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EFFECT OF *PENICILLIUM* EXTRACTS ON GERMINATION VIGOR
IN SUBSEQUENT SEEDLING GROWTH OF TOMATO
(*SOLANUM LYCOPERSICUM* L.)

ABSTRACT

Penicillium spp. are well known to produce a variety of beneficial metabolites for plant growth and survival, as well as defend their hosts from attack of certain pathogens. In this study, effects of culture filtrate of different *Penicillium* spp. were tested on tomato seeds. On the whole, presoaking of seeds in filtrates of the nine *Penicillium* isolates tested, significantly increased seed germination when compared with the control seeds. Cultural extracts of *P. expansum* and *P. billi* were highly effective in growth promotion up to 90%. It was also observed that *P. implicatum* and *P. oxalicum* significantly enhanced the root growth in tomato seedling as compare to other species. In case of shoot length, *P. verrucosum* (3.38), *P. granulatum* (2.81) and *P. implicatum* (2.62) were effective. However *P. implicatum* was quite promising to increase shoot and root length in tomato seedlings. Where as *P. simplicissimum* and *P. citrinum* were less effective on seedling growth. The plant growth promoting ability of *Penicillium* strains may help in growth promotion in other plants and crops. *Penicillium* spp. are already known for producing mycotoxin and enzymes. Plant growth promoting ability of *Penicillium* spp will open new aspects of research and investigations. The role of *Penicillium* spp. in tomato plant growth requires further exploration.

Key words: Fungal extract, *Penicillium*, metabolites, seedling growth, tomato

INTRODUCTION

Fungi produce metabolites in their metabolic processes. These metabolites are products of some amino acids, cyclic peptides, aromatic, phenols, terpenoids and plant growth regulators (Griffin, 1981; Madhosing, 1995; Nema, 1992). These metabolites are many and diverse and they are known to cause diseases in plants, animals and humans who eat infected food (Schumann 1991, Singh *et al.*, 1991). Fungi

of the genera *Aspergillus*, *Fusarium*, *Penicillium* and *Rhizoctonia* are commonly known to produce toxic substances that affect health of plants.

Species of *Penicillium* are ubiquitous saprobes, whose numerous conidia are easily distributed through the atmosphere and detected with high frequency in soils (Domsch *et al.*, 1993). *Penicillium* species have been reported to be able to cause different diseases in many plant species (Umemoto *et al.*, 2009, Valdez *et al.*, 2006, Payne 1999, Jarvis 1990). However, antagonistic potential of *Penicillium* species to suppress plant pathogens is also well reported (Phuwiwat and Soy 2001). Beneficial effects of the introduction of specific microorganisms on plant growth have been reported for numerous crops, including tomato (*Solanum lycopersicum* L.) grown under field (Kokalis-Burelle *et al.*, 2002; Guo *et al.*, 2004) or greenhouse conditions in organic media (Gagne' *et al.*, 1993). A little is also known about plant growth stimulants/regulators produced by the *Penicillium* spp. (Hamayun *et al.*, 2010, Alfredo *et al.*, 2007, Phuwiwat and Soy 2001). The present study was therefore, undertaken to evaluate the effect of filtrates of *Penicillium* species on Tomato seeds germination and seedling vigor.

MATERIALS AND METHODS

Fungal Isolates

Penicillium species were isolated from different sources (Table. 1) using the dilution plate technique (Waksman, 1922) on 2% Malt Extract Agar (20g malt extract, 17g agar and 1,000 ml of distilled water). The test fungi were grown on MEA medium in Petri plats at 28 °C for 7 days.

Table 1

List of *Penicillium* isolates

FCBP #	<i>Penicillium</i> species	Sources
022	<i>P. simplicissimum</i>	Onion
024	<i>P. citrinum</i>	Bread
025	<i>P. oxalicum</i>	Rhizosperic soil of Grapes,
1052	<i>P. verrucosum</i> var. <i>cyclopium</i>	Lemon fruit,
1069	<i>Penicillium</i> sp.	Bread
1079	<i>P. billii</i>	Bread
1080	<i>P. granulatum</i>	Grape fruit,
1102	<i>P. expansum</i>	Garlic, Lahore
1109	<i>P. implicatum</i>	Pomgranate fruit,

Culture Filtrate Preparation

Fungal filtrate of each test species was prepared. A disc (0.5 cm diameter) of mycelia was taken from the periphery of 6-day-old culture of each fungus grown on (MEA) medium was introduced into 250-ml conical flasks, each containing 100 ml of 2% Malt Extract (ME) broth. The flasks were incubated at 25 ± 2 °C for 12 days. The fungal filtrates were obtained by passing the 12 days old culture broth through sterile membrane filter No.0.20 μ to obtain a cell free filtrate.

Seed Selection and Effect of Fungal Filtrated on Seed Germination

Healthy seeds were selected and surface sterilized for 5 minutes with 1% sodium hypochlorite solution. Seeds were then rinsed three times with sterile water and were air dry on metallic mash. Sterile dry tomato seeds were soaked in 12-day-old undiluted fungal filtrates of each of the fungi for 30 minutes. The control seeds were pre-soaked in sterile distilled water for the same periods of time. At the end of each pre-soaking period, the seeds were removed from the filtrates and were placed in Petri plates lined with two layers of Whatman filter paper soaked in sterile distilled water. In each Petri plate, ten seeds were placed. Plates were incubated at 25 °C under dark. Germination counts were made after 12 days of incubation period.

Germination percentage ($GP_{[\%]}$) of tomato seeds were calculated as:

$$GP_{[\%]} = \frac{N}{T} \times 100$$

Where N —number of germinated seeds, T —number of seeds

The seedling shoot and roots length were also measured. The experimental design was a complete randomized design with 3 replicates.

RESULTS AND DISCUSSIONS

The effect of presoaking seeds in undiluted filtrates of *Penicillium* species fungi for 30 minutes is shown in Fig. 1. On the whole, presoaking of seeds in filtrates of the nine *Penicillium* isolates tested, significantly increased seed germination when compared with the control.

The percentage of germination also varied also with respect *Penicillium* species. Cultural extracts of *P. expansum* and *P. billi* were the highly effective in growth promotion up to 90% as compare to control. While filtrates of *P. oxlaicum* and *Penicillium* sp. gave the lowest percentage of germination in all test *Penicillium* species, but the percentage of seed germination was significantly higher than control. *Penicillium bilaii*, is also reported to increase inorganic P uptake, dry matter accumulation and yield by wheat (*Triticum aestivum* L.), canola (*Brassica napus* L.), field pea (*Pisum sativum* L.) and field bean (*Phaseolus vulgaris* L.) (Kucey and Leggett 1989 ; Downey and van Kessel 1990). However, the host-growth promotion caused by *P. bilaii* cannot always be attributed to an increase in plant P status (Heisinger 1998).

Although a stimulation in P uptake is a common observation with *P. bilaii*-mediated increases on plant growth (Downey and van Kessel 1990; Kucey and Leggett 1989).

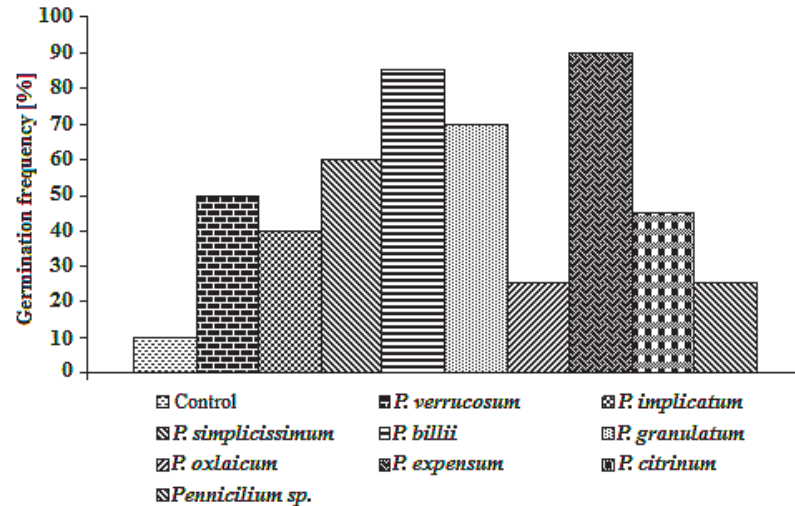


Fig.1. Effect of *Penicillium* extracts on seed germination of tomato

The results in Fig. 2 showed that fungal filtrates of all the test fungi had significant effects on the seedling root length when compared with control. The low growth was still evident in control treatment.

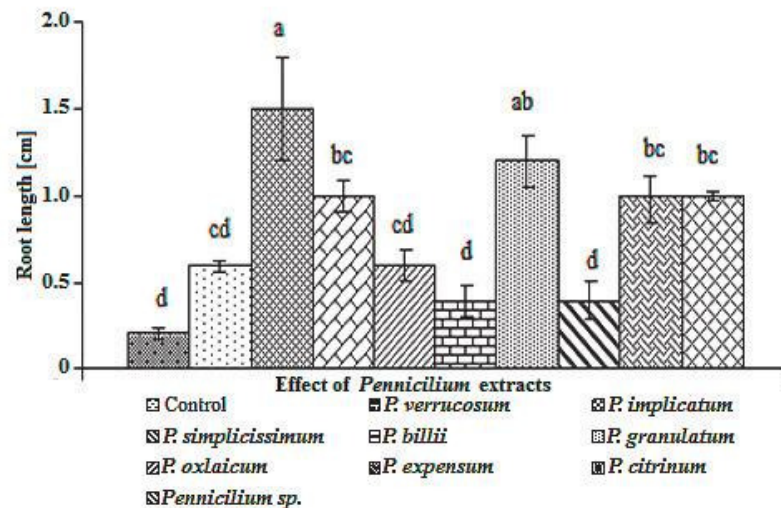


Fig.2. The effect of *Penicillium* extracts on the root length of tomato seedling

It was observed that *P. implicatum* and *P. oxlalicam* significantly enhanced the root growth in tomato seedling as compare to other species (Fig 2.). In case of shoot length, *P. verrucosum* (3.38), *P. granulatum* (2.81) and *P. implicatum*

(2.62) extracts showed effective results. However *P. implicatum* was quite promising to increase shoot and root length in tomato seedlings. Where as *P. simplicissimum*, *P. citrinum* and *Penicillium* sp. were least effective on seedling growth (Fig. 2,3). The production of plant growth regulators by the microorganisms is an important mechanism often associated with growth stimulation (Vessey, 2003). The balance between vegetative and reproductive growth is controlled by hormone signalling within the plant and can therefore be highly influenced by it (Taiz and Zeiger, 1991). Many workers found a plant growth regulator in the culture filtrate of *Penicillium* sp. (Maes *et al.*, 1986; Sedlock *et al.*, 1994). These compound can stimulate seed germination root length and shoot length in many crops (Kimura, *et al.*, 1996). Some studies explain that *P. oxalicum* inoculum can promotes growth of tomato plants (Garcia-Lepe *et al.*, 1997) in different growth conditions which also supports our results.

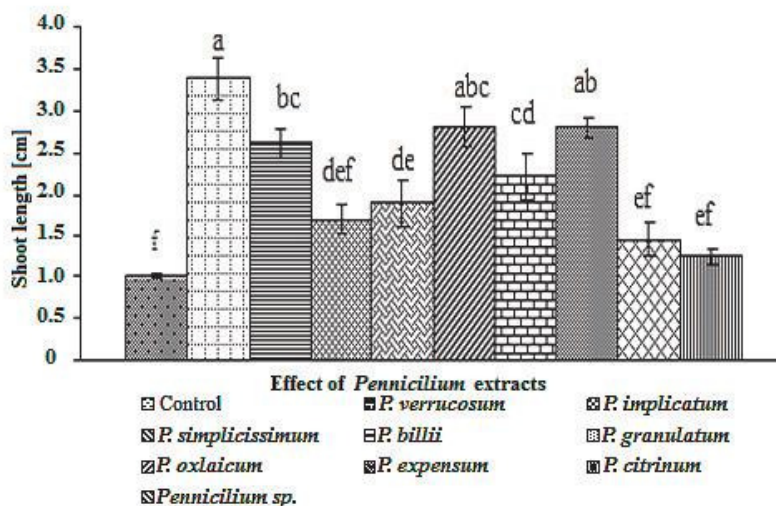


Fig.3. The effect of *Penicillium* extracts on the shoot length of tomato seedling

P. citrinum has been reported as common endophytic fungus of cereal plants like wheat and soybean (Khan *et al.*, 2008) and also as growth promoter. However in present study, *P. citrinum* and *Penicillium* sp. was least effective in our study. We can conclude that the cultural filtrate of *Penicillium* spp. can play very important role for supporting effective strains in case of tomato seeds. Current findings contradiction to the previous reports of shoot length promotion by *P. citrinum* culture filtrate application (Hamayun *et al.*, 2010; Choi *et al.*, 2005).

The microbial extracts had been and will continue to be a productive source of biologically active compounds (Tejesvi *et al.*, 2007). From their host growth and possibility of new basis for further investigation in *Penicillium* metabolites.

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