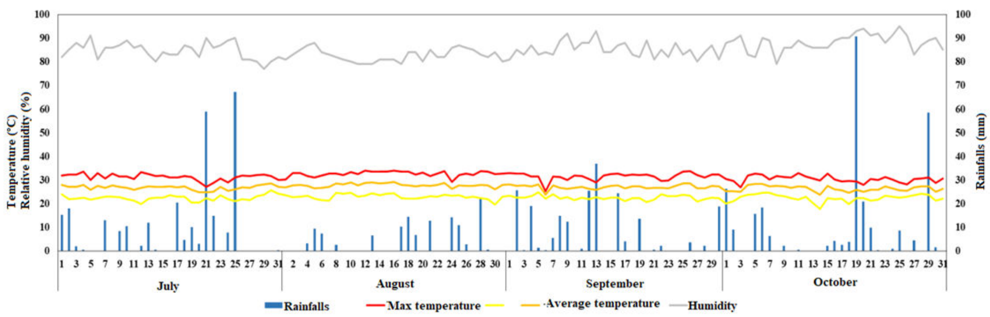
Figure 2 shows the difference in the response of the treatment factor for the emergence of plants per linear meter; it is evident how the seed treatment applied to the seven soybean cultivars had a positive influence, with respect to the untreated seeds, coinciding with the satisfactory results found by the same authors under controlled conditions.

Of the treated seeds that were sown, 92 % of the plants emerged per linear meter, 20.6 % more plants emerged in a linear meter, compared to the untreated seeds. This reflects a difference in favor of 4.76 plants m emerged among the seeds sown with the treatment, which, taken to one hectare, means an increase in the population of 63.308 more productive plants, which directly affects the significant increase in yield per area.

These results are similar to those obtained (13) for times and densities similar to those of this work with INCASoy-24 and INCASoy-27 cultivars in Villa Clara. However, in Argentina (14), using plant densities of 40 plants m, when decreasing to values close to 28 plants m, they did not find such a strong relationship with yield.

Variables related to plant growth and development

Table 2 shows the interactions of the factors under study with respect to variables related to plant growth and development of the seven soybean cultivars at flowering initiation stage (R1). As can be seen, seed treatment did not



Tapaste meteorological station, San José de las Lajas, Mayabeque

Figure 1. Behavior of climatic variables in the summer season 2020

Table 1. Factorial analysis for the variable emergence of plants per linear meter, at 10 das, in the summer period

Origin of the variation	Root mean square	Sig.
Cultivar	3.224	.272
Treatment	6797.280	.000
Replica	.502	.994
Cultivar * treatment	1.488	.709
Cultivar * replica	1.035	1.000
Treatment * replica	.445	.996
Cultivar * treatment * replica	.957	1.000
Error	2.527	

$R^2 = 0.717$

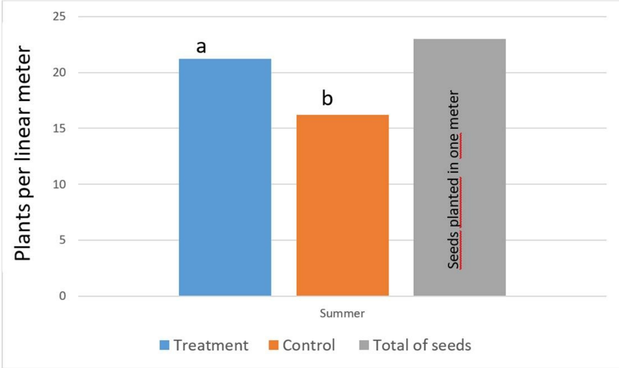


Figure 2. Analysis of plants emerged per linear meter (comparison between treatment and control) and, as a reference, the total number of seeds sown in one meter

Table 2. Factor analysis for growth and development variables of soybean plants evaluated at the flowering stage (R1)

Variables	Height plant		Branch/plants		Cutting height	
Origin of variation	Root mean square	Sig.	Root mean square	Sig.	Root mean square	Sig.
Cultivar	651618.065	0	72.218	0	72.218	0
Treatment	92716.92	0.337	1.688	0.019	261.333	0.616
Replica	77950.168	0.638	0.345	0.341	659.937	0.768
Cultivar * treatment	82020.05	0.537	0.894	0.013	357.275	0.887
Cultivar * replica	113790.99	0.253	0.31	0.449	707.974	0.947
Treatment * replica	61223.166	0.789	0.117	0.944	604.743	0.813
Cultivar * treatment * replica	94494.39	0.582	0.552	0.001	1317.758	0.114
Error	100293.32		0.306		1040.135	
	R^2= .117 (R^2 corrected = .020)		R^2 = .554 (R^2 corrected = .505)		R^2 = .387 (R^2 corrected = .320)	

affect the final growth of emerged plants, because there was no significance among treatments for plant height, number of branches and cutting height.

Table 2 shows that, although seed treatment did not affect the growth and development variables evaluated, each cultivar presented its own characteristics, with significant differences between cultivars.

Variables related to yield components

The variables that contribute to the definition of final yield are characteristics of each cultivar, as well as plant height and variables related to plant development. Table 3 shows that, with respect to heat treatment, there were no significant differences for the number of pods/plant and grains/pod, only differences between cultivars were observed, as was to be expected for both variables, i.e. seed treatment does not affect these two variables. The values shown in Figures 3 and 4 for the variables number of pods/plant and number of grains/pod correspond to the patterns that have been studied for some of these cultivars in spring-summer sowings (12,13). Similar results were presented in other cultivars in Mexico (14).

In Vera Cruz (15) similar results were presented, being the populations of 300 to 400 thousand plants ha^{-1}, those that presented the best results of yields and number of pods and grains responded to the characteristics of the cultivars, little influenced by the different sowing densities.

INCASoy-24 cultivar (Figure 3) had the highest number of pods per plant, which is a variable with yield potential.

In the number of grains per pod (Figure 4), INCASoy-27 and the transformed cultivar CIGBCC6 showed the best results, followed by INCASoy-24.

Grain yield (t ha^{-1})

Table 4 shows the factorial analysis for yield, where significant differences are reflected in terms of treatment, which shows that there were marked differences between plants with seed treatments and plants without seed treatments (control).

The seed treatment significantly increased the number of vigorous plants in the hectare, with a significant increase in yield with plants similar in number of grains per plant and weight of grains to the untreated plants, so the effect is due to the significant result of the increase in plant density per area.

Figure 5 shows that the plants obtained from treated seeds achieved almost one-third higher yields than the untreated ones. Figure 6 shows that the cultivars INCASoy-36 and INCASoy-2 presented the best yields among the seven cultivars evaluated (3.2 and 3.1 t ha^{-1}, respectively), the potential of these cultivars is similar to those found in comparative evaluations in spring-summer sowings of different years (16-20).

Table 3. Factorial analysis of the variables number of pods per plant and number of grains/pod at the physiological maturity stage (R8)

Variables	Pod number/plant		Grains per pods	
Origin of variation	Root mean square	Sig.	Root mean square	Sig.
Cultivar	16359.008	0	21.583	0
Treatment	35.363	0.07	29.768	0.102
Replica	7.228	0.733	0.97	0.894
Cultivar * treatment	10.317	0.44	2.786	0.24
Cultivar * replica	5.726	0.995	0.977	0.999
Treatment * replica	14.863	0.19	2.508	0.28
Cultivar * treatment * replica	8.115	0.88	0.931	0.999
Error	10.731		2.059	
	R^2 = .887 (R^2 corrected = .864)		R^2 = .108 (R^2 corrected = .010)	

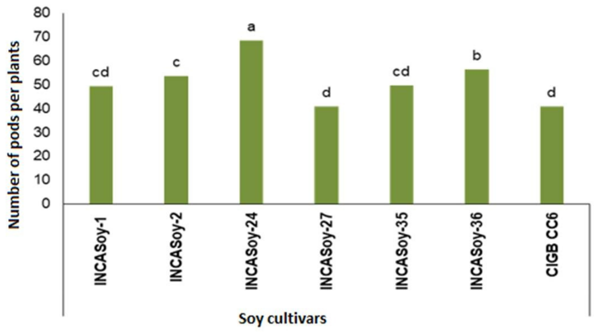


Figure 3. Mean values of the number of pods per plant of the seven cultivars studied

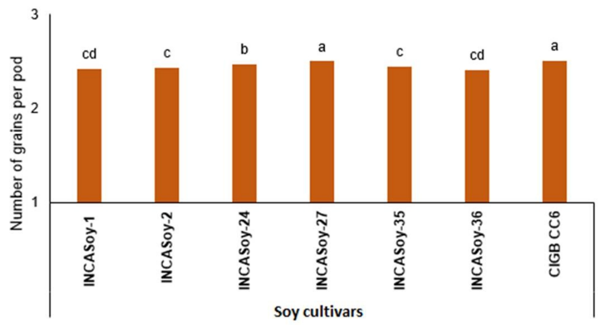


Figure 4. Mean values of grains per pod of the seven cultivars studied

Table 4. Factorial analysis for the variable yield in tons per hectare, evaluated at the physiological maturity stage (R8)

Origin of the variation	Estimated yield t ha^{-1}	
	Root mean square	Sig.
Cultivar	28.926	.000
Treatment	**143.672**	**.000**
Replica	.775	.498
Cultivar * treatment	1.471	.117
Cultivar * replica	.790	.570
Treatment * replica	.755	.518
Cultivar * treatment * replica	.779	.595
Error	.833	

R^2 = .297 (R^2 = .220)

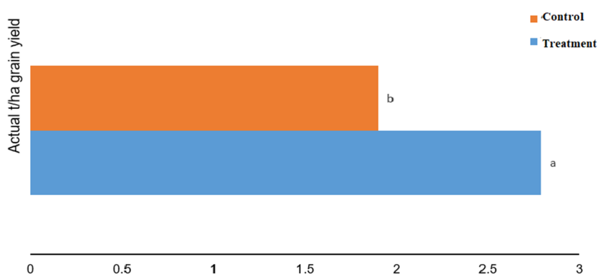


Figure 5. Yield of cultivars with treated and untreated seeds in t ha^{-1}

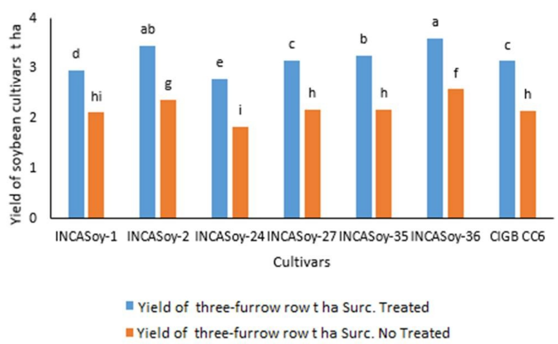


Figure 6. Yield of soybean cultivars with seed treatments and their controls

CONCLUSIONS

The heat treatment (50 ºC for 5 hours) to the soybean seed prior to summer sowing defined significant productive increases of any cultivar at the time of harvesting executed to each cultivar according to its productive cycle, also demonstrating the effectiveness of the yield estimation executed in R8. The treated plants showed higher mean germination values than the controls at all times of the evaluation, improved the emergence and vigor of the plants, a fundamental aspect to achieve high yields, since vigor determines the activity and performance in the normal growth and development of the plants. This is reflected in the first vegetative stages of the crop in greater uniformity, speed and percentage of emergence, and in later vegetative stages, plants with greater adaptability to the environment and survival in the field are obtained, guaranteeing high potentially productive populations.

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