



EVALUATION AND DETERMINATION OF THE BEST TIME OF PRIMING AND PRIMING SOLUTION LEVELS FOR GERMINATION INDEXES OF *Trigonella foenum graecum*

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ABSTRACT

Optimal germination and plant establishment is an important problem for agricultural productivity in arid and semi-arid areas. Priming is an approach for increasing plant establishment in undesirable conditions. An experiment as factorial in RCBD with three replications was conducted during 2011 at Research Laboratory of Faculty of Agriculture, Lahijan University in Iran. Factors were time of priming ($T_1 = 12$ and $T_2 = 24$ hours) and priming solution levels ($S_1 =$ water, $S_2 = 250$ mg/l humic acid, $T_3 = 500$ mg/l humic acid, $T_4 = 250$ mg/l vermiwash, $T_5 = 500$ mg/l vermiwash). The results showed that time of priming only on plumule dry weight had a significant difference in 5% probability level. The results showed that priming solution levels treatment significantly affected all of germination indexes except radicle dry weight to plumule dry weight ratio.

Keywords: *Trigonella foenum graecum*, germination indexes, priming time, priming solution levels.

INTRODUCTION

Trigonella foenum graecum wild or cultivated is widely distributed throughout the world and belongs to the Fabaceae family. It is an old medicinal plant and has been commonly used as a traditional food and medicine (Mehrafarin *et al.*, 2010). Seed priming has been successfully demonstrated to improve germination and emergence in seeds of many crops, particularly seeds of vegetables and small seeded grasses (Bradford, 1986; Heydecker and Coolbaer, 1977). Humic substances (humic and fulvic acids) constitute 65-70% of the organic matter in soils, and are the subject of study in various areas of agriculture, such as soil chemistry, fertility, and plant physiology as well as environmental sciences, because the multiple roles played by these materials can greatly benefit plant growth (Knicker *et al.*, 1993; Friedel and Scheller, 2002). Humic acids (HA) are a major component of organic fertilizers, and as heterogeneous molecules of different sizes that are self-organized in supramolecular conformations (Piccolo *et al.*, 2002; Piccolo, 2002), they are also the most reactive ones. HA effects on plant physiology are mainly positive, and they include enhancement of biomass yields (Ayuso *et al.*, 1996; Arancon *et al.*, 2006), induction of lateral roots emergence and ATPase activity (Canellas *et al.*, 2002), increase of cell respiration and membrane uptake of nutrients, and exertion of hormone-like activities (Nardi, 2002). Given the HA structural complexity, a structure-activity approach aimed at linking effects on plant physiology with specific humic chemical properties is a difficult task. One way to partly reduce the HA heterogeneity is to carry out a size-fractionation of humic matter and characterize the separated size-fractions by combined pyrolysis and NMR spectroscopy (Piccolo *et al.*, 2002; Piccolo, 2002). The characteristics of such more homogenous humic molecules may be related to well define soil-plant process of ecological importance. Humic acids are considered to be

compounds increasing permeability of cellular membranes in plants (Kaya, 2005), and recent studies prove that these substances significantly affect an increase in seed germination energy, the intensification of seedling growth, the growth in root weight and shoot development (Katkat, 2009). Vermiculture is a mixed culture containing soil bacteria mixed and an effective strain of earth worms (Niir, 2008). Earthworm has efficiency to consume all types of organic rich waste material including vegetable waste, industrial and other organic waste. Vermicomposting refers to the production of plant nutrient rich excreta of worms. Earthworms play a vital role in plant growth. It is a quite possible to effect quick change over for sustainable agriculture by harnessing brand new vermicompost technology to the soil. In recent times, the commercial vermin culturists have started promoting a product called vermiwash. This vermiwash would have enzymes, secretions of earthworms which would stimulate the growth and yield of crops and even develop resistance in crops receiving this spray. Such a preparation would certainly have the soluble plant nutrients apart from some organic acids and mucus of earthworms and microbes (Shivsubramanian and Ganeshkumar, 2004). But so far there are no experimental evidences to quantify the effect of such spray. vermiwash is considered to be compounds increasing permeability of cellular membranes in plants, and recent studies prove that these substances significantly affect an increase in seed germination energy, the intensification of seedling growth, the growth in root weight and shoot development (Shivsubramanian and Ganeshkumar, 2004; Zambare, 2008). The aims of the study are Evaluation and Determination of the Best Hydro and Osmopriming Treatments for Germination indexes of *Trigonella foenum graecum*.



MATERIALS AND METHODS

The experiment was carried out at the Seed Research Laboratory of Faculty of Agriculture, University of Lahijan, Iran in the year 2011. The experiment was a factorial with two factors on the basis of a completely randomized design with four replicates. *Trigonella foenum gracum* seeds were primed with water, humic acid, fulvic acid and verminvash solutions (S_1 = water, S_2 = 250 mg/l humic acid, T_3 = 500 mg/l humic acid, T_4 = 250 mg/l vermiwash, T_5 = 500 mg/l vermiwash) for T_1 = 12 and T_2 = 24 hours, at 20°C. After priming, seeds were put in a wire mesh strainer and washed with tap water for 5 minutes and then rinsed with distilled water. Following this, seeds were dried between two filter papers. Primed and non-primed seeds were placed in 9cm glass petri dishes on a layer of filter paper (Whatman # 41). Fifty seeds were placed in each petri dish. Seeds were allowed to germinate $25 \pm 1^\circ\text{C}$ under dark condition for 7 days. Germination was considered to have occurred when radicles were 2mm long. Germination percentage was recorded every 24 hours for 7 days. Radicle dry weight, plumule dry weight, radicle dry weight to plumule dry weight ratio, radicle length, plumule length, radicle length to plumule length ratio and seedling length were measured to day 14 after the start of the experiment. Speed germination, Mean Germination Time, Percent germination and Seedling Vigor Index were calculated according to the following bottom equations.

$$\text{Speed Germination (SG)} = \Sigma \frac{Ni}{Di}$$

$$\text{Mean Germination Time (MGT)} = \frac{\Sigma (Ni Di)}{\Sigma N}$$

$$\text{Percent germination} = \Sigma \frac{Ni}{N}$$

Where

$$\begin{aligned} \text{Seedling Vigor Index (SVI)} &= \text{Vigor Index} \times \\ &\quad (\text{radicle length average} + \\ &\quad \text{plumule length average}) \\ \text{Vigor index} &= \% \text{ germination} \times \text{seedling} \\ &\quad \text{length} \\ Ni &= \text{number of seeds germinated on} \\ &\quad \text{the day } i \\ Di &= \text{days of germination test} \\ N &= \text{total number of seeds} \end{aligned}$$

Data analyses were carried out using SAS.

RESULTS AND DISCUSSIONS

The process of seed germination of *Trigonella foenum gracum* show that maximum of seed germination in all treatment obtained in 5 days after priming and then decreases (Figure-1). Results of variation analysis show that the effect time of priming only on plumule dry weight had a significant difference in 5% probability level and traits radicle dry weight, radicle dry weight to plumule dry weight ratio, radicle length, plumule length, radicle length to plumule length ratio, speed germination, Mean Germination Time, percent germination, Seedling Vigor Index and seedling length were not significantly influenced by time of priming (Tables 1 and 2). Comparison of Mean between studied traits show that the highest plumule dry weight was obtained of 12 hours with 0.245 g (Table-3). Results of variation analysis show that the effect of priming solution levels on traits radicle dry weight, plumule dry weight, radicle length, plumule length, radicle length to plumule length ratio, speed germination, Mean Germination Time, percent germination, Seedling Vigor Index and seedling length was significantly and radicle dry weight to plumule dry weight ratio was not significantly influenced by seed priming treatment (Tables 1 and 2). Comparison of Mean show that the highest traits radicle dry weight (0.165 g), plumule dry weight (0.301 g), radicle length (0.515 cm), plumule length (1.058 cm), speed germination (11.5 number/day) Seedling Vigor Index (43.32) and seedling length (1.572 cm) were obtained of treatment with 250 mg/l vermiwash concentration. Comparison of Mean show that the lowest radicle length to plumule length ratio was obtained of treatment with 500 mg/l humic acid concentration (0.415 cm) (Tables 3 and 4). Comparison of Mean between priming solution levels show that the lowest Mean Germination Time was obtained of treatment with 250 mg/l vermiwash concentration (4.89 day) (Tables 1 and 2). Comparison of Mean between priming Solution levels show that the lowest percent germination were obtained of treatment with 500 mg/l humic acid (91.33%) and 250 mg/l vermiwash (93.33%) concentration (Tables 1 and 2). With attention to variance analysis table the interaction effects time of priming and seed priming treatment on traits plumule dry weight, radicle length, plumule length, percent germination and seedling length had a significant difference in 1% probability level (Tables 1 and 2). The mean comparison of interaction effects show that (Table-3) the highest amounts of plumule dry weight (0.36 g), radicle length (0.61 cm), plumule length (1.21 cm) and percent germination (100%) were found from 12 hours and 250 mg/l vermiwash concentration (Table-5). The mean comparison of interaction effects show that the lowest amount of seedling length (0.77 cm) was found from 24 hours priming with water (Table-5). Similar results were reported by Makai, (1996) and Farahani and Maroufi, (2011).

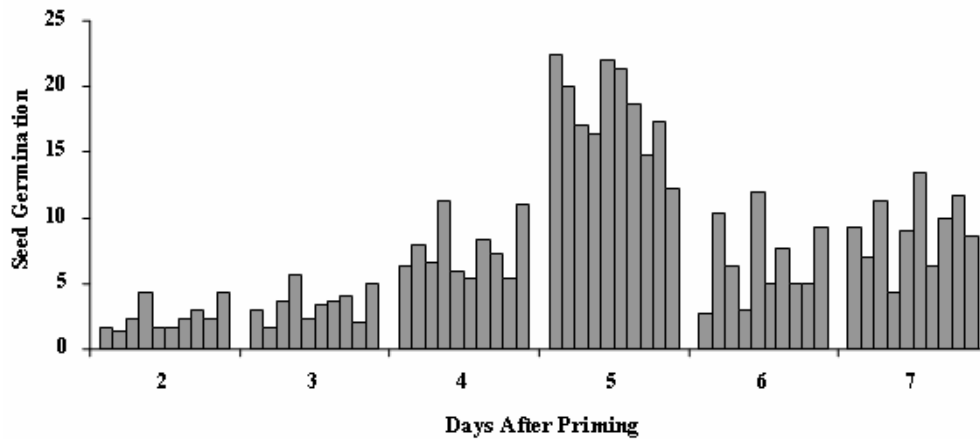


Figure-1. The effect of time of priming and priming solution levels of process seed germination.

Table-1. Analysis of variance of the traits growth in seedling *Trigonella foenum gracum*.

Sours of variance	df	Radicle dry weight	Plumule dry weight	Radicle dry weight / radicle dry weight	Radicle length	Plumule length	Radicle length / radicle length	Seedling length
Replication	3	0.003	0.0005	0.092	0.009	0.023	0.067	0.006
Time of priming (T)	1	0.0003	0.01*	0.06	0.00009	0.003	0.003	0.004
priming solution levels (S)	4	0.015**	0.017**	0.139	0.051**	0.304**	0.062*	0.5556**
T×S	4	0.001	0.008**	0.097	0.031**	0.091**	0.027	0.220**
Error	27	0.002	0.002	0.066	0.004	0.016	0.019	0.021
C.V %		61	21	63	16	16	26	12

** and * respectively significant in 1% and 5% area; ns: none significant

Table-2. Analysis of variance of the germination indexes in *Trigonella foenum gracum*.

Sours of variance	df	Speed germination	Mean germination time	Percent germination	Seedling vigor index
Replication	3	0.425	0.034	8.888	11.752
time of priming (T)	1	0.815	0.034	11.374	49.328
solution levels (S)	4	3.599**	0.36**	70.4**	504.16**
T×S	4	0.245	0.042	89.6**	41.201
Error	27	0.733	0.028	14.91	45.25
C.V %		8	3	4	22

** and * respectively significant in 1% and 5% area; ns: none significant

**Table-3.** Comparison of means of the traits growth in seedling *Trigonella foenum gracum*.

Treatment	Radicle dry weight (g)	Plumule dry weight (g)	Radicle dry weight / radicle dry weight (g)	Radicle length (cm)	Plumule length (cm)	Radicle length / radicle length (cm)	Seedling length (cm)
Time of priming							
12 hours	0.084A	0.245A	0.366A	0.384A	0.763A	0.521A	1.148A
24 hours	0.09A	0.212B	0.444A	0.387A	0.781A	0.54A	1.168A
Priming solution levels							
Water	0.06B	0.18B	0.348A	0.315C	0.525C	0.61A	0.84
250 mg/l humic acid	0.07B	0.217BC	0.346A	0.357BC	0.743B	0.516AB	1.102B
500 mg/l humic acid	0.07B	0.203BC	0.392A	0.335C	0.83B	0.415B	1.165B
250 mg/l vermiwash	0.165A	0.301A	0.635A	0.515A	1.058A	0.485AB	1.572A
500 mg/l vermiwash	0.07B	0.242B	0.305A	0.407B	0.702B	0.627A	1.112B

Means followed by the same letter in the same column are not significantly different at the 5% probability level by Duncan test

Table-4. Comparison of means of the traits growth in seedling *Trigonella foenum gracum*.

Treatment	Speed germination (number/day)	Mean germination time (day)	Percent germination (%)	Seedling vigor index (-)
Time of priming				
12 hours	10.36A	5.28A	94.40A	31.18A
24 hours	10.64A	5.22A	95.46A	28.96A
Priming solution levels				
Water	9.97B	5.37B	95.33AB	23.17C
250 mg/l humic acid	10.16B	5.27A	95.33AB	27.43CB
500 mg/l humic acid	9.99B	5.41A	91.33B	25.46BC
250 mg/l vermiwash	11.5A	4.89B	93.33B	43.32A
500 mg/l vermiwash	10.88AB	5.34A	99.33A	30.96B

Means followed by the same letter in the same column are not significantly different at the 5% probability level by Duncan test.

Table 5. The interaction effect of time of priming and priming solution levels for germination indexes of *Trigonella foenum gracum*.

Treatment	Plumule dry weight	Radicle length	Plumule length	Seedling length	Percent germination
T1S1	0.19B	0.33CD	0.57EF	0.91EF	90.67BCD
T1S2	0.19B	0.35CD	0.69DE	1.04CDE	96.67AB
T1S3	0.24B	0.30D	0.71CDE	1.01DE	94.67ABC
T1S4	0.36A	0.61A	1.21A	1.81A	90.00CD
T1S5	0.24B	0.33CD	0.63DEF	0.97DEF	100.00A
T2S1	0.17B	0.30D	0.48F	0.77F	100.00A
T2S2	0.24B	0.36CD	0.80BCD	1.16BCD	94.00ABCD
T2S3	0.17B	0.37CD	0.95B	1.32B	88.00D
T2S4	0.24B	0.42BC	0.91BC	1.33B	96.67AB
T2S5	0.24B	0.48B	0.77BCDE	1.26BC	98.67A

Means followed by the same letter in the same column are not significantly different at the 5% probability level by Duncan test.



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