

EFFECT OF VERMIWASH AND GIBBERELLIC ACID ON SEED GERMINATION IN FENUGREEK (TRIGONELLA FOENUM-GRAECUM L.).

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Abstract

In the present study, experiments were carried out to study the influence of Vermiwash (20%) and Gibberellic acid (50 ppm) on fenugreek (Trigonella foenum-graecum L.) var. early bunching during seed germination by using Petri-plate method. Distilled water was considered as control. Both the treatments stimulated the germination percentage, vigour index and also showed enhancement in fresh and dry weights of root and shoot. GA treatment remarkably enhanced shoot length while vermiwash treatment showed upper hand in root length. Vermiwash and GA treatments significantly increased amount of total carbohydrates, soluble proteins and activities of enzymes like amylase, catalase and protease. The study revealed that vermiwash and GA exhibit superiority in germination and growth process and significantly improve phyto-chemical values.

Keywords: Vermiwash, Gibberellic acid, Fenugreek, Vigour index.

INTRODUCTION

Organic farming is productive systems which rely in use of organic fertilizers, crop rotation, biocontrol and biofertlizers while strongly deny synthetic agrochemicals, plant growth regulators and genetically modified organisms. It works in harmony with nature and to enhance the health of agricultural soil for sustainable agriculture play a crucial role. Organic fertility inputs, such as composted farm yard and green manure, improve the soil physical properties by lowering bulk density, increasing water-holding capacity and improving infiltration rates (Tester 1999; Werner 1997). Organic farming systems are more sustainable than conventional agricultural techniques and represent commercially viable alternative (Marangoni et al 2004). Organic substance like oxygenated peptone can be successfully used for soil conditioning under organic farming (Patil et al 2008; Thakare et al 2011; Jagtap 2012).

The casts of earthworm is one of the most useful and active agents in introducing changes in the soil there by, directly increasing the fertility and crop producing power in the soil (Joshi & Kelkar 1951). Earthworms play variety of important roles in agro-ecosystems. Their feeding and burrowing activities incorporate organic residues and amendments into soil, enhancing decomposition, humus formation, nutrient cycling and soil structural development (Kladivko & Mackay 1985; Kladivko et al 1986). Vermicompost and and vermiwash improve physico-chemical properties of soil (Tharmaraj et al 2010). Vermiwash is a liquid manure obtained from earthworms while vermicompost preparation. It contains phytohormones like auxins and cytokinins. According to Zambare et al (2008), Vermiwash contain nitrogen fixing bacteria such as Azotobacter sp., Agrobacterium sp., and Rhizobium sp. and also enzyme cocktail of proteases, amylases, urease and phosphatase.

Fenugreek (Trigonella foenum-graecum L.) is a member

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of papilionaceae, cultivated as leafy vegetable and forage crop across the Asian and the African continents. It was introduced to Europe and America. The plant has medicinal values also. It is extensively cultivated in most regions of the world for its medicinal value. Fenugreek seeds are good source of protein, fat, minerals and dietary fiber (Kochhar et al 2006). The notable chemical constituents of fenugreek are proteins that are rich in lysine and tryptophan, flavonoids such as quercetin, trigonelline, saponin, phytic acid and polyphenols (Billaud et al 2001). Seeds are used in colic, flatulence, dysentery, diarrhea, dyspepsia with loss of appetite, diarrhea in puerperal women, chronic cough, dropsy, and enlargement of the liver and spleen (Nadkarni 2005).

These days, vermicompost and vermiwash is widely used in organic farming. Since fenugreek has high medicinal and nutritionally important, the plant was selected for the work. Seed germination is the first and very important event in the life of plants, hence in the present investigation attempts were made to study the effect Vermiwash and Gibberellic acid on fenugreek during germination of seeds.

MATERIAL AND METHOD

The earthworm species used for vermiwash preparation was *Eudrilus eugineae* (Fig.1). Large plastic container with a tap attached at the bottom for vermiwash draining was used. A base layer of gravel was covered by a layer of coarse sand. Partially composted cow dung with leaf litter was placed on the top of sand and afterwards, earthworms were introduced. Moisture level was maintained by sprinkling water once in 2 or 3 days. Vermiwash was collected after 10 day.

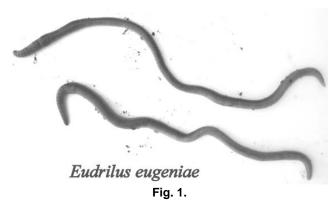
Seeds of fenugreek (*Trigonella foenum-graecum* L. var. early bunching) were surface sterilized with 0.05 % HgCl₂ for minute and rinsed thoroughly with tap water and then with distilled water. The surface sterilized seeds were arranged in petri plates (10 seeds/plate) containing germination paper. 10 ml distilled water (control), 50 ppm gibberellic acid 20% vermiwash was poured in the respective petri-dishes. Numbers of seeds germinated were recorded daily.

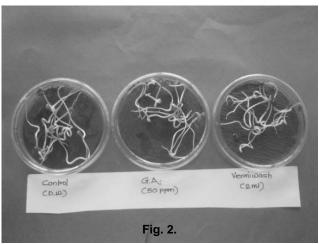
Germination parameters like germination percentage, shoot length, root length, shoot/root ratio, fresh weight and dry weight were studied on 6 DAS using routine method. Vigour Index (VI) was calculated according to the method suggested by Abdul and Anderson (1993). Biochemical constituents were analyzed on 8 DAS using methods proposed by Lowery *et. al.* (1951) for soluble proteins and Sadasivam and Manickam (2005) for total carbohydrates. The enzyme activity was studied on 10 DAS. The enzyme activity of Amylase (E.C. 3.2.1.1) and Catalase (E.C. 1.11.1.6) was scored by the methods of Sadasivam and Manickam and that of Protease (E.C. 3.4.2.2) by the method of Penner and Ashton (1967).

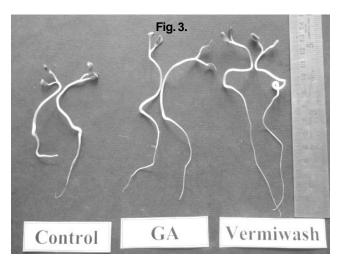
RESULTS AND DISCUSSION

The experiments revealed that GA and Vermiwash treatments improve germination performance of fenugreek (*Trigonella foenum-graecum* L. var. early bunching). Table 1 exhibit the effect GA and vermiwash on morphological parameters of germinating seeds of fenugreek 6 days after sowing (DAS). Seeds showed 100 % germination under both GA and vermiwash treatments while 98 % under control (distilled water). Shlrene et al (2012) reported that vermiwash contain higher amount of potassium (K) than sodium (Na). Potassium, as one of the essential macro-nutrients, is needed in higher amount for better plant growth (Bumb & Hammond 2005). Riley (1987) stated that GA plays direct role in stem elongation. Shoot elongation was greatly enhanced by GA (21.39%) than that by vermiwash (12.52%) (Fig. 2 and Fig. 3.), while vermiwash (53.54%) showed dramatic root elongation than control and GA (9.16%). Due to elongation of shoot, shoot/root ration was positively improved by GA (1.33), on the contrary vermiwash (0.87) exhibited less than GA and control (1.19). Both treatments also showed positive effect on fresh as well as dry weights of shoot and root. GA showed superiority in fresh (21.47) and dry (45.58%) of shoot than vermiwash (15.50% and 12.50% respectively). Vermiwash exhibited upper hand in fresh weight (59.63%) and dry weight (50%) of roots than GA (31.65% and 25% respectively). Vigour Index (VI) is generated by using values of root length, shoot length and germination percentage. There was significant increase in VI values under vermiwash (1384) than that of GA (1222) and control (1033.9).



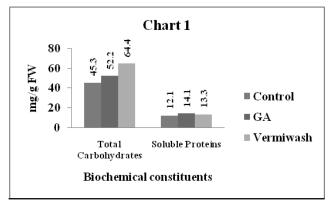






Biochemical constituents were analyzed at 8 DAS and enzyme activity 10 DAS. Table 2. exhibit effect of GA and vermiwash on seeds of fenugreek (*Trigonella foenum-graecum* L. var. early bunching) on biochemical constitu

ents and enzyme activity. Graph 1. exhibit effect of treatments on biochemical constituents. Both the treatments showed significant increase in total carbohydrates and soluble proteins content. Ascending order for total carbohydrates was control (45.3 mg/gram fresh tissue) < GA (52.2 mg/gram fresh tissue) < vermiwash (64.4 mg/gram fresh tissue), while for soluble proteins, it was control (12.152 mg/gram fresh tissue) < vermiwash (13.368 mg/gram fresh tissue) < GA (14.143 mg/gram fresh tissue).



Experiments also revealed significant increase in enzyme activities. Variety of environmental stresses like high/low temperature, water stress, air pollution, UVlight and chemicals results in the excess production of active oxygen species such as super oxide, hydrogen peroxide and hydroxyl radicals. Superoxide dismutase (SOD) and catalase (CAT) plays a vital role in scavenging reactive oxygen species. Sangeetha (2010), in fenugreek, reported SOD & CAT increases significantly under the influence of carbendazim. The catalase activity was significantly high under vermiwash treatment (7.048) mole $H_2O_2/min/g$ FW) than that of control (2.296) i mole $H_2O_2/\min/g$ FW) and GA (5.152) i mole H_2O_2/m min/g FW). For amylase activity vermiwash (1.06 mg maltose/5 min./g fresh weight) treatment showed supremacy over GA (0.86 mg maltose/5 min./g fresh weight) and control (0.68 mg maltose/5 min./g fresh weight). GA (164.358%) showed upper hand for protease activity over vermiwash (102.742%) and control. According to Muhyaddin and the enzymes are activated with an accompanying mobilization of reserve material ending in transport of reserve material in the embryo in somatic conditioning and thus stronger seedlings are obtained as a result of embryo growth.

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Table 1. Effect of GA and Vermiwash on seeds of Fenugreek (*Trigonella foenum-graecum* L. var. early bunching) on morphological parameters of seedlings (6 DAS).

| Parameters | Control | Treatments | | | | | |
|----------------------|--------------------|--------------------|--------------|-------------------|--------------|--|--|
| | Distilled water | Gibberellic acid | Increase (%) | Vermiwash | Increase (%) | | |
| Germination | 98 | 100 | 2. | 100 | 2 | | |
| percentage | 70 | 100 | 2 | 100 | 2 | | |
| Shoot length (cm) | 5.75 ± 0.31 | $6.98*\pm0.15$ | 21.39 | $6.47*\pm0.54$ | 12.52 | | |
| Root length (cm) | 4.80 ± 0.54 | $5.24*\pm0.62$ | 9.93 | $7.37** \pm 1.55$ | 53.54 | | |
| Shoot/Root Ratio | 1.19 ± 0.17 | $1.33*\pm0.15$ | 11.76 | $0.87* \pm 0.01$ | -26.89 | | |
| Shoot: Fresh wt. (g) | 1.122 ± 0.075 | $1.384*\pm0.044$ | 21.47 | $1.296*\pm0.006$ | 15.50 | | |
| Shoot: Dry wt. (g) | 0.136 ± 0.006 | $0.198* \pm 0.007$ | 45.58 | $0.153**\pm0.003$ | 34.48 | | |
| Root: Fresh wt. (g) | 0.218 ± 0.006 | $0.287*\pm0.005$ | 31.65 | $0.348**\pm0.015$ | 59.63 | | |
| Root: Dry wt. (g) | 0.016 ± 0.006 | $0.020*\pm0.002$ | 25 | $0.024*\pm0.002$ | 50 | | |
| Vigour Index (VI) | 1033.9 ± 27.07 | $1222*\pm 35.01$ | 18.19 | $1384*\pm 20.65$ | 33.86 | | |

Table 2. Effect of GA and Vermiwash on seeds of Fenugreek (*Trigonella foenum-graecum* L. var. early bunching) on biochemical constituents (8 DAS) and enzyme activity (10 DAS).

| Parameters | Control | Treatments | | | | | | | |
|---|-------------------|------------------|--------------|--------------------|--------------|--|--|--|--|
| | Distilled water | Gibberellic acid | Increase (%) | Vermiwash | Increase (%) | | | | |
| Biochemical constituents | | | | | | | | | |
| Total carbohydrates (mg/g FW) | 45.3 ± 6.90 | 52.2*± 2.85 | 15.23 | 64.4**± 3.85 | 42.163 | | | | |
| Soluble Proteins (mg/g FW) | 12.152 ± 0.21 | 14.143*± 0.25 | 16.38 | 13.368*± 0.20 | 10.00 | | | | |
| Enzyme activity | | | | | | | | | |
| Amylase (mg maltose/5min/g FW) | 0.68 ± 0.012 | 0.86*± 0.008 | 26.471 | 1.06*± 0.016 | 55.882 | | | | |
| Catalase (µ mole H ₂ O ₂ /min/g FW) Protease | 2.296± 0.021 | 5.152*± 0.022 | 124.39 | $7.048* \pm 0.011$ | 206.969 | | | | |
| (μg tyrosine/hr/mg protein) | 0.069 ± 0.001 | 0.183*± 0.002 | 164.35 | 0.140*± 0.004 | 102.74 | | | | |

^{*}Values are the mean of triplicates, *Significance at 5% level ——(*), *Significance at 1 % level ——(**)

CONCLUSION

The present investigation showed positive effect of vermiwash & GA on the seedgermination. Various morphological parameters such as root length, shoot length as well as biochemical constituents dramatically improved by the treatments during seed germination. Although GA improve performance of seed germination, but being a hormone, enter the metabolic pathways and alters them. Vermiwash contain beneficial microbes es-

sential nutrients, improve germination performance and amend soil, hence it is better choice for organic farming.

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