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Influence of seed inoculation with biological fertilizer on fennel (*Foeniculum Vulgare*) and coriander (*Coriandrum Sativum*) germination

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## Abstract

In order to study effects of seed inoculation with Nitragin as bio-fertilizer at concentrations of 2, 3 and 4 cc (ml) and distilled water as control on seed germination and early growth of fennel, a laboratory experiment was conducted in Islamic Azad University of Tabriz in completely randomized design (CRD) with three replications. Results showed that fennel seeds were inoculated with 4 cc (ml) Nitragin, root length increased by 6.5%, compared with the control. Inoculation of fennel seeds with 4 cc (ml) Nitragin was increased shoot length around 3%, compared with un-primed. Seeds which treated with the same treatment seedling length increased up to 179 mm, and root/shoot ratio increased 3.8%, in comparison to control plots. When Nitragin concentration reduced up to 3 cc (ml), shoot dry weight increased nearly 1.4g. In concentration of 2 cc (ml) Nitragin, root/shoot dry weight ratio increased by 45%. In the highest nitragin concentration GR increased by 43%, compared to control. Also, the lowest GR was occurred in seeds treated with distilled water. It was concluded that fennel producers could improve germination and early growth of fennel by seed priming with nitragin.

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#### Introduction

Fennel (Foeniculum vulgare Mill.; Fam. Umbelliferae) is one of the most important medicinal and aromatic plants due to its estrogenic activities and uses as a carminative, diuretic, antiinflammatory, antimicrobial, and galactogogue. It is a substance which is used to increase the production of milk in humans and other animals. In addition, the volatile oils of fennel are used to control flatulent dyspepsia and colic in children (Mahfouz and Sharaf-Eldin, 2007). Using bio-fertilizers that contain different microbial strains has led to a decrease in the use of chemical fertilizers and has provided high quality products free of harmful agrochemicals for human safety (Mahfouz and Sharaf-Eldin, 2007). One of the methods that reduce fertilizer usage is seed preplanting treatments called priming that include water absorption at enough level to begin germination events that is accomplished by the subsequent drying. The purpose of priming is increasing germination percent, decreasing mean of germination time and improving growth and vigor of seedling at very wide favor and un-favored environmental conditions. This method is successful in small seed plants and the most medicinal plants that have great economic value with quick and uniform emergence requirement (Ellis and Roberts, 1981). These techniques include hydro priming and hormonal priming or soaking prior to sowing (Basra et al., 2005; Ashraf and Foolad, 2005). Seed priming techniques are used to reduces emergence time, accomplish uniform emergence, better allometric (changes in growth of plant parts over time in many horticultural and field crops (Ashraf and Foolad, 2005; Farooq et al., 2005). Badran and Safwat (2004) and El-Ghadban et al. (2006) found that fennel responded to bio-fertilizer by increasing growth and oil yield and changing the chemical composition. Yousry et al. (2003) indicated that inoculation of pea (Pisum sativum) plants with Bacillus megatherium increased plant dry matter by 10.9%, while the combined application of *B*. megatherium and P-fertilizer increased 19.7% of dry matter by Amin (1997), on coriander (Coriandrum sativum), fennel (Feoniculum vulgare), and caraway (Carum carvi), showed that the growth was influenced by seed inoculation (Azotobacter and Azospirillum) with a half dose of inorganic fertilizer. In this condition plant growth was equal to full dose of inorganic fertilizer. Tehlan et al. (2004) reported that plant growth and seed yield of fennel varied according to the Azotobacter applied. These results are in agreement with research results of Gad (2001) for fennel (Foeniculum vulgare) and dill (Anethum graveolens), who reported that biofertilizers on these plants increased growth and yield. Viable seed is capable of producing new plant under both favorable and unfavorable climatic conditions. High number of seed emergence cause vigorous seedling and better stand establishment (Baleševi-Tubi and colleagues., 2007). Excessive use of fertilizers in order to achieve higher yields and food shortages can increase production costs and degradation of water resources and soil healthy and sustainable farming systems in terms of ecological interest is to specialists (Tilak et al., 1992). Kennedy and Tychan (1997) indicated that germination of corn seed accelerated by inoculated with Azotobacter. Inoculation of plants with Azospirillum was caused a significant changes in various growth parameters, such as increase in plant biomass, nutrient uptake, tissue N content, plant height, leaf size and root length of cereals (Bashan et al., 2004). Thus it has been shown that Azospirillum and Pseudomonas had the potential for agricultural exploitation and could use as natural fertilizers (Bashan et al., 1989; Cakmakc et al., 2006). In recent years, a lot of work has been done on the invigoration of seeds to improve the germination rate and uniformity of growth and reduce the emergence time of many vegetables and some field crops (Basra et al., 2003; Tehlan et al., 2004). The aim of the present study was to evaluate the effects of seed inoculation with biological fertilizer (Nitragin) on germination and early growth of fennel (Foeniculum vulgare) and study effects of Nitragin in agriculture in order to achieve the goals of sustainable agriculture.

#### Material and methods

#### Laboratory experiment

The laboratory experiment was done by complete randomized design (CRD) with three replications was

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conducted in Islamic Azad University of Tabriz, Iran. This study was assessment of seed inoculation with Nitragin as biological fertilizer in concentrations of 2, 3 and 4 cc (ml) and distilled water as control on seed germination and early growth of fennel (Foeniculum vulgare). The seeds were surface sterilized with 5% NaOCL (sodium hypo chloride) for 5 min to avoid fungal invasion, followed by washing with distilled water. The seeds were primed by solutions of Nitragin which involves free-living and associative bacteria such as Azospirillum, Azotobacter and Pseudomonas. Germination tests were conducted by using 3 replicates of 50 seeds in petri dishes (dimention of petri dishes are 9 cm). There are 10<sup>8</sup> viable bacterial cells on 1 ml fluid of Nitragin (Azotobactin) active water-soluble (Okon, 2002). Petri dishes and filter paper after disinfection with sodium hypo chloride, and also in order to ensure the absence of any contamination were in UV radiation for 24 hours under a sterile electric hood. For each treatment 50 seeds with three replications incubated in growth chamber at 28°C. After 7 days the number of germinated seeds was counted. In each petri dishes, 50 fennel healthy seeds was considered. In all treatments, seeds were disinfected with a solution of starch and sugar-coated, then inoculated with Nitragin treatments. Petri dishes in plastic bags were transferred inside the container grown 25±1 °C. In order to calculate the germination rate, Petri dishes containers from the second day of testing each day until the tenth day of out-grown and the number of germinated seeds were counted. In daily counting if

 Table 1. Analysis Variance of traits in fennel.

necessary, distilled water was added to the wet filter paper. Traits measured including, root length, shoot length, seedling length, root to shoot ratio, root dry weight, shoot dry weight, Seedling dry weight, root to shoot ratio of dry weight, germination percentage and Crop growth rate.

The germination rate (GR) was calculated by using the equation below (Maguirw, 1962).

 $GR = \Sigma n / N \times 100$ 

Where:  $\Sigma n$  - total number of germinated seeds at each counting, *N* - number of total seeds.

Finally, after the fourteenth day, 10 samples randomly selected from each Petri and measuring root length, shoot length and seedling length, seedling dry weight determination. Then, samples placed in the oven for 24 hours at  $75^{\circ}$  C and weighed on a milligram.

#### Statistical analyisis

The statistical data were analyzed using MSTAT-C software. The means of the treatments was compared using the least significant difference test at P < 0.05 by LSD method.

### **Result and discussion**

#### Coriander (Coriandrum sativum)

The germination test was performed according to previously bio-fertilizer inoculation on coriander. Nitragin concentrations on germination was not showed reaction.

S.O.V	df	Root	Shoot	length	Lr/ls	Root	Shoot	Seedling	Wr/ws	Gp	Gr
		length	length	Seedling		weight	weight	weight			
		(lr)	(ls)			(wr)	(ws)				
Treatment	3	152.5**	47.14*	363.93**	0.004*	0.06**	0.05ns	0.051 ns	0.058 **	491*	8.86**
Error	8	5.22	7.57	23.21	0.001	0.005	0.048	0.066	0.003	70	0.63
C.V (%)		2.44	3.73	2.88	2.29	14.12	17.38	14.63	13.12	10.52	11.54

### Fennel (Foeniculum vulgare)

### Radicle length

Analysis of variance for data showed that bio-priming of seed with Nitragin as biological fertilizer at concentrations of 2, 3 and 4 ml and distilled water as control had significantly effect (at 5% level) on crop root length of fennel (Table 1). Results revealed that in those fennel seeds were inoculated with 4 ml Nitragin, root length increased by 6.5%, compared with the control (Table 2). The highest (101 mm) and lowest (84 mm) of root length was obtained at biological fertilization concentration of 4 and 2 ml, respectively. However the amount of the increase in seed inoculation with Nitragin as biological fertilizer at concentrations of 2, 3 ml, on root length was 94 and 84 mm, respectively. For maximum root length 4 ml Nitragin recommended. Moreover, that the root length and number subsidiary divisions that will increase and this enhances the absorption and root uptake of water and nutrients and ultimately increasing the performance (Saleh rastin., 2001).

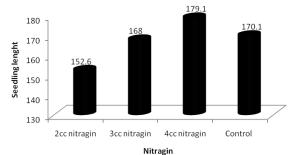
	Root	Shoot	length	Lr/ls	Radicle	Plumule	Ratio of Radicle dry	Germination%	Growth rate
	length	length	Seedling		dry	dry weight	weight to Plumule		(g.m <sup>-2</sup> . day <sup>-1</sup> )
	(mm)	(mm)			weight	(g)	dry weight		
					(g)				
2cc nitragin	84.10	68.48	152.6	1.22	0.700	1.20	0.58	85	6.007
3cc nitragin	94.80	73.20	168	1.29	0.370	1.40	0.27	62.33	7.470
4cc nitragin	101.3	77.73	179.1	1.30	0.483	1.10	0.43	78.33	8.973
Control	94.63	75.50	170.1	1.25	0.423	1.35	0.31	92.33	5.043

Table 2. Comparison of means on fertilizer nitragin on biological traits of fennel.

Means with similar letter were not significant at the 5% probability level.

#### Plumule length

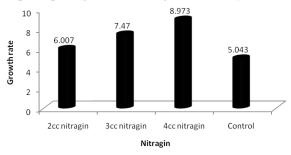
Effect of seed priming with Nitragin on shoot length was significant at 1% level of probability (Table 1). Inoculation of fennel seeds before planting with 4 cc Nitragin gave rise in shoot length around 3%, compared with un-primed ones (Table 2). Effect of seed inoculation with Nitragin as biological fertilizer at concentrations of 2, 3 and 4 cc and distilled water as control on shoot length of fennel. If the amount of the increase in inoculation with Nitragin as biological fertilizer at concentrations of 2 and 3 cc plumule length, Nitragin 68 mm, 73 mm respectively. Sorial *et al.*, (1992), have reported that inoculation of tomato with some of the nitrogen fixing bacteria such as *Azotobacter*. Increase in seedling length, stem length and shoot dry weight was crop.



**Fig. 1.** Comparison of biological effects of fertilizer seedling length fennel.

#### Seedling length

In seeds treated with 4 cc Nitragin seedling length increased 5% in comparison to control (179 mm). Effect of seed inoculation with Nitragin as biological fertilizer at concentrations of 2, 3 on seedling length of fennel, 152 mm and 168 mm, respectively (Table 2). Sorial *et al.*, (1992) have reported that inoculation of tomato with some of the nitrogen fixing bacteria such as azotobacter Increase in seedling length, stem length and shoot dry weight was crop. The results with the findings Bhadauria *et al.*, (2000) also is consistent. Impregnated with seed, fertilizer to improve plant growth in biological (EL-Zeiny., 2007).



**Fig. 2.** Comparison of biological effects of fertilizer, growth rate fennel.

### Ratio of radicle length to plumule length

The effectiveness of ratio of radicle length to plumule length of fennel seed inoculation with the biofertilizer Nitagin, expected that root to shoot ratio is also affected by the treatments, Compared to the

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averages (Table 2) indicate that fennel seeds were inoculated with 4 ml of distilled water Nitagin compared with control, The value of this attribute increases by about 3.84%. If the amount of the increase in inoculation with Nitragin as bio-fertilizer at concentrations of 2 and 3 cc ratio of radicle length to plumule length of fennel 1.22 mm and 1.29 mm respectively. Cultures of fennel seeds inoculated with 4 cc Nitagin recommended. Kennedy and Tychan (1997), accelerate seed germination of corn inoculated with Azotobacter have reported. Azotobacter and Azospirillum have a synergistic relationship. Inoculation of plants with Azospirillum could result in significant changes in various growth parameters, such as increase in plant biomass, nutrient uptake, tissue N content, plant height, leaf size and root length of cereals (Bashan et al., 2004).

## Radicle dry weight of fennel

The highest root dry weight of fennel was observed with the treatment biological fertilizer 2 ml (0.70 g). As the results of the treatments compared (Table 2) showed, fennel seeds were inoculated with 2 ml of Nitragin before planting, root dry weight of fennel around 0.70 g gave rise. The increase in the amount of seed treatment with 3 and 4 Nitragin ml 0.37 and 0.48 respectively. The lowest amount of root dry weight of fennel was obtained with the treatment of biological fertilizer 3 ml (0.37 g). The lowest root dry weight of fennel occurred with 3 ml Nitragin 0.37 g.

## Plumule dry weight of fennel

Inoculated fennel seeds with 3 cc Nitragin before planting, gave rise in shoot dry weight of fennel around 1.4 g (Table 2). Impregnated by the seed treatment with 3 ml Nitragin shoot dry weight 1.4 g of the other treatments were and the classes were statistically different. The effect Nitragin 2 ml with 1.2 g, and the effect Nitragin 4 ml with 1.1 g. The highest shoot dry weight of fennel equivalent to 1.4 by 3 ml Nitragin obtained and lowest shoot dry weight of fennel equal to 1.2 g with 2 ml Nitragin priming seeds obtained.

Ratio of radicle dry weight to plumule dry weight

The results showed, in those fennel seeds were inoculated with 2 ml Nitragin, root dry weight to shoot dry weight ratio increased by 45% compared with control. Root dry weight to shoot dry weight ratio is also affected by the treatments, compared to the averages (Table 2) indicate that fennel seeds were inoculated with 2 cc of distilled water Nitragin compared with control, The value of this attribute increases by about 45 percent. If the amount of the increase in inoculation with Nitragin as bio-fertilizer at concentrations of 3 and 4cc on root dry weight to shoot dry weight ratio of fennel 0.27 g and 0.43 g respectively. Cultures of fennel seeds inoculated with 2cc Nitagin recommended.

## Percent of Germination fennel

The highest fennel germination percentage equivalent to 92% by water priming obtained and lowest germination percentage equal to 85% with 2cc Nitragin priming seeds obtained. Treatments were compared (Table 2) showed , fennel seeds were inoculated with water priming, the germination percentage than the 92 percent increase and the 2 and 4cc Nitragin, the germination percentage than the 85 and 62 percent respectively. In a study in India, medicinal plants inoculated seeds Emblica officinalis, With bacteria brasilense A., Increased 11 percent during the shoot and consequently an increase in 16 percent that were of the seedlings (Bhadauria et al., 2000) .The results with the findings Sorial et al. (1992) is also consistent. The effect of treatments on the germination of seeds with azotobacter cumin by Rezai et al., (2005) concluded that the seeds coated with bacteria as compared to 6.25 g per kg of seed germination increases. The germination of the plant cumin epigeal. Relative length of seedlings that seem to improve the use of Nitragin. Early in the green and uniform product under field conditions may play a role.

## Growth rate (Gr)

In the highest Nitragin concentration GR increased by 43%, compared in comparison with control. Also, the lowest GR occurred in seeds treated with distilled water. It was concluded that fennel producers could

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improve germination and early growth of fennel by seed priming with Nitragin. The results of variance analysis showed that bio-priming of different seed priming showed significant difference at 5% level. The results showed, in those fennel seeds were inoculated with 4 cc Nitragin, crop growth rate increased by 43% compared with control. Results of comparing different levels showed that priming had positive role in increasing crop growth rate. If the amount of the increase in seed inoculation with nitragin as biofertilizer at concentrations of 3 and 4 cc on crop growth rate of fennel 7.47 and 8.97 g.m<sup>-2</sup>.day<sup>-1</sup> respectively. The lowest crop growth rate of fennel occurred with distilled water about 5 g.m<sup>-2</sup>.day<sup>-1</sup>. The decrease crop growth rate was 5 g.m<sup>-2</sup>.day<sup>-1</sup> without priming (distilled water as control). Cultures of fennel seeds inoculated with 4 cc Nitagin recommended.

#### Conclusion

The results showed that inoculation with 4 cc Nitragin fennel seeds before planting can, it significantly increased the seedling. Obviously, under these conditions, the seedlings are produced from high vegetative power. This product will eventually increase. Increase the effect of inoculation with the root of this plant Nitragin and the effect of these changes on the length and number of branches and sub-root, fennel seed germination affected by seed treatment with Nitragin rose sharply. The germination of seeds of fennel, it seems that the relative improvement over the use of seedlings in the green timely Nitragin and uniform seed is effective under field conditions.

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