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Dill (*Anethum graveolens* L.) Growth and Yield Responses to Gamma Radiation and Bio-Fertilization

Hussein A. H. Said-Al Ahl[1, *], Atef M. Z. Sarhan[2], Abou Dahab M. Abou Dahab[2], El-Shahat N. Abou-Zeid[1], Mohamed S. Ali[1], Nabila Y. Naguib[1]

[1]Department of Medicinal and Aromatic Plants, National Research Centre, Dokki, Giza, Egypt
[2]Department of Ornamental Horticulture, Faculty of Agriculture, Cairo University, Cairo, Egypt

Abstract

A field experiment was carried out to investigate the effect of gamma radiation doses (0, 2, 4, 8, 16, 32 and 64 k-rad) with or without bio-fertilizers (at 2 kg/fed.) on growth and yield of *Anethum graveolens* L. plants.Adding bio-fertilizer to dill plants significantly increased plant height, number of branches, root length, fresh and dry weights. The treatment of dill seeds pre-sowing to gamma radiation doses (up to 32 k-rad) led to significant increase in plant height, branches number, fresh and dry weights of root, but root length, fresh and dry weights of herb up to 16 k-rad. The highest values of plant height, root length, fresh and dry weights of plant were recorded at the treatment of 2 k-rad combined with bio-fertilizers. While, the treatment of 4 k-rad combined with bio-fertilizer was superior in increasing numbers of branch compared to the other ones.

Keywords

Anethum graveolens L., Bio-Fertilizer, Growth, Yield, Gamma Radiation

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1. Introduction

Dill (*Anethum graveolens* L.) is an annual herb of the family Apiaceae, which is native to south-west Asia or south-east Europe (Bailer et al., 2001). Itis used as a vegetable, a carminative, an aromatic and an antispasmodic (Hornok, 1992; Sharma, 2004). Herbs contain a wide variety of antioxidant photochemical or bioactive molecules that can neutralize the free radicals and thus retard the progress of many chronic diseases associated with oxidative stress and reactive oxygen species (Sun et al., 2002; Liu, 2003). *A. graveolens* has been reported to contain flavonoids, phenolic, and essential oil (Delaquis et al., 2002). The increased consumption of vegetables and herbs containing high levels of photochemical has been recommended to prevent or reduce oxidative stress in the human body (Sun et al., 2002; Liu, 2003;Chu et al., 2002).The intake of natural antioxidants has been associated with reduced risk forcancer, cardiovascular disease, diabetes, and diseases associated with aging (Ani et al., 2006)]. The extract of *A. graveolens* could protects the liver against high-fat-diet-induced oxidative damage in rats (Bahramikia et al., 2009).

Bio-fertilizers are microbial inoculants, used for application to either seeds or soil for increasing soil fertility with objective of increasing the number of such microorganisms and to accelerate certain microbial processes. Free living N_2-fixing microorganisms are widely distributed and found in almost every ecological rich-in soil associated with plants, in aquatic systems and sediments (Knowles, 1978; Subb-Rao, 1981; Alaa El-Din, 1982). Bio-fertilizers are produced and

*Corresponding author
Email address: hussein_saidalahl@yahoo.com (H. A. H. Said-Al Ahl)

practiced in different countries as well as in Egypt under commercial names. Bio-fertilization is generally based on altering the rhizosphere flora, by seed or soil inoculation with certain organisms, capable of inducing beneficial effects on a compatible host. These organisms may affect their host plant by one or more mechanisms such as nitrogen fixation, production of organic acids, enhancing nutrients uptake, synthesis of vitamins, amino acids, auxins and gibberellins which stimulate growth, or increasing the resistance against plant pathogens (Sprenat, 1990).

The effect of ionizing radiation on different parts of plant i.e, (bulb, tuber, stem-cutting, fruits and seeds) were investigated. Irradiated seeds with gamma rays induced biochemical, physiological and cytological changes, the biochemical contents, i.e, enzyme, protein and phytohormone were severely changed by exposing seeds to gamma rays. It is worth to be noticed that gamma irradiation induced either stimulation or inhibition of growth and endogenous hormones. Lower dosage of gamma rays could stimulate growth. There is general agreement that the low dose irradiation of seeds stimulates the germination and increases the mobilization of stored compounds towards the embryo resulting in an enhanced early development and growth (Korosi and Pal, 1989;Rabie et al., 1996).Applications of the use of low dose (< 10 kGy) irradiation in controlling insect infestation, particularly in fresh fruits and vegetables, controlling parasites in pork products, reducing microbial numbers, and in prolonging the shelf life of fruits and vegetables by inhibiting sprouting of tubers and bulbs and by delaying fruit ripening. It is considered that correctly irradiated food presents no toxicological problems and has the same nutritional value. (Bande et al., 1990).

The aim of this study was to investigate the effect of bio-fertilizers, gamma radiation on growth and yield of *Anethum graveolens* L. plants.

2. Materials and Methods

2.1. Plant Material and Experimental Procedure

The experiments of this study were carried out at the Farm Station of National Research Centre, at Shalakan Kalubia Governorate during the two successive seasons of 2000/2001 and 2001/2002. Seeds of dill (*Anethum graveolens* L.) were obtained from Medicinal and Aromatic Research Dep., Ministry of Agriculture, Egypt. The physical and chemical characteristics of the soil were presented in Table (1). The analysis of the soil was conducted using the methods described by Jackson (1967).

Dry seeds of dill were irradiated with gamma rays doses at 2, 4, 8, 16,32 and 64 k-rad before sowing. The source of irradiation is installed at the Middle Eastern Radioisotope Centre for the Arab Countries, Dokki, Giza, Egypt. The bio-fertilizers used in this study were Rhizobacterin (a mixture of Azotobacter and Azospirillum) and Microbein (a mixture of Azotobacter, *Azospirillum, Pseudomonas, Rhizobium* and *Bacillus*). Bio-fertilizers produced by General Organization for Agriculture Equalization Fund (G.O.A.E.F.). The soil was prepared and divided into plots 3x3.5m with five rows. The dill seeds were sown in 15thoctober in the two seasons. The distance between each row was 60cm a part and 20cm between the hills. The seedlings were thinned one month after sowing to leave two plants per hill. The used bio-fertilizers (1kg Rhizobacterin+1kg Microbein/feddan) added as inoculated the seeds and sowing. The experimental design of the three experiments was factorial and planned in a complete randomized block design having three replications and each replicate contained one plot, the plot contained 175 plants.

Table 1. The physical and chemical properties of the experimental soil.

Physical and chemical properties	2000/2001	2001/2002
Sand (%)	48.8	50.8
Silt (%)	28	26
Clay (%)	23.2	23.2
Soil texture	Sandy loam	Sandy loam
pH	8.08	8.22
E.C (m. mohs/cm)	0.68	0.89
Organic matter (%)	2.07	2.18
N (%)	0.11	0.12

2.2. Data Recorded for Growth Characters

Data for growth characters and chemical constituents were estimated for the all treatments during vegetative stage (90 days after sowing) as follows: plant height (cm), root length (cm), number of branches per plant, fresh and dry weights of herb and root g/plant.

2.3. Statical Analysis

The data of the three experiments were statistically analyzed and the differences between the means of the treatments were considered significant when they were more than least significant differences (L.S.D) at 5% level according to Steel and Torrie (1980).

3. Results and Discussion

3.1. Plant Height

The effect of bio-fertilizer on plant height of dill plants is shown in Table (2). Difference between with and without bio-fertilizer treatments was significant in the two seasons. In addition, the radiation dose from 2 to 16 k-rad significantly increased plant height, whereas, 64 k-rad of gamma rays significantly stunted this character in the two seasons, when compared with that of unirradiated plants. Moreover, the combination of bio-fertilizer with gamma radiation (2-32 k-rad) resulted in increases in plant height during both seasons. The tallest plants were observed from the treatment of interaction between bio-fertilizer with 2 k-rad which recorded by (34.03, 31.15 (cm)) in both seasons, respectively. It is clear that 2 k-rad gamma radiation combined with bio-fertilizer was considered the best treatments for plant height due to stimulation of gamma rays on cell division and its elongation (Pitirmovae, 1979). Radiation affected the genetic components of the treated plants which increased the genetic variation and caused greater frequencies of unadaptive (including lethal) genotypes. Thus, radiation can break down the balance in genotype, which has been kept adaptive to the environment, and reduce adaptability function of the plant (Ichikawa, 1981). The adapted (survived) plants were nearly similar in their heights which resulted in narrow ranges, not enlarged variances and low coefficient of variation values, compared with the control. Hence the increase in variability by plant height following certain mutagen treatment, compared with the control, may be attributed to induced genetic variability (Larik, 1975; Morsi et al., 1977 and El-Mahrouk, 2000). Also, plant height was enhanced by bio-fertilizer and gamma radiation as cited by (El-Sherbeny et al.,1992; El-Shafie et al.,1993; Yadavet al.,1998; Youssef and Moussa, 1998; Youssef et al.,2000; Khalil et al.,2001; Rashed, 2002;Hassanein et al.,2003; Al-Humaid, 2004; Kandeel et al., 2004, a and b; Youssef et al.,2004).

3.2. Number of Branches

Data shown in Table (2) resulted that the treatments of radiation from 2 to 64 k-rad caused a significant increases in branch numbers per plant in both seasons in comparison to that of unirradiated plants. Difference between with and without bio-fertilizer treatments was significant in both seasons. Also, the interaction between bio-fertilizer and gamma radiation had significant effect on branch numbers per plant in the two seasons. The highest branch numbers per plant was found by the combination treatment of irradiation at 4 k-rad combined with bio fertilizer in the first season and 8 k-rad + bio-fertilizer in the second season which gave (4.66 and 4.50 branch number/plant, respectively). The increment of branches number/plant as a result of exposing the seeds to gamma radiation before planting. One possibility could the activation of certain morphogenic genes caused by the alteration in the level of growth regulators, (Meins, 1986). Similar results have been reported by (El-Sherbeny et al., 1992;El-Shafie et al.,1993; Hussein et al.,1995; El-Sherbeny et al.,1997; Youssef and Moussa, 1998; Youssef et al.,2000; El-Kashlan, 2001; Hassanein et al.,2003; Al-Qadasi, 2004; Kandeel et al., 2004, a and b).

3.3. Plant Fresh Weight

From the data in Table (3) it may be remarked that, generally, inoculating the plants with bio-fertilizer as well as irradiating with gamma rays at various doses that used singly or collectively tended to a significant effect on the plant fresh weight of dill grown in both seasons in comparison to that of untreated plants. The heaviest fresh weight of dill herb was obtained by combined treatment of radiation at 2 k-rad with inoculation, giving the values of (25.29, 23.99g/plant, respectively). This trend could be attributed to the stimulation effect of lowest gamma dose on physiological and biochemical processes in plants owing to the best activation of enzymatic system, sustaining natural plant hormons, enhancing metabolism of various organic and non-organic components in plant as well as photosynthetic processes reflexing upon the mass growth production of dill plants. In addition, the maximum enhancement of fresh mass production was occurred by combined treatment of least effective gamma dose and efficient bio-fertilizer. This trend may be attributed to the presence of additive or cumulative and/or synergistic action of both factors on plant growth yield. The favorable effects of bio-fertilizers on the growth and productivity of the treated plants may be explained on the basis of the beneficial effects of bacteria on the nutrient availability, vital enzymes, hormonal stimulating effects on plant growth or increasing of photosynthetic activity. Also, bio-fertilizer inoculation playsa organic form of nutrients such as N to mineral N. This led to increase in the uptake of nutrients from the soil by root of plant and hence promots plant growth, leading to an increase in plant yield (Lampkin, 1990).In this connection, Pitirmovae (1979) revealed that the simulative effect of low dosage of gamma rays on growth might be due to the increase of cell length or cell number and size, shifting in metabolism which promoted the stimulating effect of photohormones on biosynthesis of nucleic acid. Grossmann and Craig (1982) reported that the difference in germination and plant morphology of *Pelargonium hortorum*seed exposed to 1, 2 or 3 k-rad of acute gamma irradiation appeared to be related to internode elongation and it is suggested that irradiation may have altered the gibberellin status. On the other hand, it is clear that high irradiation doses used had unfavorable effect on all growth and yield character, and this may be due to the sensitivity of dill seed to high gamma irradiation doses. In this respect, Kaul et al., (1973) found that irradiation of *Datura metel* seeds with relatively higher doses (10-30 k-rad) resulted in shorter plants with less leaves which indicated also morphological abnormalities. However, Roy and Clark (1970) and Ragab and Mohamed (1983) interpreted the reduction of growth due to high doses of irradiation by inhibiting the translocation of photoassimilation or impairing water uptake and translocation from roots. These results fairly agreed with those obtained by (Hussein et al.,1990; El-Sherbeny et al.,1992; El-Shafie et al.,1993; Youssef and Moussa, 1998; Ibrahim, 2000; Youssef et al.,2000; Khalil et al.,2001; Rashed, 2002; Hassanein et al.,2003; Al-Qadasi2004; Hamed, 2004;Kandeel et al., 2004, a and b; Youssef et al.,2004; Harb et al., 2005; Singh et al.,2005).

3.4. Plant Dry Weight

Table 2. Effect of bio-fertilizers, gamma radiation and their interaction treatments on some growth characters of dill plants during the two seasons (2000/2001 and 2001/2002).

Bio-fertilizers / Gamma radiation (k-rad)	Plant height			Number of branches			Herb fresh weight (g)			Herb dry weight (g)		
	Without bio-fert.	Bio-fert.	Mean	Without bio-fert.	Bio-fert.	Mean	Without bio-fert.	Bio-fert.	Mean	Without bio-fert.	Bio-fert.	Mean
First Season												
0	22.51	26.27	24.39	2.60	3.30	2.95	18.50	24.02	21.26	4.32	4.37	4.34
2	32.37	34.03	33.20	4.50	4.60	4.55	20.65	25.29	22.97	4.67	5.29	4.98
4	30.94	33.18	32.06	4.40	4.66	4.53	16.88	21.64	19.26	4.37	4.60	4.48
8	29.16	30.02	29.59	4.60	4.56	4.58	16.61	19.02	17.81	4.33	4.39	4.36
16	25.63	26.80	26.21	4.26	3.90	4.08	16.50	17.96	17.23	4.17	4.31	4.24
32	23.65	24.99	24.32	3.60	3.66	3.63	14.72	15.67	15.19	4.15	4.14	4.14
64	19.96	22.12	21.04	3.33	3.56	3.44	13.57	13.81	13.69	3.30	3.33	3.31
Mean	26.31	28.20		3.89	4.03		16.77	19.63		4.18	4.34	
L.S.D. at 5%	Bio-fertilizers = 0.58 Radiation = 1.09 Interaction = N.S			Bio-fertilizers = 0.08 Radiation = 0.15 Interaction = 0.22			Bio-fertilizers = 0.97 Radiation = 1.82 Interaction = 2.58			Bio-fertilizers = 0.15 Radiation = 0.29 Interaction = N.S		
Second Season												
0	22.49	23.76	23.12	2.73	3.40	3.06	17.81	22.71	20.26	4.14	4.65	4.39
2	31.12	31.15	31.13	4.40	4.43	4.41	20.49	23.99	22.24	4.85	5.15	5.00
4	29.53	29.48	29.50	4.16	4.46	4.31	20.04	20.45	20.24	4.42	4.39	4.40
8	26.51	27.01	26.76	4.30	4.50	4.40	17.64	18.87	18.25	4.03	4.41	4.22
16	25.07	26.65	25.86	4.06	4.40	4.23	16.91	17.71	17.31	3.79	4.12	3.95
32	23.86	25.38	24.62	3.56	4.06	3.81	14.41	15.33	14.87	3.76	3.93	3.84
64	18.92	21.29	20.10	3.26	3.70	3.48	12.56	12.88	12.72	3.35	3.55	3.45
Mean	25.35	26.38		3.78	4.13		17.12	18.84		4.04	4.31	
L.S.D. at 5%	Bio-fertilizers = 0.59 Radiation = 1.10 Interaction = 1.50			Bio-fertilizers = 0.08 Radiation = 0.15 Interaction = 0.22			Bio-fertilizers = 1.08 Radiation = 2.02 Interaction = N.S			Bio-fertilizers = N.S Radiation = 0.53 Interaction = N.S		

The effect of bio-fertilizer on palnt dry weight of dill plants is shown in Table (3). Difference between with and without bio-fertilizer treatments was significant in the two seasons. In addition the radiation dose at 2 k-rad significantly stimulated plant dry weight in the two seasons. The heaviest weight of dry herb was recorded by the inoculating + irradiation treatment at 2 k-rad giving (5.29, 5.15 g/plant, respectively). This trend colud be attributed to the stimulation effect of lowest gamma dose on physiological and biochemical processes in plants owing to the best activation of enzymatic system sustaining natural plant hormons, enhancing metabolism of different components in plant as well as photosynthetic processes reflexing upon the mass growth production of dill plants. In addition, the maximum enhancement of dry mass production was occurred by combined treatment of least effective gamma dose and efficient bio-fertilizer. This trend may be attributed to the presence of additive or cumulative and/or synergistic actions of both factors on plant growth yield. The superiority of bio-fertilizers treatments in plant height, number of branches, and the fresh and dry weights than the control may be due to fix N by microorganisms as well as vitamins, amino acids, cytokinin and gibberellins as growth regulators for the plant (Subb-Rao, 1981; Sprenat, 1990).Low doses of gamma radiation induced plant recognized with higher levels of promoters (gibberellins, auxin and cytokinins) and lower levels of inhibitors (ABA). This change in endogenous hormones which regulate growth and induce low shedding and high yield (Tamas and Engels, 1981; Stajkov et al.,1986). Furthermore, the increment in the obtained fruits yield as a result of exposing the seeds before planting to gamma radiation could be explained through the effective role of radiation in raising the mean of branches/plant (El-Shafie et al., 1993). These results are in agreement with the findings of (Hussein et al., 1990; El-Sherbeny et al.,1992; El-Shafie et al.,1993; Youssef and Moussa, 1998; Ibrahim, 2000; Khalil et al.,2001; Rashed, 2002; Hassanein et al.,2003; Barakat et al.,2004;Kandeel et al.,2004, a and b; Youssef et al.,2004; Garcia-Gonzalez et al.,2005).

3.5. Root Length

It was obvious from Table (3) that, using bio-fertilizer treatments significantly increased the root length (cm) during the two stages of growth in both seasons when compared to untreated plants. In both seasons the greatest root length (cm) were resulted from the treatment of gamma irradiation at rate of 2 k-rad combined with bio-fertilizer that recorded as [28.69, 31.36 (cm)], in the both seasons, respectively). The least root length of treated with 64 k-rad of gamma rays at both stages in both seasons. In addition, the radiation dose from 2 to 16 k-rad in the two seasons significant increased root length in comparison to unirradiated plants. Moreover, there were significant differences between the interaction treatments in the two seasons. Similar results were recorded by (Farooqi et al.,1990; El-Sherbeny et al.,1992; Narimanov and Korystov, 1996; El-Naggar, 1998).

Table 3. Effect of bio-fertilizers, gamma radiation and their interaction treatments onsome growth characters of dill plants during the two seasons (2000/2001 and 2001/2002).

Bio-fertilizers / Gamma radiation (k-rad)	First Season								
	Root length (cm)			Root fresh weight (g)			Root dry weight (g)		
	Without bio-fert.	Bio-fert.	Mean	Without bio-fert.	Bio-fert.	Mean	Without bio-fert.	Bio-fert.	Mean
0	22.20	23.62	22.91	2.41	2.87	2.64	0.70	0.71	0.70
2	28.07	28.69	28.38	3.62	3.97	3.79	1.07	1.23	1.15
4	26.45	26.77	26.61	3.04	3.75	3.39	1.03	1.04	1.03
8	25.49	25.17	25.33	2.83	3.06	2.94	0.93	1.05	0.99
16	24.77	24.97	24.87	2.78	2.77	2.77	0.76	0.78	0.77
32	22.15	22.25	22.20	1.85	1.91	1.88	0.71	0.72	0.71
64	16.52	19.88	18.20	0.99	1.67	1.33	0.34	0.68	0.51
Mean	23.66	24.47		2.50	2.85		0.79	0.88	
L.S.D. at 5%	Bio-fertilizers = 0.59 Radiation = 1.10 Interaction = 1.56			Bio-fertilizers = 0.16 Radiation = 0.31 Interaction = N.S			Bio-fertilizers = 0.06 Radiation = 0.11 Interaction = N.S		
	Second Season								
0	21.29	24.25	22.77	2.25	3.25	2.75	0.70	0.81	0.75
2	27.86	31.36	29.61	3.84	3.87	3.85	1.15	1.27	1.21
4	26.76	27.82	27.29	2.86	3.47	3.16	1.06	1.06	1.06
8	25.28	26.90	26.09	2.66	2.93	2.79	0.91	1.10	1.00
16	24.82	25.10	24.96	2.63	2.65	2.64	0.70	0.74	0.72
32	21.62	23.94	22.78	1.75	1.77	1.76	0.69	0.69	0.69
64	17.76	21.19	19.47	0.97	1.57	1.27	0.32	0.64	0.48
Mean	23.62	25.79		2.42	2.78		0.79	0.90	
L.S.D. at 5%	Bio-fertilizers = 0.64 Radiation = 1.20 Interaction = N.S			Bio-fertilizers = 0.12 Radiation = 0.23 Interaction = 0.33			Bio-fertilizers = 0.04 Radiation = 0.09 Interaction = 0.13		

3.6. Root Fresh Weight

The results reported in Table (3) indicate the effect of bio-fertilizer on fresh weight of dill roots. The effect of bio-fertilizer was significant in the two seasons.The results recorded in the two seasons Table (3) also showed that, the radiation dose at 2 and 4 k-rad significantly increased fresh weight of root, whereas, the radiation from 2 to 32 k-rad significantly increased this character. Also, there was a significant difference between bio-fertilizer and gamma radiation in the second season. The heaviest fresh weights of roots were obtained by the combination of gamma irradiation at 2 k-rad with bio-fertilizer giving (3.97, 3.87 g/plant) in the two seasons, respectively. The beneficial effect of bio-fertilizer and gamma radiation at least dose led to promote the growth substances such as auxins, gibberellins and cytokinins due to their activation for cell division and their elongation causing the enlargment of their voluminous densities. Several investigators reported that application of bio-fertilizers may benefit the plant growth not only by nitrogen fixation but also by production of growth hormones that affect root development and consequently their function in the uptake of both water and nutrients (Hauka, 2000). These results confirm the findings of (Korosi and Pal, 1989; Farooqi et al., 1990; Korystov and Narimanov, 1997; Badawi, 2000; Hassanein et al., 2003; Singh et al., 2005).

3.7. Root Dry Weight

The data presented in Table (3) showed the effect of bio-fertilizer treatments on the dry weight of dill root. In the two seasons, bio-fertilizer resulted in a significant increase in the dry weight of roots compared to the unfertilized control.It can also be seen from the data in Table (3) that, in both seasons the radiation doses from 2 to 8 k-rad significant increased dry weight of root. Also, there was a significant difference in the interaction between bio-fertilizer and gamma radiation doses in the second season. In this concern, the heaviest dry weight of root was givin by combination treatment of irradiation at 2 k-rad with bio-fertilizer during both seasons evaluating the data of (1.23, 1.27 g/plant, respectively). The beneficial effect of bio-fertilizer and gamma radiation at least dose led to promote the growth substances such as auxins, gibberellins and cytokinins due to their activation for cell division and their elongation causing the enlargement of their voluminous denesities. These results are similar to those obtained by (Korosi and Pal, 1989; Farooqi et al.,1990;Korystov and Narimanov, 1997; Badawi, 2000; Hassanein et al.,2003).

4. Conclusion

The recorded results showed that in the two seasons, the most effective treatment on the plant height, root length, fresh and dry weights was 2 k-rad + bio-fertilizers, while combining 4 k-rad with bio-fertilizers gave the best results for numbers of branch.

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