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# Efficacy of seed treatment of fungicides, bio agents and botanicals on seed mycoflora, seed germination and seedling vigour index of mung bean

**RM Dolas, SB Gawade and MC Kasture**

**Abstract**
The seed treatment of mung bean seed with carbendazim @ 0.2 % was found most effective among all the seed treatments. It showed 8.4 per cent seed mycoflora as against 51.2 per cent in control treatment. The reduction in seed mycoflora due to this fungicidal treatment was 83.59 per cent over control. Among the bioagents, *Trichoderma viride* @ 0.6 % showed 20.0 per cent seed mycoflora as against 51.2 per cent in control. The reduction in seed mycoflora due to this bioagent treatment was 60.93 per cent over control. Among botanicals, Garlic extract @ 0.5 % showed 21.5 per cent seed mycoflora as against 51.2 per cent in control. The reduction in seed mycoflora due to this botanical treatment was 58.0 per cent over control. Further, the seed treatment with carbendazim @ 0.2 % showed 87 per cent seed germination as against 69 per cent in control. The increase in seed germination due to this fungicidal treatment was 20.68 per cent over control. Among bioagents, *Trichoderma viride* @ 0.6 % showed 78 per cent seed germination as against 69 per cent in control. The increase in seed germination due to this bioagent treatment was 11.53 per cent over control. Among botanicals, Garlic extract @ 0.5 % showed 75 per cent seed germination as against 69 per cent in control. The increase in seed germination due to this botanical treatment was 8.0 per cent over control. Similarly, the seed treatment with carbendazim @ 0.2 % showed 1592.1 seedling vigour index as against 1166.1 in control. The increase in seedling vigour index due to this fungicidal treatment was 26.75 per cent over control. Among the bioagents, *Trichoderma viride* @ 0.6 % showed 1380.6 seedling vigour index as against 1166.1 in control. The increase in seedling vigour index due to this bioagent treatment was 15.50 per cent over control. Among botanicals, Garlic extract @ 0.5 % showed 1297.5 seedling vigour index as against 1166.1 in control. The increase in seedling vigour index due to this botanical treatment was 10.12 per cent over control.

**Keywords:** Triclosan, TCS, determination, detection, sensor

## Introduction
Green gram (*Vigna radiata* (L.) Wilczek) is commonly known as mung bean or mung. It is very ancient annual crop in Indian farming. Mung bean is especially grown in Southeast Asia but some are also grown in Africa and America. In India, it is one of the most important pulse crops. It is grown in almost all parts of the country. This crop is sown usually as dry land crop in almost all the states of India, namely Madhya Pradesh, Bihar, Uttar Pradesh, Andhra Pradesh, Rajasthan, Karnataka and Maharashtra. It is an excellent source of high quality protein and consumed in different ways. Ascorbic acid (Vitamin C) is synthesized in sprouted seeds of mung bean with increment in riboflavin and thiamine. Since mung bean is a leguminous crop, it has the capacity to fix atmospheric nitrogen through symbiotic nitrogen fixation. It is also used as green manure crop. Being a short duration crop it also provides an excellent green fodder to the animals.

Green gram is a highly nutritious containing 24 per cent of high quality protein, 1.3 per cent fats, 56.6 per cent carbohydrates and 3 per cent dietary fibers. It is rich in minerals having 140 mg calcium, 8.4 per cent iron and 280 mg phosphorous. It also contains 0.47 mg vitamin $B_1$, 0.39 mg vitamin $B_2$ and 2 mg niacin. It has calorific value of 334 calories per 100 g of edible protein (Baldev *et al*., 2003) [3].

India is the world's largest producer as well as consumer of green gram. It produces about 1.5 to 2.0 million tons of mung bean annually from about 3 to 4 million hectares of area with an average productivity of 500 kg per hectare. Green gram output accounts for about 10-12 % of total pulse production in the country. Mung production in the country remained stable more than a decade through the 2000s at around 10 to 15 lakh tons. But a sudden jump in output was noted in 2010-11 to 1.75 million tonnes primarily on account of rise in production from Madhya Pradesh, Rajasthan and Tamil Nadu. In 2014-15 the mung bean production in India was 1.39 million tonnes in which, Maharashtra's contribution was about 20 %, while Rajasthan was highest having 26 % of the total production.

Mung bean production in the country is largely concentrated in five states *viz*., Rajasthan, Maharashtra, Andhra Pradesh, Gujarat and Bihar. These five states together contribute for about 70 % of total Mung production in the country. There is a distinct change in production pattern of mung bean across states. Traditionally Rajasthan, Maharashtra, Andhra Pradesh are major mung bean producing states. But there is significant rise in production from other states in recent years particularly, from Tamil Nadu, Uttar Pradesh and Gujarat. Nevertheless production remained volatile across the years with respect to most of the states. As per the latest available estimates, Rajasthan, Maharashtra occupies the first two positions, contributing over 45 %. Andhra Pradesh contributes about 10 % while together Gujarat and Bihar account for about 13 % of total production in the country (Anonymus, 2015).

Seed borne diseases are regarded as major constraints in mung bean production. Infected seeds serve as the source for the spread of the pathogen in disease free area. Seed infection affects the import and export adversely because the seed affected with microbes is not acceptable in international market.

Seeds are the carrier of fungal flora either externally or internally. The variety and intensity of fungal flora changes area-wise and depends upon climate under which seed produced storage or in field, if, not controlled. Also they reduce seed quality i.e. seedling vigour and germination percentage. So it is necessary to control harmful fungi before causing damage by using suitable available measures i.e. chemicals or bio agents.

The seed borne pathogens associated with seeds externally or internally may cause seed rot, seedling blight and resulting into low germination. Some fungi are associated with testa and cotyledon of seeds infected in form of mycelium, pycnidium and conidia or spores, after germination the infection translation to hypocotyls and stem bases as well as dicotyledonary leaves of seedling. Some fungal seed borne pathogens having ability to kill the seedling or plants and substantially, reduce the productive capacity (Shamsur Rahman *et al.,* 1999) [15]. Seed mycoflora play an important role in determining the quality and longevity of seed.

As many as 16 diseases have been reported on mung bean, many of these diseases have been reported as seed borne. Seedling blight, root rot, stem rot, leaf and pod rot caused by *Macrophomina, Curvularia, Alternaria* are some major fungal diseases of mung bean. Species of *Alternaria, Cladosporium, Fusarium* and *Rhizoctonia* are known to cause seed rot and pre and post-emergence losses in green gram (Khare and Chaubey, 1978; Saxena, 1986 and Patil *et al.,* 1990) [14, 11].

Several workers reported *Alternaria* spp., *Aspergillus flavus, Aspergillus Niger, Cladosporium* spp., *Colletotrichum* spp., *Curvularia lunata, Fusarium* spp., *Macrophomina phaseolina, Phoma medicaginis* and *Rhizopus* are seed borne and seed transmissible (Raut and Ahire, 1988; Patil *et al.*, 1990) [11]. The above mentioned fungi are potentially harmful for cultivation of mung bean. So it is better to use some protective measures to control these pathogens.

## Material and Method

### Mung bean seeds

To study the mycoflora associated with seeds of mung bean and to test the efficacy of bio agents, botanicals and fungicides on seed mycoflora, seed germination and seedling vigour index, the seeds of mung bean varietiy Vaibhav were collected from Pulses Improvement Project, Mahatma Phule Krishi Vidyapeeth, Rahuri, Dist. Ahmednagar and Oilseed Research Station, Jalgaon.

### Glass wares

The standard corning brand glasswares *viz.,* petriplates, conical flasks, slides and test tubes were used.

### Equipments

The laboratory equipments *viz.,* autoclave, laminar flow cabinet, incubator, sterio-binocular microscope, research binocular microscope and weighing balance were used.

### Incubation room

The incubation room was used for keeping the blotter plates. The temperature of incubation room was $20 \pm 2^0$C controlled automatically with alternate cycle of 12 hrs. light and 12 hrs darkness (Automatically controlled by electronic timer).

### Miscellaneous material

Pointed needles, inoculating needle, forceps, blotting papers, scissor, glass marking pencil, glass rods, cover slips, towel papers, mercuric chloride, spirit lamp and sterilized water etc. were used.

### Seed treatment with bio agents, botanicals and fungicides

The bio agents i.e. *Pseudomonas fluorescens* @ 0.6 per cent and *Trichoderma viride* @ 0.6 per cent alone were used to find out their effect on seed mycoflora, seed germination and seedling vigour index. Talc based formulations of these bio agents were used for the seed treatment in mung bean. The weight of talc based formulations of bio agents were taken on weighing balance as per the dose and mixed with seeds of mung bean. The material was slightly moistened with sterilized water, shakes slightly so as to cover the entire seed surface by bio agent and then was used for blotter test and seed germination.

In botanical extracts, the crude extracts prepared from ginger rhizome and garlic cloves were used for treatment to the mung bean seeds. The concentration of crude extract was taken with the help of sterilized pipette and mixed with seeds so as to smear total surface of the seeds. Then this seed was used for blotter test and seed germination.

In fungicidal seed treatments, required quantity of the fungicides, on the basis of their concentration was weighted accurately. The fungicide was mixed in seed, moistened with sterilized water, shake the petridish containing seeds and fungicides gently so as to cover the seed surface by fungicide. After drying, the seeds were used for blotter test and seed germination.

**Table 1:** Fungicides, bioagents used and their botanicals

| S. No. | Fungicides, Bioagents, Botanicals | Chemical name | Active Ingredient | Conc. | Manufacturer |
|---|---|---|---|---|---|
| 1. | Captan 50 % WP | N-(trichloromethyl thio-4) Tetrachlorohexane-1-2-dicarboximide | 50 % WP | 0.2 % | Rallis India Ltd. Mumbai |
| 2. | Carbendazim 50 % WP | 2-(Methoxy arbonyl amino) benzimidazole | 50 % WP | 0.2 % | BASF India Ltd., Mumbai |
| 3. | Mancozeb 75 % WP | Manganese ethylene bisdithiocarbamate zink sulphide | 75 % WP | 0.2 % | Indofil Chemicals Ltd., Mumbai |
| 4. | Carboxin (37.5 %) + Thiram (37.5 %) (Vitavax power) | 5,6- dihydro-2- Methyl-1,4- oxathiin-3-carboxamide (56) | 75 % WP | 0.2 % | Uniroyal Chemical Co. |
| 5. | Propineb | Zinc propylenebis -dithiocarbamate | 70 % WP | 0.2 % | Bayer India Ltd., Mumbai |
| 6. | *Trichoderma viride* | | | 0.6 % | |
| 7. | *Pseudomonas fluorescens* | | | 0.6 % | |
| 8. | Ginger extract | | | 0.5 % | |
| 9. | Garlic extract | | | 0.5 % | |

**SVI:** [Mean root length (cm) + Mean shoot length (cm)] x Seed

**Germination (%)**

The per cent reduction in seed germination and seedling vigour index with the inoculation of pathogens over control was calculated. The data was subjected to statistical analysis (Panse and Sukhatme, 1985)

**Result and Discussion**

The seed treatment of mung bean seed with carbendazim @ 0.2 % was found most effective among all the seed treatments. It showed 87 per cent seed germination as against 69 per cent in control treatment. The increase in seed germination due to this fungicidal treatment was 20.68 per cent over control. Among bio agents, Trichoderma viride @ 0.6 % showed 78 per cent seed germination as against 69 per cent in control. The increase in seed germination due to this bioagent treatment was 11.53 per cent over control. Among botanicals, Garlic extract @ 0.5 % showed 75 per cent seed germination as against 69 per cent in control. The increase in seed germination due to this botanical treatment was 8 per cent over control. The seed treatment of mung bean seed with carbendazim @ 0.2 % was found most effective among all the seed treatments. It showed 1592.1 seedling vigour index as against 1166.1 in control treatment. The increase in seedling vigour index due to this fungicidal treatment was 26.75 per cent over control. Among bio agents, Trichoderma viride @ 0.6 % showed 1380.6 seedling vigour index as against 1166.1 in control. The increase in seedling vigour index due to this treatment was 15.50 per cent over control. Among botanicals, Garlic extract @ 0.5 % showed 1297.5 seedling vigour index as against 1166.1 in control. The increase in seedling vigour index due to this treatment was 10.12 per cent over control.

The above results on seed germination and seedling vigour index are in confirmation with Kulshreshta (1988), Pradeep *et al*. (2000) [12], Krishnamurthy *et al*. (2003) [8], Akhtar *et al*. (2005) [1], Dhutraj and Gokhale (2007) [5], Kar and Sahu (2008) [6], Suryawanshi *et al*. (2008) [16], Koche *et al*. (2009) [7], Chilkuri and Giri (2014) [4], Chilkuri and Giri (2014) [4] and Gawade *et al*. (2016). Kulshreshta (1988) tested efficacy of thiram, bavistin, difolaton, dithane M-45 and cereson and reported bavistin as the most effective fungicide for better emergence, less seedling mortality and better yield in mung bean. Pradeep *et al*. (2000) [12] recorded that seed treatment with Trichoderma viride and Pseudomonas fluorescens reduced the colonies of Aspergillus niger, Aspergillus flavus and Fusarium moniliforme with significantly increased seed germination and seedling vigour index in soybean. Krishnamurthy *et al*. (2003) [8] reported that captafol and bavistin gives effective control on Macrophomina phaseolina and Fusarium spp. upto 98 %. Also Trichoderma harzianum can control these fungi upto 92 % and improve seed germination and vigour. Akhtar *et al*. (2005) [1] observed that in mung bean, seed treatment with Carbendazim + Carbofuran was highly effective against nematode and fungal disease complex followed by seed powder of Azadirachta indica. Dhutraj and Gokhale (2007) [5] reported that thiram and bavistin was effective to control seed mycoflora of mung bean than dithane M-45 and captan. Kar and Sahu (2008) [6] noted that Trichoderma harzianum and Trichoderma viride effectively control Macrophomina phaseolina. Also these biocontrol agent enhanced germination and seedling vigour in mung bean. Suryawanshi *et al*. (2008) [16] tested seven fungicides against Macrophomina phaseolina of mung bean and found that

carbendazim is most effective in both field and lab condition. Koche *et al.* (2009) [7] reported that germination percentage and seedling vigour index of soybean was increased with seed treatment of Thiram + Carbendazim @ 3gm/kg each and also reduced seed mycoflora. Chilkuri and Giri (2014) [4] studied the seed treatment with talc based formulations of *Trichoderma viride* and *Pseudomonas fluorescens*. These bio agents were tested for their efficacy against seed mycoflora and seed germination in green gram. Among these bio agents *T. viride* was found superior in controlling the seed mycoflora and also maximum seed germination was observed in *T. viride*. Chilkuri and Giri (2014) [4] reported that seed treatment with thiram + carbendazim (2:1) @ 3 g/kg of seed was increasing the seed germination, shoot length, root length and seedling vigour index in green gram and black gram. Gawade *et al.* (2016) studied the efficacy of bio agents, botanicals on seed mycoflora and seed quality in mung bean and found that *Trichoderma viride* @ 0.6 % + *Pseudomonas fluorescens* @ 0.6 % was most effective among the bio agents and Garlic extract @ 1 % among botanicals was found most effective in controlling seed borne pathogens and increasing seed germination, seedling vigour index and field emergence

**Table 2:** Efficacy of fungicides, bio agents and botanicals on seed germination and seedling vigour index of naturally infected seeds of mung bean (Cv. Vaibhav)

| S. No. | Treatments | Seed germination (%) | Increase in seed germination over control (%) | Seedling vigour Index (SVI) | Increase in SVI over control (%) |
|---|---|---|---|---|---|
| 1 | Captan @ 0.2 % | 84 (66.50) | 17.85 | 1503.6 | 22.44 |
| 2 | Carbendazim @ 0.2 % | 87 (68.91) | 20.68 | 1592.1 | 26.75 |
| 3 | Mancozeb @ 0.2 % | 83 (65.66) | 16.86 | 1477.4 | 21.07 |
| 4 | Carboxin (37.5) + Thiram (37.5) 0.2 % | 85 (67.32) | 18.82 | 1547.0 | 24.62 |
| 5 | Propineb @ 0.2 % | 83 (65.68) | 16.86 | 1477.4 | 21.07 |
| 6 | *T. viride* @ 0.6 % | 78 (62.07) | 11.53 | 1380.6 | 15.5 |
| 7 | *P. fluorescens* @ 0.6 % | 76 (60.68) | 9.21 | 1320.0 | 11.65 |
| 8 | Ginger extract @ 0.5 % | 73 (58.70) | 5.47 | 1248.3 | 6.58 |
| 9 | Garlic extract @ 0.5 % | 75 (60.05) | 8.00 | 1297.5 | 10.12 |
| 10 | Control | 69 (56.17) | | 1166.1 | |
| | S. E. + | 0.95 | | 17.96 | |
| | CD at 5 % | 2.75 | | 51.87 | |
| | C.V. (%) | 3.01 | | 2.56 | |

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