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Biopriming: A novel seed treatment options to manage the seed-borne fungal infection of tomato

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Abstract
Tomato is the most popular vegetable crop grown in the world. Crop is severely affecting many of seed-borne fungal infection. Those seed-borne fungal infections can be managed by seed treatment with bio-control agents along with priming agents (bio-priming).Among four priming agents with the selected two bio agents were tested for their efficacy in overcoming seed borne fungal infections and seed quality parameters of tomato under *in vitro* by using rolled towel method. Seed treatment with *Pseudomonas fluorescens* + jelly were found most effective compared to *Pseudomonas fluorescens* alone with respect per cent seed germination, seed infection and vigour index it indicating the role of jelly in enhancing efficacy of *P. fluorescens*, jelly being hydrophobic polymer, can imbibe water and provides suitable conditions for the multiplication of bio agent *P. fluorescens.*

Keywords: Seed borne fungi, tomato, bioprimig, vigour index

Introduction
Tomato (*Solanum lycopersicum* Mill.) is the most popular vegetable crop grown in the world, crop offers significant nutritional advantages, as it contains significant source of dietary lycopene, β-carotene, vitamin C and antioxidant properties in a low energy dense food (Britt and Kristin, 2011) [4]. Several human studies indicated a relationship between a high intake of tomato products and a decreased risk of several types of cancer, atherosclerosis and cardiovascular diseases (Cecilia *et al*., 2010) [6]. Recently, this crop is recognized as a model for plant-pathogen interactions (Arie *et al*., 2007) [3].
Several diseases affecting tomato are caused by fungi, bacteria, viruses and nematodes and many of them are seed-borne in nature. These seed-borne pathogens are known to cause economically important diseases like early blight, late blight, Fusarium wilt, Septoria leaf spot, damping off and fruit rot. Seed-borne fungi are of considerable importance due to their influence on the overall health, germination and final crop stand in the field. The infected seeds may fail to germinate, or transmit disease from seed to seedling and/or from seedling to growing plant (Islam and Borthakur, 2012) [9]. The disease caused by seed-borne fungi may lead to seed abortion or elimination of germination capacity. Thus, the disease control programme is important at each stage of growth. Seed treatment with bio-control agents along with priming agents may serve as an important means of managing many of the soil and seed-borne diseases, the process often known as 'bio-priming". The bio-priming seed treatment developed for control of *Pythium* seed rot of sweet corn involves coating seed with a bacterial biocontrol agent such as *Pseudomonas aureofaciens* Kluyver AB254 and hydrating for 20 h under warm (23 ^{0}C) conditions in moist vermiculite or on moist germination blotters in a self-sealing plastic bag. The seeds are removed before radical emergence. The bacterial bio-control agent may multiply substantially on seed during bio priming (Callan *et al*., 1990) [5]. Seed encapsulation with beneficial microorganisms is now becoming common. The priming agents along with bio agents can protect the seeds from biotic as well as abiotic stresses like moisture stress and thereby they can protect the seeds and the seedlings for extended periods. Hence there is a need to screen the new and the available seed dressing molecule of bio agents along with priming agents for their efficacy in overcoming the seed-borne fungal infections in tomato.

Material and Method
The present investigation was carried out during the year 2015-16, to know the efficacy of different seed dressing priming agents and two best seed dressing bio agents were selected for the eliminating the seed-borne fungal infections in the infected tomato seed sample of variety PKM-1.In bio-priming, two grams of tomato seeds were treated with bacterial bio-control

Agent *Pseudomonas fluorescens* with priming agents like vermiculite, coir pith, jelly and coco peat. Same treatment was followed for fungal biocontrol agent *Trichoderma harzianum.* After Pre-soaking of seeds in sterile distilled water, seeds were coated with powder formulations of *P. fluorescens and T. harzianum.*at 0.8 per cent concentration along with moist vermiculite in the proportion of 2:1 (2 parts of vermiculite and 1 part of seed) and mixed thoroughly to give uniform coating. These seeds were dried in shade and stored at 25± 2 °C for 24 h in a self– sealing plastic bags. The treated seeds were tested three replications of 100 seeds by employing paper towel method in complete randomized block design (CRD). These paper towels were incubated at 25±2 °C for twelve days under 12 h light and 12 h darkness. After twelve days of incubation, per cent germination and per cent infection were recorded and seedling vigour was calculated as stated earlier. The same procedure was employed for coco peat, coir pith also.

For bio-priming in jelly, (a commercial preparation containing water absorbent organic polymer which absorbs, stores and then keeps releasing the moisture slowly). 10 g of jelly was poured into 500ml of cold sterilized water and kept in water 5 to 6 hours till the small pieces turned into sparkling jelly cubes. After pre-soaking of seeds in sterilized water, seeds were coated with powder formulations of *P. fluorescens* and *T. harzianum*at 0.8 per cent concentration along with the jelly cubes in 2:1 proportion as stated earlier. Such treated seeds were tested in three replications of 100 seeds by employing paper towel method. These paper towels were incubated at 25±2 °C for twelve days under 12 h light and 12 h darkness. After twelve days of incubation per cent germination, per cent infection was recorded and seedling vigour was calculated by the following formula, given by Abdul and Anderson (1973) [1] *i.e.*

Vigour Index = Seed germination (%) × Seedling length (Shoot +Root length (cm)

$$\% \text{ seed germination} = \frac{\text{No of seed germinated}}{\text{Total no. of seed used}} \times 100$$

$$\% \text{ seed infection} = \frac{\text{No of seed infected}}{\text{Total no. of seed used}} \times 100$$

Results and Discussion

Four priming agents with the selected two bio agents were tested for their efficacy in overcoming seed borne fungal infections of tomato (Variety: PKM-1) under *in vitro* by using rolled towel method and the results are presented in Table 1. Among the four priming agents treated with respect to *Pseudomonas fluorescens*, seed treatment with *Pseudomonas fluorescens* + jelly recorded significantly higher per cent seed germination (98.33%) and vigour index (1235.57) with least per cent seed infection (10.00%) compared to seed treatment with *Pseudomonas fluorescens* @ 0.8% alone, it indicating the role of jelly in enhancing efficacy of *P. fluorescens*, jelly being hydrophobic polymer, can imbibe water and provides suitable conditions for the multiplication of bio agent *P. fluorescens.*

Table 1: Efficacy of bio priming agent as seed dressers against seed-borne fungal infection and seed quality parameters of tomato

Sl. No.	Treatments	Per cent seed infection	Per cent seed germination	Vigour index
1	*P. fluorescens* alone @ 0.8%	14.00 (21.96)*	92.00 (73.57)	1291.07
2	*P. fluorescens* @ 0.8% + Jelly	10.00 (18.43)	98.33 (82.67)	2058.23
3	*P. fluorescens* @ 0.8% + Vermicullite	12.33 (20.56)	95.33 (77.87)	1738.33
4	*P. fluorescens* @ 0.8% + Coco peat	12.00 (20.27)	96.33 (78.98)	1916.53
5	*P. fluorescens* @ 0.8% + Coir pith	13.33 (21.40)	94.00 (75.82)	1316.87
6	*T. harzianum*alone @ 0.8%	14.67 (22.52)*	91.67 (73.23)	1235.57
7	*T. harzianum*@ 0.8% + Jelly	12.67 (20.85)	95.00 (77.08)	1811.33
8	*T. harzianum*@ 0.8% + Vermicullite	13.67 (21.69)	92.67 (74.30)	1640.77
9	*T. harzianum*@ 0.8% + Coir pith	15.33 (23.04)	91.33 (72.90)	1212.60
10	*T. harzianum*@ 0.8% + Coco peat	11.33 (19.67)	97.00 (80.03)	2091.97
11	Control	29.67 (33.00)	68.67 (55.96)	1007.07
	Mean	14.45 (22.13)	92.03 (74.76)	1574.58
	S.Em±	0.39	0.75	20.27
	CD at 1%	1.57	2.98	80.81

*Figures in parentheses indicate arcsine transformed values

The present findings are in accordance with the findings of El-Mohamedy and Abd El-Baky (2008) [7] who investigated / evaluated the efficacy of different types of seed treatments i.e., bio-priming, seed coating with bio-control agents (*T. harzianum, Bacillus subtilis* and *P. fluorescens*) seed priming and seed dressing with these antagonistic micro-organisms in control of root rot disease incidence compared to other treatments. Ravindra *et al.* (2014) [10] evaluate the effect of bio priming with bacterial strains of *Enterobacter* spp. on seed germination and seedling growth of tomato.

With respect to *Trichoderma*, seed treatment with *Trichoderma harzianum*+ coco peat recorded significantly higher per cent seed germination (97.00%) and vigour index (2091.07) with least per cent seed infection (11.33%) compared to seed treatment with *Trichoderma harzianum* alone. Harman *et al.* (1989) [8] reported increased plant stand in soils infested with *F. graminarum* and *Pythium ultimum* by priming of wheat seeds with *Trichoderma harzianum*. They further reported that in field trial, stands of peas were not significantly enhanced by seed treatment with *T. harzianum* Strains in the absence of priming but were improved by *T. harzianum*+ vermiculite. Ananthi *et al.* (2014) [2] evaluated the efficacy of bio-priming with the bio-control agents *Trichoderma viride* or *Pseudomonas fluorescens* in order to improve seed germination and seedling vigour using PKM 1 chilli seed.

Seed priming (or) osmo-conditioning refers to a pre-sowing hydration treatment developed to improve seedling establishment. Primed seeds can be handled in a manner similar to conventional seeds and a seed company can perform this process. On-farm seed priming seems to be a robust, widely applicable technology and its effects are generally independent of the crop variety used. This is important, because priming can be used to add value to the benefits achieved by using improved modern varieties or by

adoption of other improved technologies such as fertilizer or better crop protection.

Seed treatment with bio-control agents along with priming agents may serve as an important means of managing many of the soil and seed-borne diseases, the process often known as bio-priming. However, this may not give additional benefits under optimum conditions.

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