Physiological potential of rice seeds treated with rhizobacteria or the insecticide thiamethoxam[1]

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ABSTRACT- Development of new technologies, aiming at increasing productivity in different crops, involves constant research on the effectiveness and application of these techniques in seed treatment. In this study, it was aimed at evaluating physiological potential of rice seeds treated with plant growth-promoting rhizobacteria (PGPR) (strains DFs185, DFs223, DFs306, DFs416), or with two dosages of the insecticide thiamethoxam. The variables assessed were: germination (G); first count of germination (FCG); cold test (CT); length of seedlings aerial parts (LAP), root system (LRS), and total length (TL); emergence speed index (ESI); emergence (E), at 14 days; and dry phytomass (DP). Treatments have had a positive effect on percentage of G, FCG and E. The strain DFs185 has promoted increase in percentage emergence, for five of the six lots assessed. The variables: LAP; LRS; TL; ESI; and DP have undergone low or none influence of treatments; and there has been no toxic effect of rhizobacteria or insecticide thiamethoxam. In the cold test, a negative effect of treatments has been detected. Seed treatment with rhizobacteria, as well as with thiamethoxam, improve quality of low quality rice seeds. The strain DFs185 is promising for treating rice seeds, once it stimulates seed germination and emergence.

Index terms: *Oryza sativa*, bioactivator, PGPR, vigor.

Potencial fisiológico de sementes de arroz tratadas com rizobactérias ou tiametoxam

RESUMO - O desenvolvimento de novas tecnologias, visando o aumento de produtividade em diferentes cultivos, implica em investigações constantes sobre a eficiência e aplicação dessas novas técnicas no tratamento de sementes. O objetivo deste trabalho foi avaliar o potencial fisiológico de sementes de arroz, tratadas com rizobactérias promotoras de crescimento de plantas (RPCP) (estirpes DFs185, DFs223, DFs306 e DFs416) e duas doses do inseticida tiametoxam. As variáveis avaliadas foram: germinação (G); primeira contagem da germinação (PCG); teste de frio (TF); comprimento da parte aérea (CPA), sistema radicular (CPR) e total (CT); índice de velocidade de emergência (IVE); emergência (E) aos 14 dias; e fitomassa seca (FS). Os tratamentos conferiram efeito positivo aos porcentuais de G, PCG, e E. Estirpe DFs185 conferiu aumento do porcentual de emergência para cinco dos seis lotes avaliados. As variáveis CPA, CPR, CT, IVE e FS sofreram pouca ou nenhuma influência dos tratamentos; porém não houve efeito tóxico das rizobactérias e do tiametoxam. No teste de frio foi observado efeito negativo dos tratamentos. O tratamento das sementes com rizobactérias e tiametoxam incrementa o potencial fisiológico de sementes de arroz com baixa qualidade. DFs185 é promissor, para o tratamento de sementes de arroz, por estimular a germinação e emergência.

Termos para indexação: *Oryza sativa*, bioativador, RPCP, vigor.

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Introduction

Rice (*Oryza sativa* L.), is one of the most consumed sources of food worldwide, and is of essential importance for developing countries; once it presents high potential to combat hunger and is adapted to different environments. However, the annual production of this cereal does not meet the worldwide demand; and despite Brazilian production be ranked among the highest in the world, such amount is not sufficient to supply the nationwide demand (Embrapa, 2010).

In many crops, including rice, studies have been demonstrating the capacity of microorganisms, or chemical activators, of incrementing physiological potential of seeds treated with rhizobacteria (Freitas et al., 2003; Freitas and Aguilar-Vildoso, 2004; Ludwig and Moura, 2007) or with the insecticide thiamethoxam (Castro and Pereira, 2008).

Seed treatment with some microorganisms has been used aiming at increasing productivity; once, besides protecting plants against phytopathologic agents, it can also promote their growth. Such microorganisms are generally linked to development of plants; including beneficial effects on seed germination and emergence, and development of seedlings, as well as production of grains and fruits (Harman, 2000).

Studies carried out with bacteria isolated from rhizosphere have been showing the capacity of these microorganisms in promoting plant growth. These so called plant growth-promoting rhizobacteria (PGPR) can colonize different plant organs and exert beneficial effects on them; and are also able of promoting increase on seed germination rate under glasshouse conditions, as well as in the field (Kloepper and Schroth, 1978; Amorim and Melo, 2002; Dey et al., 2004; Suslow and Schroth, 1982). Such benefits can occur through production of hormones and/or enzymes by these bacteria (Glick et al., 1999).

The Discovery of the molecule thiamethoxam ((3-[(2-chloro-5-thiazolyl) methyl] tetrahydro-5-methyl-N-nitro-4H-1,3,5-oxadiazin-4-imine) has brought new perspectives for agriculture; mainly for seed treatment. Many studies have shown the efficiency of thiamethoxam as a bioactivator; incrementing germination, emergence, and length of seedlings (roots and aerial parts) (Acevedo and Clavijo, 2008) and the dry phytomass, thus improving performance of the seedlings under stress conditions; and by increasing the levels of total proteins and enzymes in plants (Acevedo and Clavijo, 2008). Efficiency of thiamethoxam has been proved, mainly for soybean (Castro and Pereira, 2008). However, studies with other species, including rice, have demonstrated that this product possesses a wide spectrum of action (Acevedo and Clavijo, 2008).

When used as soybean seed treatment, thiamethoxam accelerates germination and induces greater development of embryonic axis; minimizing negative effects in situations of presence of aluminum, salinity, and water deficit. According to Cataneo (2008), the product accelerates germination by stimulating activity of the enzyme peroxidase; thus preventing oxidative stress. The action of thiamethoxam on seed germination reduces time for establishment of crop in the field; diminishing negative effects of competition with weeds, or by essential nutrients present in the soil (Cataneo, 2008).

Considering the scarceness of information referring to treatments that increment physiological potential of seeds, the objective of this study was to assess the performance of rice seeds treated with PG PR and thiamethoxam.

Material and Methods

Saline suspensions, each of which containing one of four strains of the rhizobacterium or two different dosages of thiamethoxam, were applied on soybean seeds.

The rhizobacteria strains used were: DFs185 [*Pseudomonas synxantha* (Ehrenberg) Holland], DFs223; (*P. fluorescens* Migula); DFs306 (non-identified strain); and DFs416 (*Bacillus* sp. Cohn). These strains were preselected, once they are able in controlling diseases and promoting development of rice plants; according to data already published by Ludwig and Moura, 2007 and Ludwig et al., 2009.

Microbiolization of rice seeds was performed with saline suspension of rhizobacteria of each of the four strains, after each strain had been grown during 24 h, on the culture medium 523 developed by Kado and Heskett (1970). Each of suspension was prepared with a saline solution, containing 0.85% of NaCl, with the concentration adjusted to A_{540}=0.5; followed by microbiolization through immersing the rice soybean seeds in the rhizobacteria suspension that then was shaken during 30 min., at 10 ºC, with the aid of a motorized shaker.

For obtaining concentration of insecticide thiamethoxam, seed were treated with an equivalent to 100 mL or 200 mL of product per 100 kg of rice seeds. The product was directly applied in the bottom of plastic bags before introducing seeds; which were then shaken until the seeds were uniformly covered by the product. A volume of the mixture (product + distilled water) was used; what was sufficient to promote a uniform distribution of thiamethoxam on the seeds. To measure the quantities of product and distilled water were used micropipettes.

The treatments used as control have consisted of: seeds without microbiolization; and without the tiametoxan. The physiological potential of seeds was evaluated by the following tests:

Germination: this test has been performed according to Rules for Seed Testing (Brasil, 2009). Except for the first day of assessment what was performed at the seventh day.

First count of germination: performed in the seventh day after sowing, according to Brasil (2009), except for the assessment day.

Cold test, without soil: for such test, the seeds have been sown in the same manner used for those subjected to germination test. However, differently from standard germination test, rolls of paper towels containing the seeds, were placed into plastic bags and were maintained into cold chamber, at 10 ºC, during seven days, previous to germination. After this period, the rolls were removed from cold chamber and placed in a seed germinator, at 25 ºC, also during seven days. Evaluations were performed according to RSA.

Lengths of aerial parts and roots, and total length of seedlings: in this test, 16 subsamples of 10 seeds each, for each treatment, were used. These seeds were sown in the upper third of two sheets of germination paper (Germitest®), covered with another sheet of the same paper, moistened with distilled water at an amount equivalent to 2.5 times the mass of dry substrate, made into rolls and maintained into seed germinator, at 25 ºC, during seven days. Total length (aerial part + root) was obtained, by summing the values obtained for aerial part and root of each replication and dividing by number of seedlings assessed. Results were expressed in cm./plântula.

Assessments in the greenhouse:

Seedlings emergence: for such test, were used four replications, with 100 seed each, which were sown into individual cells of polystyrene trays containing commercial substrate. Assessments were performed 14 days after the sowing, by counting total number of emerging seedlings; and results were expressed in percentage.

Emergence Speed Index (ESI): was jointly conducted with the emergence test; and the assessments were daily performed until the fourteenth day; by using such data in the equation proposed by Maguire (1962) for computing the ESI.

Lengths of aerial parts and roots and total length of seedlings: for such tests were used four replications with 10 seeds each, which were obtained from test of emergence; and assessments were carried out 14 days after the sowing. Lengths of aerial parts and roots were obtained, and then total length (aerial part + roots) was obtained by summing the values of measurements acquired in each replication and dividing by the total number of normal seedling assessed. The results were expressed in cm./plântula.

Dry phytomass: here, were used the same seedlings obtained from the length test, which after being measured, were stored in Kraft paper bags and then placed into a drying oven, at 60 ºC, until reaching constant weight. The total dry phytomass of seedlings was obtained by summing dry masses of seedlings from each replication, and then dividing result of this addition by the total number of seedling assessed; with the results being expressed in mg./plântula.

Experimental design: a completely randomized experimental design was used in the experiment, which was carried out with four replications, and with treatments arranged in a factorial scheme 7 x 6 [7 treatments (4 strains of rhizobacteria; 2 dosages of thiamethoxam; and control treatment) x 6 different lots of rice seeds]. Data with values expressed in percentages were computed after transformation by arc sine ($\sqrt{x} \div 100$). The Tukey test was used for comparing means of treatments, at 5% probability level.

Results and Discussion

Based on results obtained in tests of germination and first count of germination (vigor), the rice seed lots assessed were sorted as possessing: high vigor (lots 2 and 6); medium vigor (lots 1, 3, and 5; and low vigor (lot 4), as presented in Table 1.

In Table 2 it is possible to observe the general performance of rice seed lots with high vigor (lots 2 and 6), which have not responded to treatments, as much to the strains of rhizobacteria as to application of thiamethoxam; although, for the seed lots of medium vigor (lots 1, 3, and 5) and low vigor (lot 4) there have been positive and statistically significant responses to the treatments with rhizobacteria strains and thiamethoxam. Lot 1, with 74% of normal plants, and that initially was not presenting the minimum germination percentage established by Ministry of Agriculture, Livestock and Food Supply (MAPA, 2005), has reached percents of until 88% germination when under microbiolization with the strain DFs416 of rhizobacterium; and such percentage represents an increase of 14 percentage points on seed germination of such lot. Rhizobacteria of the strains DFs306 and DFs223; as well as thiamethoxam, in dosages of 100 or 200 mL 100 kg of seeds^{-1}, have also conferred increases on germination percentage to seed lot 1, which then have reached the minimum limit established by legislation.

Table 1. Initial characterization of rice seed lots selected for an experiment of seed treatment with Plant Growth-Promoting Rhizobacteria (PGPR) and the insecticide thiamethoxam.

Seed lots	Moisture content (%)	First count of germination	Germination (%)
1	11.0	69 b*	74 bc*
2	11.0	81 a	90 a
3	12.0	70 b	82 b
4	11.0	49 c	71 c
5	11.0	62 b	80 bc
6	12.0	84 a	90 a

*Means followed by the same lower case letters in the column do not statistically differ between each other by Tukey test, at 5% probability.

Seed lot 4, with initial germination of 71%, in control treatment, has presented germination percentage of 83% after microbiolization with the strain DFs306 of rhizobacteria (Table 2). The stimulus to germinations, induced by rhizobacteria strains, may be related to production of hormones, such as gibberellins (Holl et al., 1988) and auxins (Araujo et al., 2005), as well as by enzymes, such as ACC (1-aminocyclopropane-1-carboxylate) deaminase. For Glick et al. (1995), this enzyme can stimulate vegetal growth, particularly growth of roots, by kidnapping and hydrolyzing ethylene of the seeds. As example, it can be cited that ethylene stimulates germination and dormancy overcoming of seeds of majority of plant species (Taiz and Zeiger, 2004).

The results positive and statistically significant herein obtained with thiamethoxam corroborate results achieved by Acevedo and Clavijo (2008), who have observed increase in percentage of germination in seeds of bean (*Phaseolus vulgaris* L.), maize (*Zea mays* L.), and soybean [*Glycine max* (L.) Merrill]. Almeida et al. (2009) have observed increase on vigor of seeds of carrot (*Daucus carota* L.) treated with this bioactivator; and Lauxen et al. (2010) have reported the same effect on seeds of cotton (*Gossypium hirsutum* L.) treated with thiamethoxam. According to reports of Cataneo (2008), thiamethoxam accelerates germination, thus preventing oxidative stress.

Through data presented in Table 2, it can be verified that results obtained in the test of first count of germination have presented the same behavior found in the germination test, once there has been no response for seed lots with high vigor. Lot 5 has presented statistically significant response to three of the four strains of rhizobacteria: i.e., increase of 15 percentage points after microbiolization with the strain DFs416; 13 percentage points for the strains DFs223; and 12 percentage points for the strain DFs306. For thiamethoxam, in dosage of 100 mL 100 kg of seed^{-1}, the increase has reached 11 percentage points. Lot 4 has presented positive responses to strains DFs 306 and DFs185 of rhizobacteria, with increases of 14 and 11 percentage points, respectively, on germination, seven days after microbiolization.

Dileep et al. (1998) have found increase on germination on seeds of rice, peanut (*Arachis hypogaea* L.), and okra [(*Abelmoschus esculentus* (L.) Moench.], inoculated with species of *Pseudomonas* spp. fluorescent, isolated from rhizoplane of rice and green pepper (*Capsicum annuum* L.) plants. For Araujo et al. (2005), rhizobacteria can produce hormones, such as acid indoleacetic (IAA) and indolebutyric acid (IBA), besides the secretion of enzymes that are important for nutrition of plants (Powar and Jagannathan, 1982). In a study on action of hormones in the germination of soybean seeds, Vieira and Castro (2001) have observed that the intermediary concentration of auxin hormone has significantly increased germination.

Within this study, the cold test has detected differences statistically significant on seed quality among the six seed lots of rice studied (Table 2); including lot 4 that, although presenting low vigor, has shown lower values than the remaining lots in the germination test, after application of some treatments. An hypothesis that can explain such event is that the cold possibly inhibited development of a given pathogen, which could be present in seeds of lot 4, causing the germination percentage, except for the treatment with thiamethoxam in dosage of 200 mL 100 kg of seeds^{-1}, was lower after the cold test.

By data herein obtained, it was not possible to identify beneficial effects of thiamethoxam and rhizobacteria; once, after the cold test there has been reduction on germination of rice seeds of 34 a 14 percentage points, for lots 4 and 6, respectively, after microbiolization with the strain DFs223. Neither rhizobacteria nor thiamethoxam have conferred significant and positive effects on the rice seeds; similar to what was observed by Bittencourt et al. (2000) with seeds of maize, in detecting significant reductions on vigor of seeds treated with insecticides; among them the thiamethoxam. Notwithstanding, as the cold might have inhibited development of pathogens, possibly present in seed lot 4, likewise might have inhibited or impaired development of rhizobacteria, and thus prevented their beneficial effects.

Table 2. Percentages of germination, first count of germination, and germination after the cold test, of six different lots of rice seeds without treatment (W/treat.) and treated with two dosages of thiamethoxam (Thiam., in dosages of 100 and 200 mL 100 kg of seeds^{-1}) and four distinct strains of rhizobacteria (DFs).

Germination (%)							
Lots	W/ treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	74 bcC*	83 bABC*	85 abAB*	78 bBC*	85 aAB*	86 aAB*	88 aA*
2	90 aA	86 abA	90 aA	90 aA	85 aA	90 aA	83 aA
3	82 bAB	79 bAB	77 bAB	83 abAB	85 aA	75 bB	85 aA
4	71 cBC	68 cC	65 cC	78 bAB	73 bBC	83 abA	72 bBC
5	79 bcB	81 bAB	88 aA	81 bAB	82 aAB	84 abAB	86 aAB
6	90 aA	91 aA	88 aA	89 aA	85 aA	89 aA	91 aA
CV = 4.74%							
First count of germination (%)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs 185	DFs223	DFs306	DFs 416
1	69 bAB	75 bA	65 bcB	69 cdAB	65 bB	64 cdB	76 bA
2	81 aA	73 bAB	73 bAB	78 bAB	76 aAB	75 abAB	70 bB
3	70 bAB	61 cBC	59 cC	72 bcA	79 aA	73 bcA	79 bA
4	49 cC	38 dD	55 cABC	60 dAB	53 cBC	63 dA	49 cBC
5	62 bB	70 bcAB	73 bA	69 cdAB	75 aA	74 abA	77 bA
6	84 aA	83 aA	85 aA	86 aA	81 aA	82 aA	88 aA
CV = 4.88%							
Germination after cold test (%)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs 185	DFs223	DFs306	DFs 416
1	78 aAB	78 abAB	81 abAB	87 aA	81 aAB	77 aB	78 abAB
2	78 aA	78 abA	85 aA	78 abA	80 abA	79 aA	78 abA
3	79 aA	80 abA	85 aA	78 abA	80 abA	79 aA	78 abA
4	83 aA	85 aA	77 abAB	67 bBC	49 dD	59 bCD	68 bBC
5	74 aA	71 bA	71 bA	81 aA	70 bcA	74 aA	77 abA
6	80 aAB	71 bBC	74 bABC	84 aA	66 cC	83 aA	83 aA
CV = 5.83%							

*Means followed by the same lower case letter in the column and upper case letter in the line do not statistically differ between each other by Tukey test, at 5% probability. CV = coefficient of variation; DFs185 = *Pseudomonas synxantha*; DFs223 = *P. fluorescens*; DFs306 = strain non-identified; and DFs416 = *Bacillus* sp.

There have been statistically significant differences, among seed lots, for almost all strains of rhizobacteria and for the two dosages of thiamethoxam, for length of aerial parts of seedlings in the experiment carried out under glasshouse conditions (Table 3); except for lot 4, where the thiamethoxam, in the dosages of 100 or 200 mL 100 kg of seeds^{-1}, as well as for all strains of rhizobacteria have not caused statistically significant differences for increase in length of aerial parts, as compared to control treatment. For seed lot 3, the treatment with thiamethoxam, in dosage of 100 mL 100 kg of seeds^{-1} has been responsible for an increase of 3.34 cm (30%), as compared with seedling from non-treated seeds.

The seed lots were quite uniform, as for length of root system for seedlings grown under glasshouse conditions, where only a few seedlings have shown statistically significant differences between each other, by Tukey test at 5% probability (Table 3). Generally, for seed lot 3, there has been no statistically significant response to strains of rhizobacteria, or to thiamethoxam bioactivator, for such variable. However, the treatment with thiamethoxam, in the dosages of 200 or 100 mL 100 kg of seeds^{-1}, or the strains DFs306 and DFs416 of rhizobacteria have induced increases in length of root system of seedling of the seed lot 2; while, for lot 4, only the strain DFs306 has caused statistically significant difference, in relation to control treatment.

For total length of seedlings grown under glasshouse conditions, likewise, effect of thiamethoxam was not observed for both dosages used, and for almost all the strains of rhizobacteria (Table 3); only lot 3 differed statistically from control treatment, when treated by microbiolization with the strains of the rhizobateria DFs223 and DFs306.

Table 3. Lengths of aerial part and root system, and total length of rice seedling from six different lots of rice seeds without treatment (W/treat.) and treated with two dosages of thiamethoxam (Thiam., in dosages of 100 and 200 mL 100 kg of seeds^{-1}) and four distinct strains of rhizobacteria (DFs), after 14 days, under greenhouse conditions.

Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
Length of aerial part (cm.seedling^{-1})							
1	14.18 aAB*	14.50 aAB*	12.51 aB*	12.53 aB*	15.41 aA*	13.32 abAB*	13.88 aAB*
2	14.54 aA	14.26 aA	11.78 aA	13.64 aA	13.35 abA	13.03 abA	12.23 aA
3	10.90 bB	12.45 aAB	14.24 aA	12.89 aAB	14.05 abA	14.02 abA	12.85 aAB
4	14.00 aA	14.27 aA	13.83 aA	13.50 aA	14.73 aA	14.62 abA	12.07 aA
5	15.16 aA	15.19 aA	13.34 aA	14.29 aA	14.05 abA	15.01 aA	14.03 aA
6	13.29 abA	12.92 aA	13.75 aA	11.83 aA	11.95 bA	12.04 bA	13.24 aA
CV = 9.94%							
Length of root system (cm.seedling^{-1})							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	4.14 aAB	4.47 aA	4.21 aAB	3.78 aB	4.12 aAB	4.28 aAB	4.03 aAB
2	3.95 aD	4.18 aBCD	4.85 aA	4.00 aCD	3.98 aD	4.78 aAB	4.66 aABC
3	3.86 aA	4.23 aA	4.51 aA	4.30 aA	3.98 aA	4.25 aA	4.19 aA
4	4.02 aB	4.04 aB	4.27 aAB	3.92 aB	3.97 aB	4.87 aA	4.11 aB
5	4.04 aA	4.46 aA	4.37 aA	4.06 aA	4.12 aA	4.26 aA	4.35 aA
6	4.39 aAB	3.92 aAB	4.44 aAB	3.90 aB	3.90 aB	4.58 aA	4.08 aAB
CV = 7.46%							
Total length (cm.seedling^{-1})							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	17.36 abAB	18.52 abA	16.17 aAB	15.17 bB	18.39 aA	16.02 aAB	17.03 aAB
2	17.97 aA	18.07 abA	16.89 aA	16.75 abA	16.37 abA	17.18 aA	16.06 aA
3	14.75 bB	16.76 bAB	17.57 aAB	17.33 abAB	18.05 abA	18.28 aA	16.09 aAB
4	17.27 abAB	18.34 abA	16.97 aAB	17.41 abAB	18.14 aA	17.84 aAB	15.25 aB
5	18.48 aAB	20.08 aA	17.69 aAB	18.38 aAB	17.13 abB	18.41 aAB	17.93 aAB
6	17.68 aA	16.83 bA	17.67 aA	14.83 bA	15.33 bA	17.54 aA	15.30 aA
CV = 7.93%							

*Means followed by the same lower case letter in the column and upper case letter in the line do not statistically differ between each other by Tukey test, at 5% probability. CV = coefficient of variation; DFs185 = *Pseudomonas synxantha*; DFs223 = *P. fluorescens*; DFs306 = strain non-identified; and DFs416 = *Bacillus* sp.

These results for length of seedling (aerial parts, root system, and total length) demonstrate that the rhizobacteria, as well as the bioactivator thiamethoxam, do not present phytotoxic effect on rice seedlings. Results on length of seedlings with the bioactivator, are in agreement with those results obtained by Castro et al. (2008), who, in evaluating growth of soybean roots, grown from seeds treated with thiamethoxam, have concluded that such insecticide has not promoted greater growth of roots. Araujo (2008) has assessed growth of plants of soybean, maize, and cotton promoted by inoculation with the bacteria *B. subtilis*, and found that only the maize plants have had increase on plant height and also on foliar area. Therefore, these results demonstrate that, for plant growth promotion, interaction between the plant species and the strain of rhizobacteria is necessary.

For the dry phytomass of root system and aerial parts, there were no statistically significant differences among the majority of treatments; except for reductions on dry mass of root system, as well as of aerial parts induced by the strain DFs185 of rhizobacteria for lot 6, in relation to control treatment (Table 4).

Likewise, for the ESI variable there have not been statistically significant differences among the majority of treatments; only excepting a reduction on the ESI for seed lot 1, which was induced by treatments with thiamethoxam, in dosage of 100 mL 100 kg of seeds^{-1}, and by the strains DFs185 and DFs223 of the rhizobacteria (Table 5). For Dan et al. (2010), the speed of emergence plays a major role in the rapid establishment of seedlings under field conditions; despite the same authors have confirmed that effects of treatments with thiamethoxam on emergence speed of soybean seedlings have not been found.

All the strains of the rhizobacteria, as well as the dosage of 100 mL.100 kg of seeds^{-1} of thiamethoxam have shown positive responses and statistically significant differences on emergence of at least one of the six seed lots assessed (lot 1) (Table 5).

Table 4. Dry phytomass of root system and aerial part of rice seedlings from six different lots of rice seeds without treatment (W/treat.) and treated with two dosages of thiamethoxam (Thiam., in dosages of 100 and 200 mL.100 kg of seeds^{-1}) and four distinct strains of rhizobacteria (DFs), after 14 days, under greenhouse conditions.

Dry phytomass of root system (mg)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	38.9 aA*	36.4 aA*	49.9 aA*	31.6 aA*	35.8 aA*	40.5 aA*	41.1 aA*
2	35.5 aA	37.3 aA	48.3 aA	35.6 aA	35.1 aA	46.9 aA	46.2 aA
3	34.8 aA	31.5 aA	44.8 aA	34.2 aA	39.0 aA	36.8 aA	39.7 aA
4	30.6 aA	35.8 aA	50.9 aA	34.5 aA	33.2 aA	45.1 aA	37.0 aA
5	40.0 aA	35.3 aA	50.9 aA	32.5 aA	41.2 aA	49.3 aA	39.2 aA
6	31.3 aAB	32.9 aAB	46.8 aAB	17.1 aB	48.2 aAB	54.6 aA	34.3 aAB
Dry phytomass of aerial part (mg)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	72.0 aA	83.4 aA	72.0 aA	56.6 abA	71.1 aA	61.3 aA	66.5 aA
2	72.2 aA	84.3 aA	70.6 aA	63.1 abA	62.9 aA	65.5 aA	65.8 aA
3	60.8 aA	65.9 aA	72.6 aA	70.3 abA	64.4 aA	55.1 aA	63.9 aA
4	61.3 A	69.5 aA	70.4 aA	72.9 aA	58.4 aA	63.2 aA	46.1 aA
5	80.8 aA	76.1 aA	77.1 aA	61.0 abA	69.8 aA	73.9 aA	69.5 aA
6	74.5 aA	72.7 aAB	64.9 aAB	41.9 bB	58.0 aAB	66.1 aAB	55.7 aAB
CV= 22,39%							

*Means followed by the same lower case letter in the column and upper case letter in the line do not statistically differ between each other by Tukey test, at 5% probability. CV = coefficient of variation; DFs185 = *Pseudomonas synxantha*; DFs223 = *P. fluorescens*; DFs306 = strain non-identified; and DFs416 = *Bacillus* sp.

Table 5. Emergence Speed Index (ESI) and final emergence of rice seedlings from six different lots of rice seeds without treatment (W/treat.) and treated with two dosages of thiamethoxam (Thiam., in dosages of 100 and 200 mL 100 kg of seeds^{-1}) and four distinct strains of rhizobacteria (DFs), after 14 days, under greenhouse conditions.

ESI							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs85	DFs223	DFs306	DFs416
1	4.93 aAB*	4.24 abBC*	3.84 abC*	3.60 bC*	3.30 abC*	4.27 bBC*	5.59 aA*
2	3.87 bcAB	4.81 aA	4.04 abAB	3.44 bB	4.31 aAB	4.32 bAB	4.61 bA
3	4.42 abA	3.79 bA	4.13 abA	3.93 abA	3.91 abA	4.23 bcA	4.31 bA
4	3.36 cA	3.56 bA	3.51 bA	3.55 bA	3.31 bA	3.31 cA	3.13 cA
5	4.39 abAB	3.85 bB	4.28 abAB	3.79 abB	4.05 abAB	5.02 abA	4.25 bAB
6	4.79 abAB	4.08 abB	4.64 aAB	4.57 aAB	4.08 abB	5.28 aA	3.96 bcB
CV = 11.16%							
Final emergence (%)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	65 cC	69 cC	80 bB	86 aAB	82 abAB	88 abA	85 aAB
2	75 abBC	85 abA	67 dC	84 aA	80 bAB	82 bAB	67 Cc
3	72 bcBC	78 bB	71 cdBC	86 aA	70 cBC	70 cBC	68 cC
4	73 bcB	86 aA	71 cdBC	67 bBC	63 cC	66 cBC	62 cC
5	82 aBC	84 abABC	79 bcC	90 aA	88 aAB	82 bBC	79 abC
6	83 aCD	82 abCD	88 aABC	90 aAB	84 abBCD	92 aA	78 bD
CV = 4.44%							

*Means followed by the same lower case letter in the column and upper case letter in the line do not statistically differ between each other by Tukey test, at 5% probability. CV = coefficient of variation; DFs185 = *Pseudomonas synxantha*; DFs223 = *P. fluorescens*; DFs306 = strain non-identified; and DFs416 = *Bacillus* sp.

Despite negative result for the ESI of seeds of lot 1, considering mean quality, such seed lot was the most positively influenced by treatments with thiamethoxam (100 mL 100 kg of seeds^{-1}) and by all strains of rhizobacteria on emergence of seedlings, after 14 days (Table 5). It has to be emphasized that the strain DFs185 has increased final percentage emergence of five of the six lots assessed, inducing 21 and 14 additional percentage points, respectively, for lots 1 and 3, in relation to seeds of control treatment. The treatment with thiamethoxam, in the dosage of 200 mL 100 kg of seeds^{-1}, has been the only treatment that promoted increase on emergence of seedlings in the lot 4 (13 percentage points); however it has been also efficient for seeds of lot 2, which possessed high quality seeds (10 percentage points). The strain DFs306 induced positive and statistically significant responses for seeds of lots 1 and 6; which for lot 1, with low quality seeds, represents an increase of 23 percentage points, in relation to non-treated seeds.

Positive effect of microbiolization by the Gram-positive rhizobacteria *B. cereus* and *P. fluorescens*, has already been reported by Saravanakumar et al. (2007), who have observed increases varying from 37% to 50% on percentage emergence of seeds of Tea (*Camellia sinensis* L.). Kishore and Pande (2007) have observed increase that ranged from 12% to 19%, on emergence of peanut seeds, induced by Gram-positive bacterium *B. megaterium*.

Considering the possibility that the root growth of rice seedlings cultivated under glasshouse conditions could be limited by dimension of tray cells; the length of aerial parts and root system were also assessed by the germination method into rolls of paper towels (Table 6). By data therein presented it can be verified that there have been no statistically significant differences, so much among treatments as for among seed lots, for both factors (aerial parts and root system). Among rhizobacteria, the strains DFs185 (*P. synxantha*) and DFs306 (non-identified) have been the ones that propitiated the best performance for seeds among the variables assessed (Figure 6). However, strain DFs185, which was pre-eminent in the emergence of seedling for presenting positive effect in five of the six lots assessed (Table 5); have not induced a positive effect in lot 4, for both factors. Nevertheless, when treated with thiamethoxam, in dosage of 200 mL 100 kg of seeds^{-1}, these seeds have presented emergence greater than non-treated seeds (Table 5).

Table 6. Length of aerial parts and root system of rice seedlings from six different lots of rice seeds without treatment (W/treat.) and treated with two dosages of thiamethoxam (Thiam., in dosages of 100 and 200 mL 100 kg of seeds^{-1}) and four distinct strains of rhizobacteria (DFs), incubated in roll of paper towels and assessed after seven days.

Length of aerial parts (cm)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	2.95 aA*	2.92 aA*	3.15 aA*	2.83 abA*	2.96 aA*	3.58 aA*	2.90 aA*
2	2.79 aA	2.81 aA	3.22 aA	3.22 aA	3.33 aA	3.50 aA	3.25 aA
3	2.69 aA	2.70 aA	2.54 aA	3.03 abA	2.45 aA	3.27 aA	2.60 aA
4	2.77 aAB	2.78 aAB	2.62 aAB	2.00 bB	2.65 aAB	2.41 aA	2.73 aAB
5	2.87 aA	2.26 aA	2.77 aA	3.03 abA	2.93 aA	3.10 aA	3.26 aA
6	2.64 aA	2.86 aA	2.51 aA	2.90 abA	3.00 aA	2.84 aA	2.74 aA
CV = 17.6%							
Length of root system (cm)							
Lots	W/treat.	Thiam. 200	Thiam. 100	DFs185	DFs223	DFs306	DFs416
1	4.56 aA	4.26 aA	3.99 aA	4.53 aA	4.48 aA	4.86 aA	3.80 abA
2	4.12 aA	5.02 aA	4.46 aA	5.03 aA	4.87 aA	4.77 aA	4.41 abA
3	3.72 aA	4.80 aA	3.82 aA	4.58 aA	4.41 aA	3.83 aA	4.52 abA
4	3.15 aAB	4.47 aA	3.00 aAB	2.53 bB	3.89 aAB	3.60 aAB	2.99 bAB
5	4.53 aA	4.78 aA	4.68 aA	4.52 aA	4.69 aA	4.65 aA	5.37 aA
6	4.67 aA	5.36 aA	4.44 aA	5.30 aA	4.99 aA	4.29 aA	4.98 aA
CV = 16.8%							

*Means followed by the same lower case letter in the column and upper case letter in the line do not statistically differ between each other by Tukey test, at 5% probability. CV = coefficient of variation; DFs185 = *Pseudomonas synxantha*; DFs223 = *P. fluorescens*; DFs306 = strain non-identified; and DFs416 = *Bacillus* sp.

Araujo (2008) has verified that presence of *B. subtilis* in lots of maize, cotton, and soybean seeds has positively influenced the metabolism and physiology of plants, what is reflected on increase of emergence, growth, and nutrition of plants. According to Glick et al. (1999), bacteria may promote vegetal growth by direct stimulus so much by providing plant hormones, such as auxins and cytokinins, as for lowering the levels of ethylene, by means of the ACC deaminase enzyme. For Bewley e Black (1978) the emergence of seedling can be stimulated by acidification of cell wall during germination process, in such case being essential the action of auxin.

Conclusions

The seed treatment with rhizobacteria or thiamethoxam increments physiological potential of low quality rice seeds.

The plant growth-promoting rhizobacterium (PGPR) (strain DFs185) is promising for the treatment of rice seeds, since it increases germination and emergence percentages.

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