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PRELIMINARILY EVALUATION OF BIO-PRIMING MAIZE GRAINS WITH SOME STORED BIOAGENTS AS BIOLOGICAL CONTROL MEASURES AGAINST ROOT ROTS UNDER GREENHOUSE CONDITIONS

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Abstract

Stored formulated antagonistic some fungal and bacterial agents were evaluated for their activity against pathogenic fungi causing Maize root rot disease under *in vitro* and greenhouse conditions. The tested antagonists were *Trichoderma harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens*, meanwhile pathogenic fungi were *Rhizoctonia solani*, *Macrophomina phaseolina* and *Fusarium solani.* The antagonistic agents applied asbio-priming Maize grains with stored formulated bioagents were tested as inhibitor factor against pathogenic fungi *in vitro*. Meanwhile, surface-sterilized Maize grains bio-primed with fresh prepared fungal or bacterial suspensions and used as comparison treatment. Under laboratory conditions, all tested bioagents either used as fresh suspension or formulated on carrier material of CMC, sawdust and talc powder had significant inhibitor growth reduction of pathogenic fungi. *Fusarium solani* showed more sensitivity against tested bioagents followed by *M. phaseolina* and *R. solani*, respectively.

Under greenhouse conditions, the highest influence of bioagents was observed at treatment of applied grain bio-primed with fresh suspension which recorded 4.1-4.9% as a mean range of root rot incidence compared with 33.3% in un-primed grains control treatment. Similarly, the bioagents formulated on sawdust carrier showed higher significant low disease incidence than that of formulated on CMC and talc powder, in respective order. These treatments recorded 6.2-7.7%, 8.7-11.2% and 11.1-13.2%, as a mean range in respective order according to the used carrier.

On light of the obtained results, the use of bio-priming grains with bioagents might besuggested as promising applicable techniques for controlling such soilborne plant pathogens, especially it characterized as safe, cheap and easily applied.

Key words : Biocontrol, formulated bioagents, Maize, root rot.

Introduction

Maize (*Zea mays* L.) is an important grain crop for human food security, fodder, and biofuel production. Soil-borne diseases significantly reduce maize yield in irrigated systems where maize follows winter wheat, resulting in a significant reduction in yield due to root rot disease. Corn roots are infected with optional parasitic fungi that occur in the soil and under the seed layer (Chambers 1987). *Rhizoctonia solani*, *Macrophomina phaseolina* and *Fusarium solani* are the most common fungi that affect corn root causes of root rot disease (Sumner and Bell 1982; Ocamb and Kommedahl 1994; Pal *et al.,* 2001). The investigation of root rot disease is particularly important given its widespread prevalence in Egypt, especially in sandy soils. To date, because of scientific and practical difficulties, there is no economic way to control root rot disease in many crops.

Growing demand for a steady food supply for the growing world population will require control of plant diseases that significantly reduce crop yields. Current plant disease control practices are largely dependent on the development of resistant and synthetic pesticides (Emmert and Handelsman 1999). Biocontrol is receiving greater attention due to its low cost and environmentally friendly application.

The application of biological controls using

antimicrobials has proved successful in combating various plant diseases in many countries (Sivan 1987). In recent years, hostile microorganisms have been applied to overcome this obstacle during several attempts. As an effective agent in biological control of plant diseases caused by soil-borne fungi, *Trichoderma* spp. was documented (Sivan and Chet 1986; Whipps and Lumsden 2001; McLean *et al.,* 2004). Grain root diseases caused by *R. solani* and *S. rolfsii* were reduced by applying wheat bran colonized by *T. harzianum* to soil affected by these pathogens (Hadar *et al.,* 1979 and 1984; Elad *et al.,* 1980).

Kim *et al.,* (1997) Effective control of three fungal wheat root diseases was found by seed treatment using *Bacillus* spp. Also, the applied pea seeds worked with *Pepudomonas cepacia* or *P. fluorescens* as a biological control agent against the erosion of lithium and Aphanomyces root rot and managed to reduce the incidence of disease (Parke *et al.,* 1991; King and Parke 1993). In addition, *Bacillus* sp. recorded to give a highly antagonistic effect against some pathogenic fungi including *F. solani* (Sunick *et al.,* 1997).

The aim of this study was to evaluate the activity of some stored fungal and bacterial antimicrobial agents in reducing corn root rot disease when applied as a treatment for biofilm grains and planted into artificial infested soil with pathogens, *R. solani*, *M. phaseolina* and *F. solani* under Greenhouse conditions.

Materials and Methods

Pathogens and antagonists

The tested pathogenic fungal isolates of *Rhizoctonia solani*, *Macrophomina phaseolina* and *Fusarium solani* were isolated during survey carried out as a part of in house project 11030132, National Research Centre (NRC). Meanwhile, the antagonistic microorganisms, *T. harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens* were obtained from the Plant Pathology Dept., NRC, Giza, Egypt. The antagonists were isolated from the rhizosphere of various healthy and root rot infected various crops, grown in the Delta and Middle Egypt regions, and proved their high antagonistic ability during previous work at the same department. Fungal and bacterial cultures were maintained on potato dextrose agar (PDA) and nutrient agar slant media at 5±1°C as stock cultures until use. All isolates were refreshed by growing at the optimum growth conditions at the beginning of the present experiments.

Plant material

Maize grains (cv. M84) used in the present work was obtained from the Cereal Crops Research Institute, Agricultural Research Centre, Ministry of Agriculture, Giza, Egypt.

Growth media

PDA and nutrient media (Difco Laboratories, Detroit, MI) were used for growing fungal and bacterial isolates tested in the present work.

Laboratory tests

Preparation of fungal spores and bacterial cells suspensions

Antagonistic fungal bioagent inoculum was grown on PDA medium at 25±2°C until an abundant heavy growth of conidia was evident. Conidia were harvested by scraping the surface of the colonies with a spatula and then transferring them to sterilized distilled water and filtering them through nylon mesh. All spore suspension was adjusted with sterile water to give a spore concentration of 10^5-10^6/mL. Meanwhile, bacterial bioagents were grown for 48 h in nutrient broth medium, and then cells were harvested by centrifugation. Bacteria were re-suspended in sterile distilled water and the concentration adjusted to give 10^7-10^8 cells/mL.

Formulation of fungal and bacterial bio-agents

Carboxymethyl cellulose (CMC), sawdust and talc powder were used as different carrier materials for bioagents propagules formulation. CMC, sawdust and talc powder were autoclaved (120°C/1.5lb for 60 min.) before use. Fungal and bacterial spores/cells suspensions were added individually to sterilized carriers to prepare different formula at the rate of 2:1 (carrier: suspension, w:v), then mixed thoroughly to even ensure equal distribution of microorganism suspension through the mixed carriers. The prepared mixture then placed onto paper sheet and left for air dry 2-3 hrs at room temp. (23-25°C). Then, the mixtures were passed through sieve (0.25 mm-diameter) to get the formulated bio-agent in granules shape. The obtained granules were packed into dark glass bottles (500 mL) sealed with cotton plugs and stored in incubator at 20 ±2°C for six months.

Bio-priming Maize grains and antagonism test

Three groups of surface-sterilized Maize grains (using 3% sodium hypochloride for 5 min, then air-dried) were used in this test. Arabic gum (0.5%) was added to grains before dressing for getting a sticky purpose. For the first group, Maize grains were bio-primed individually with each of the prepared stored formulated bioagents for 6 months at the rate of 3g/100 grain. The second group was surface-sterilized Maize grains bio-primed with fresh prepared fungal or bacterial suspensions at the rate

of 3ml/100 grain and used as comparison treatment. Meanwhile, the third group was only surface-sterilized Maize grains left as control treatment. All treated Maize grains were air-dried on filter paper for 1 h in a laminar flow hood.

The antagonistic activity of fungal and bacterial antagonist's bio-primed Maize grains was evaluated using the dual culture technique (Ferreira *et al.,* 1991) on PDA medium in 9-cm-diameter Petri dishes. Maize grains bio-primed with formulated bioagents (stored for 6 months) as well as bio-primed grains with the same bioagents as fresh suspensions (for comparison treatment) were evaluated on PDA plates. Bio-primed Maize grains were used in this test instead of the growth culture disks of tested biocontrol agents.

The pathogenic fungal isolates of *Rhizoctonia solani*, *Macrophomina phaseolina* and *Fusarium solani* were used in this test. One previously bio-primed treated or un-primed grain was placed onto the PDA, 10mm from the edge of the Petri dish. Another disk of the same diameter of each pathogenic fungal growth culture was placed on the opposite side of the dish at the same distance. The control treatment was inoculated with a culture disk of either a pathogenic or antagonistic culture alone at the same conditions. All Petri dishes were assigned to a completely randomized design, with five replicates per treatment and incubated at 25±1°C. Afterthe pathogenic fungal growth in the control treatment had reached the edge of the Petri dish, the fungal growth diameter away from and towards the antagonist agent in all treatments was measured. This test was repeated three times and the inhibition was calculated as the reduction (%) in pathogenic colony diameter growth compared with the control.

Greenhouse experiments

Evaluation of the bio-primed Maize grains with stored formulated biocontrol agents against root rot incidence caused by *R. solani*, *M. phaseolina* and *F. solani* was performed in pot experiments under greenhouse conditions.

Experiments were carried out in a sandy loam soil artificially infested with the root rot pathogens. Fungal mass production used for soil infestation was obtained by growing the tested isolates on sand barley medium. This natural medium was prepared by mixing sand and barley (1:1, w:w and 40% water), then the mixture was packed into glass bottles sealed with cotton plugs and sterilized for three successive periods at121°C. The autoclaved medium was then inoculated individually with a 5-mm disk of each pathogenic fungal growth and incubated at 28±2°C for 2 weeks. Soils were infested individually at a ratio of 5% (w:w) with pathogenic fungal cultures and mixed thoroughly to ensure equal distribution of fungal inoculum, then filled in plastic pots (25-cm-diameter) and irrigated every second day for 1 week before sowing. After 12 hrs the bio-primed Maize grains with the stored formulated antagonistic bioagents *T. harzianum*, *B. subtilis* and *P. fluorescens* were sown at the rate of five grains per pot (40cm diameter) and six pots were used as replicates for each particular treatment. A set of un-primed Maize grains were used for control treatment. The average percentage of pre- and post-emergence root rot incidence was recorded up to 45 days of sowing (the experimental period). All of the previous procedures were repeated three times.

Disease assessment

Percentage of root rot incidence at the pre-emergence stage was calculated as the number of absent seedlings relative to the number of grains sown. Meanwhile, the percentage of root rot incidence at the post-emergence stage was calculated as the number of diseased plants relative to the number of emerging seedlings.

At the end of the experiment, Maize plants were carefully pulled out from pots after being flooded with water in order to leave the root system undamaged. Plant roots showing rot lesions in addition to the visual root rot symptoms on the shoot system were considered diseased plants. Isolation from infected germinated Maize grains at the pre-emergence stage as well as infected Maize plants at the post-emergence stage was carried out. Undeveloped, germinated grains which were picked up from the soil, and the diseased Maize plants were both water washed and surface sterilized with 3% sodium hypochlorite then subjected to re-isolation trials for the causal pathogens. The fungi obtained were compared with those used in soil infestation to prove their identity.

Statistical analysis

The obtained data were subjected to IBM SPSS software version14.0. Analysis of variance was determined and the mean values were compared by Duncan's multiple range test at $P < 0.05$.

Results and Discussion

The inhibitory effect of fungi and stored antibacterial bacteria stored in the laboratory in this study were tested as antifungal agents applied as biologically prepared granules. The percentages for reducing the growth of pathogenic fungi in response to antimicrobial agents are shown in table 1). The data in table 1 indicate that all the

Table 1: Growth reduction of pathogenic fungi in response to stored formulated antagonistic agents applied as Maize grains dressing *in vitro*.

Maize grains dressing		Fungal growth reduction (%)		
Carrier	**Formulated antagonist***	***R. solani***	***M. phaseolina***	***F. solani***
CMC	*T. harzianum*	82.7 ±3.9a	83.3 ±2.9a	84.2 ±1.5a
	B. subtilis	74.3 ±2.5b	73.3 ±1.4b	75.6 ±1.8b
	P. fluorescens	66.4 ±1.0c	63.0 ±1.6c	67.6 ±0.8c
Sawdust	*T. harzianum*	84.2 ±2.8a	85.0 ±0.2a	86.7 ±1.4a
	B. subtilis	76.6 ±0.9b	74.7 ±9.0b	79.6 ±0.8b
	P. fluorescens	63.6 ±3.0c	62.2 ±2.6c	66.6 ±2.2c
Talc powder	*T. harzianum*	82.4 ±6.2a	82.2 ±1.0a	83.3 ±1.3a
	B. subtilis	72.7 ±1.7b	70.5 ±1.6b	73.7 ±1.0b
	P. fluorescens	63.9 ±2.7c	60.0 ±4.2c	64.2 ±2.4c
Fresh cultures**	*T. harzianum*	86.8 ±1.1a	86.6 ±0.7a	88.8 ±0.0a
	B. subtilis	70.5 ±1.5b	67.8 ±1.2b	76.7 ±0.7b
	P. fluorescens	67.8 ±1.4c	69.4 ±0.7c	69.6 ±0.6c

* Formulated stored Antagonistic microorganism (after 6 month of storage)

** Fresh cultures of fungal and bacterial suspensions

*** Pathogenic fungal growth reduction relatively to control

Means ± standard deviations within a column followed by the same letter are not significantly different by Duncan multiple range test at $P < 0.05$

tested biomarkers either used as a new suspension or formulated on a carrier of CMC, sawdust and talc powder had a significant reduction in the growth of pathogenic inhibitors, *R. solani*, *M. phaseolina* and *F. solani*. In this concern, *T. harzianum* had a superior inhibitory effect against the growth of pathogenic fungi followed by *B. subtilis* and *P. fluorescens*, respectively.

No significant differences were observed between the effects of the inhibitor of fresh suspension and the six-month antimicrobials stored for all tested biological agents, although the highest pathogen growth rate was observed in the suspension of fresh biological agents followed by biocompatible compounds on sawdust, CMC and powder. Talc in descending order. *Fusarium solani* exhibited greater sensitivity against the tested biological agents followed by *M. phaseolina* and *R. solani*, respectively. *Fusarium solani* growth decreased by between 69.6-88.8% in the treatment of new bio-suspension followed by 66.6-86.7%, 67.6-84.2% and 64.2-83.3% in stored biomaterials placed on sawdust, CMC and Talc powder carriers, customized. A similar trend was also observed in treatments of *M. phaseolina* and *R. solani*.

In this study, records of corn grains containing six months of stored fungal and bacterial biological agents showed an inhibitory effect against the pathogens of root rot under laboratory conditions. These findings are also confirmed by many researchers (Bell *et al.,* 1982; Abdel-Kader 1997; El-Mougy 2001).

The results obtained in the current work table 1 confirmed the frequency of the El-Mougy and Abdel-Kader (2008). They tested the effect of laboratory inhibitors on anti-fungal and bacterial infection agents applied either to growth-growing tablets or bean seeds. The researchers found that the effect of the inhibitor of *T. viride*, *T. Harzianum*, *B. subtilis* and *P. fluorescens* highly significantly control than *T. hamatum* and *B. cereus*, respectively. They added that, similar results were also obtained when antifungal agents were applied as biologically primed seeds.

Greenhouse experiment

The efficacy of bio- primed corn grains with six months of stored different fungi and bacteria anti-root rot pathogens was evaluated in a pot experiment using artificial infested soil with root pathogens under greenhouse conditions. Incidence of maize root rot before and after appearing above soil surface is shown in (Table 2 and Fig. 1). The data indicate that bio-coated grains used in soil infested with *F. solani* showed a very significant effect causing a complete reduction (100%) in the occurrence of root rot in both stages of plant growth before and after its appearance above soil surface compared to 30.0 and 35.0% in untreated control therapy. Furthermore, the lowest root rot rate of 3.3 and 6.8%, respectively, was recorded in both plant growth phases of fresh bio-primed grains when infected with *M. phaseolina* compared with six months of stored biological agents and untreated control as well.

Bio-priming Corn grains with six-month stored bioagents showed a less protective effect against the occurrence of the disease. Bio-primed grains showed a marked decrease in the incidence of pre-emergence root rot, ranging from 3.3 to 12.2%; 10-16.6%; 3.3-6.6% for fungal and bacterial bio grains grown in artificially infested soil with *R. solani*, *M. phaseolina* and *F. solani* compared with untreated control treatments, which recorded 33.3, 30.0 and 33.3%, in the respective order. Also, post-emergence root rot ratios ranged from 6.8-13.9%, 8.6-14.6%; 7.1-11.8% for fungal and bacterial bio-treated grains in artificially infested soils with *R. solani*, *M. phaseolina* and *F. solani* compared to check control treatment, which recorded 35.0, 33.3 and 35.0%, according to each relevant treatment.

On the other hand, the data in table 2 revealed that the highest effect of bio-factors was observed in treatment of applied grains bio-primed with fresh bioagents suspension which recorded the average root rot rate in the range of 3.9-4.5% compared to 33.3% in grains in untreated. Furthermore, biomarkers designed for sawdust showed a significantly lesser incidence of disease than those placed on CMC and Talc powder, in respective order. The incidence of root rot in these treatments varies in mean between 6.2-8.0%, 8.6-11.2% and 11.2-13.6%, in the order adopted according to the carrier used. Referring to presented data in table 2 the general mean of disease incidence for formulated bioagents stored in different carriers revealed 7.0, 9.9 and 12.6%. In other word, Talc powder affected the antagonistic efficacy of tested bioagents than those of CMC and Sawdust carriers in descending order.

Illustrated data in Fig. 1 demonstrate that in artificially infested soil with root rot pathogens, *R. solani*, *M. phaseolina* and *F. solani* the sown maize grains primed with fresh cultures of tested bioagents revealed superior efficacy for reducing root rot disease incidence at both plant growth stages compared with the same formulated stored bioagents and untreated control as well. In this regard, fresh cultures of tested bioagents caused disease reduction calculated as a range of 79.5 up to 100% and from 50.8 up to 100% at pre-, and post-emergence growth stages, respectively. Meanwhile, at the same growth plant stages lower disease reduction was recorded with stored bioagents on different carriers although they highly differed significantly than untreated control. The calculated disease reduction was in a range of 69.9-90.0%, 54.0-98.8% for *R. solani*; 44.6-71.4%, 51.9-77.7% for *M. phaseolina* and 80.1-90.0% for *F. solani*, in relevant order at both plant growth stages.

The results obtained in this study are in harmony with other reports by several investigators. In this regard, the biological control of seedling diseases that use fungi and hostile bacteria has received increasing attention. Taylor & Harman (1990) and (Wheeler 1983) reported that antimicrobial coated seeds have the ability to colonize the ground roots of the target seedlings, and therefore they are able to present or close to a pathogenic infection court, and thus can work by producing anti-compounds for fungi or antibiotics. Biological preservatives with any fungal or bacterial biological agents applied as a seed treatment are an attractive delivery system that may protect colonized seeds and protect roots and may increase plant growth (Sivan and Chet 1986; Chao *et al.,* 1986; Chang *et al.,* 1986; Chang *et al.,* 1986; Wright *et al.,* 2003). A process known as "awareness" or "preparation" has been identified by Conrath *et al.,* (2002) as an antibiotic effect on plant resistance to pathogens by stimulating the basal level of defensive reactions after treatment as well as enhancing the activity of cellular defense responses.

Moreover, many researchers used the terms "bio-preparation to protect seeds and seedlings from pathogen invasion. In this concern, two different techniques were used to achieve bio-preparation against the erosion of lithium in tomatoes and sweet corn. The first was the addition of *T. harzianum* directly to the priming of the solid matrix (Harman and Taylor 1988). In the meantime, the suspension of *P. fluorescens* was added to methyl cellulose (1.5%) and sterile surface-coated sweet corn grains before wetting the grain between wet paper towels (Callan *et al.,* 1990).

In this study, bio-maize preparation treatments were applied through coating granules with prepared biomaterials stored on sawdust, CMC and Talc powder 12 hours before seeding to allow seed colonization before drying. Jensen *et al.,* (2004) found that *Clonostachys rosea* colonized the entire surface of the pericarp, including the apex, of carrot seeds where the main root appears. This observation is based on microscopic evaluation of the growth and distribution of the antagonist during the seed preparation process. These reports are consistent with the results obtained in this study.

Moreover, Vidhyasekaran and Muthamilan (1995) recorded that under *in vitro* studies *P. fluorescens* strains had inhibitor action against chickpea wilt disease (*Fusarium oxysporum* f. sp. *Ciceris*). They trailed various carriers for assessing their efficacy on the population of these strains during storage. In addition, they illustrated that the bacteria survived more than 8 months of storage; also the treated seeds were sown in soil, the antagonist moved and survived well in the

Table 2: Effect of Maize grains dressing with stored formulated bioagents against root rot disease incidence under greenhouse conditions.

Maize grains dressing		Root rot incidence (%)						
		Soil infestation						
Carrier	Formulated antagonist*	*R. solani*		*M. phaseolina*		*F. solani*		Mean
		Pre-emergence ***	Post-emergence ****	Pre-emergence ***	Post-emergence ****	Pre-emergence ***	Post-emergence ****	
CMC	*T. harzianum*	10.0±10.9d	7.4±1.9f	13.3±10.0c	7.6±1.8f	6.6±9.5e	7.1±1.6f	8.6
	B. subtilis	10.0±10.9d	11.1±2.0d	13.3±10.0c	11.5±1.9d	6.6±9.5e	7.1±1.6f	9.9
	P. fluorescens	10.0±10.9d	11.1±1.9d	16.6±8.6b	16.0±2.1c	6.6±9.5e	7.1±1.6f	11.2
Mean		10.0	9.8	14.4	11.7	6.6	7.1	9.9
Sawdust	*T. harzianum*	3.3±7.5f	6.8±1.6f	10.0±10.3d	7.4±1.7f	3.3±7.5f	6.8±1.6f	6.2
	B. subtilis	3.3±7.5	6.8±1.6f	10.0±10.3d	7.4±1.7f	3.3±7.5f	10.3±1.7e	6.8
	P. fluorescens	3.3±7.5f	6.8±1.6f	10.0±10.3d	11.1±1.9d	3.3±7.5f	13.7±2.6d	8.0
Mean		3.3	6.8	10.0	8.6	3.3	10.2	7.0
Talc powder	*T. harzianum*	10.0±10.3d	11.5±1.9d	16.6±8.6b	12.0±3.0d	6.6±9.5e	10.7±1.8e	11.2
	B. subtilis	13.3±10.0c	15.3±1.8c	16.6±8.6b	16.0±1.9c	6.6±9.5e	10.7±1.8e	13.0
	P. fluorescens	13.3±10.0c	15.3±1.8c	16.6±8.6b	16.0±1.9c	6.6±9.5e	14.2±1.7c	13.6
Mean		12.2	13.9	16.6	14.6	6.6	11.8	12.6
Freshcultures**	*T. harzianum*	3.3±7.5f	10.3±1.7d	3.3±7.5f	6.8±1.2f	0±0.0g	0±0.0g	3.9
	B. subtilis	3.3±7.5f	13.7±3.2d	3.3±7.5f	6.8±1.2f	0±0.0g	0±0.0g	4.5
	P. fluorescens	3.3±7.5f	17.2±3.1b	3.3±7.5f	6.8±1.2f	0±0.0g	0±0.0g	5.1
Mean		3.3	13.7	3.3	6.8	0	0	4.5
Un dressed grains control		33.3±12.9 a	35.0±4.5a	30.0±12.7 a	33.3±3.2 a	33.3±12.9a	35.0±3.4a	33.3

* Formulated stored Antagonistic microorganism (after 6 month of storage).

** Fresh cultures of fungal and bacterial suspensions.

*** Pre-emergence root rot incidence (%) was calculated as the number of absent seedlings relative to the number of grains sown.

**** Post-emergence root rot incidence (%) was calculated as the number of diseased plants relative to the number of emerging seedlings.

Means ± standard deviations within a column followed by the same letter are not significantly different by Duncan multiple range test at $P < 0.05$

rhizosphere area.

This occurred in the present study, under greenhouse conditions, when plants were examined in soil that had been artificially infested with disease incidents. The present findings demonstrate that the evaluated formulated antagonistic micro-organisms were able to survive and keep their antagonistic viability throughout the six months storage period. Therefore, it could be hypothesized that the formulated bioagents in Sawdust, CMC and Talc powder and used as grains bio-priming even after six months of storage could kept the antagonists' ability to grow and survive competitively.

Conclusion

On the light of the results obtained in the present study the evaluated technique could be recommended. Corn-coated with granules containing fresh or stored antifungal or bacterial antibiotics have proven to be an effective way to provide grain with protective effects against soil-borne pathogens, which significantly reduce the incidence of root rot disease. The use of bio-priming seeds could be considered a safe, cheap and easily applied biocontrol method to be used against soilborne plant pathogens, particularly in organic farmers for avoiding environmental pollution.

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Compliance with ethical standards

Conflict of interest

The authors declare that they have no conflict of interest.

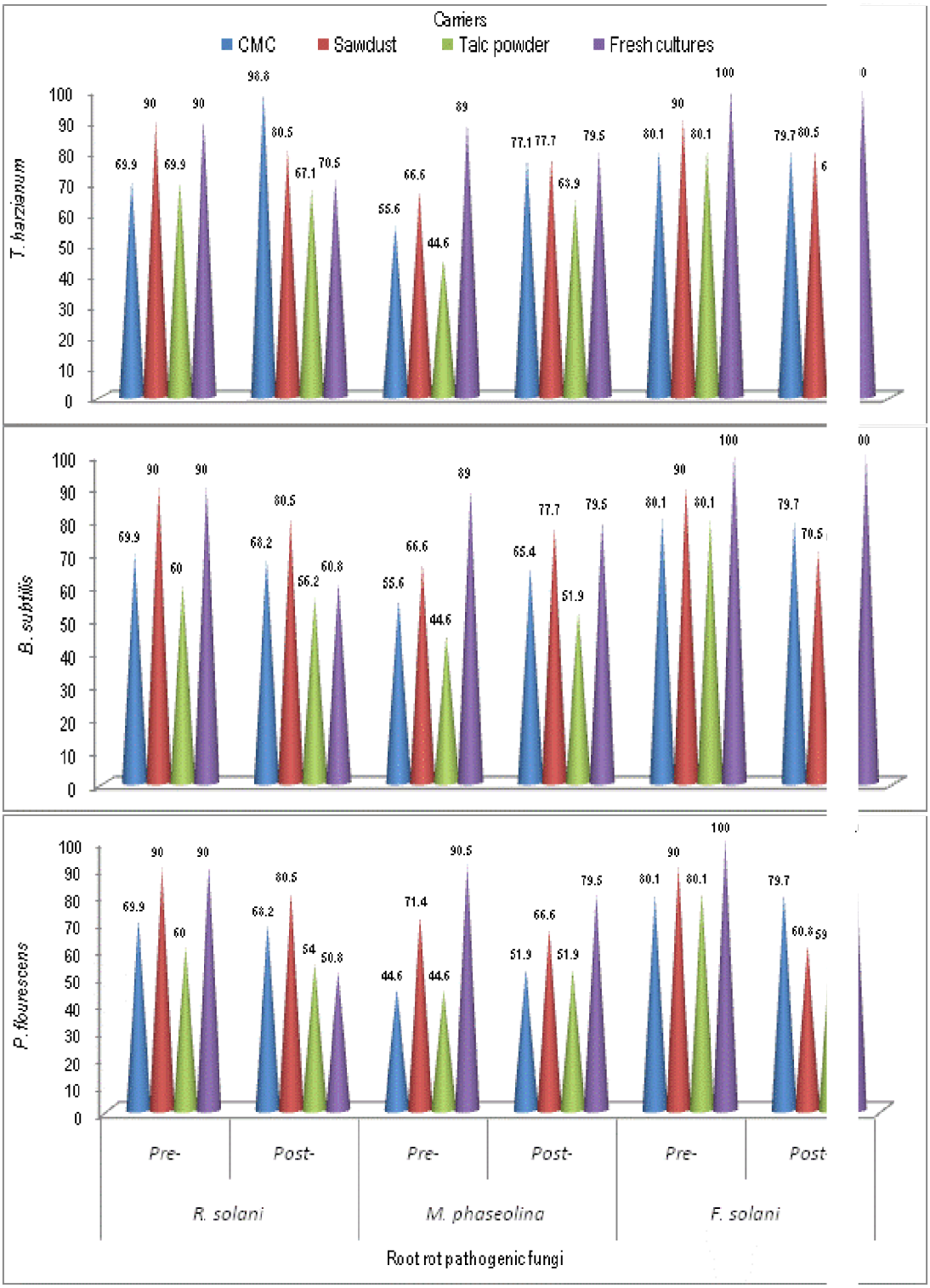

Fig. 1: Reduction in root rot disease incidence at pre-, and post-emergence plant growth stages in response to Maize grains dressing with stored formulated bioagents under greenhouse conditions.

Statement of human and animal rights

This article does not contain any studies with human or animal subjects performed by the any of the authors.

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