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Radiosensitivity of Onion Seeds With Different Moisture Contents

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ABSTRACT

Seeds of onion (*Allium cepa* L.) cv. Ailsa Craig with different moisture contents (2.92, 4.89, 6.87, 8.88, 11.63, 17.98 and 26.42%) were exposed to various doses of gamma radiation (0, 0.1, 0.2, 0.4, 0.8 and 1.0 kGy) using a Co^{60} source. Seed viability, germination percentage, germination speed, number of normal seedlings and seedling growth decreased, while electrical conductivity of seed leachates and number of abnormal seedlings increased with the increase in radiation dose. The inhibitory effects of radiation were more obvious at the lowest and the higher seed moisture contents. The radiosensitivity of onion seeds varies depending upon the radiation dose; however, levels of seed moisture can modify the radiation effects.

Key Words: *Allium cepa;* Gamma radiation; Onion; Moisture content; Seed irradiation; Seedling

INTRODUCTION

Gamma radiation has been widely used to induce chromosome variation (Aggarwal & Kaul, 1998; Latha & Nair, 1999; Viccini & Carvalho, 2001), produce mutations (Al-Safadi & Simon, 1996; Kumar & Chaudhary, 1996; Munishamanna *et al.*, 1998), enhance seed germination (Bhargava & Khalatkar, 1987; Ripa & Audrina, 1993) or to study its effects on growth, yield and quality (Ahmad & Qureshi, 1992; Al-Ashwat *et al.*, 1995; Veeresh *et al.*, 1995; Sirtautaite, 1996; Munishamanna *et al.*, 1998; Latha & Nair, 1999) in crop plants. Among the various factors, seed moisture at the time of irradiation influences the radiosensitivity of seeds (Singh & Singh, 1978; Mahama & Silvy, 1982; Viccini *et al.*, 1997). Radiosensitivity increases as seed moisture content increases or decreases from the normal level. Minimum radiosensitivity at intermediate seed water content has been demonstrated in several plant species e.g. in rice (Lal & Richharia, 1975), maize (Staikov & Antonov, 1984), and wheat (Kumar, 1978; He, 1985) etc. Singh and Singh (1978) treated the seeds of barley cv. Amber with gamma rays (15 Kr) at normal (11.2%) and different low seed moisture levels (3.0, 3.5, 4.5, 5.5, 6.0 and 7.5%). Low seed moisture levels modified the effects of radiation indicating the influence of seed moisture levels on the effectiveness of gamma rays. Ghiorghita *et al.* (1985) found that when seeds of wheat with water contents of 9.2, 24.2, 29.9, and 34.4% were irradiated with increasing doses of gamma rays, the cytogenetic effects and the inhibition of plant growth were greater with rising seed moisture. Seeds moistened for only 6 h and then irradiated showed stimulated seedling growth. Plant height became increasingly reduced with increase in time of soaking before irradiation. Viccini *et al.* (1997) found that in corn, the damage caused by radiation was more evident on seed with higher moisture content. They also observed increased damage with higher radiation doses. Latha and Nair (1999) exposed barley cv. IB-65 seeds with 4 or 10% moisture content to various doses of gamma radiation. With the increase in radiation dose, there was an almost linear increase in radiation damage but an increase in seed moisture content from 4 to 10%, there was a decrease in radiation damage at all the doses. It is evident from the literature that the majority of the workers have used a small range of moisture content in their seed irradiation experiments and contrasting results have been reported. Moreover, the radiosensitivity also varies from crop to crop and even among different cultivars of the same crop (Bhamburkar & Bhalla, 1980; Ahmad & Qureshi, 1992; Freng, 1996; Kumar & Chaudhary, 1996). Therefore, in the present study, a wide range of seed moisture content has been used to study the effects of seed irradiation and its interaction with seed moisture content in onion.

MATERIALS AND METHODS

Onion (*Allium cepa* L.) cv. Ailsa Craig seeds were obtained from Booker Seeds Ltd., Sleaford, England as a single large aluminum foil pack, containing 1 kg of seeds. The seeds were divided into smaller batches and stored in closed glass bottles in a cold room at 5°C.

Development of various seed moisture contents. Seed samples were equilibrated in atmospheres of known relative humidities for five days at 25±2°C. The atmospheres of various relative humidities were developed by mixing required amounts of sulphuric acid and water in desiccators as described by Solomon (1951). After five days of equilibration, the seed samples attained different moisture contents. Four replicates of 50 seeds were weighed from each batch and evenly spaced in 90 mm Petri dishes placed in an oven at 103±2°C for 17±1 h (ISTA, 1985), cooled in a silica gel container for 15-30 min, after which they were reweighed. The moisture content was expressed as a percentage of the wet weight basis, and calculated as:

$$\text{Percentage moisture content } (\%m.c.) = \frac{M_2 - M_3}{M_2 - M_1} \times 100$$

Where, M_1 is the weight of dish, M_2 is the weight of dish and its contents before drying and M_3 is the weight of dish and its contents after drying.

Irradiation of seeds. Seed samples with different moisture contents were irradiated by gamma radiation using a ^{60}Co source at a dose rate of 20 Gy/minute. The exposure doses used were 0.1, 0.2, 0.4, 0.8 and 1.0 kGy.

Seed viability (TTC test). 1 g of 2,3,5-triphenyl tetrazolium chloride (TTC) was dissolved in 100 mL of distilled water to make a 1% solution. The test was conducted with three replicates of 100 seeds soaked in distilled water for 18 - 20 h. Each seed was cut longitudinally without completely separating the two halves and submerged in 1% TTC solution for at least 8 h at 35 °C in darkness, after which the staining patterns were recorded.

Electrical conductivity test. Three replicates of onion seeds, each of 100 seeds, were placed in 100 mL beakers separately, each containing 75 mL distilled water. The seeds were gently stirred to ensure that all seeds were completely immersed and evenly distributed. The beakers were placed in an incubator at 20±2 °C. After 24 h, the seeds were gently stirred and the conductivities of the soaking waters were measured without filtration using a digital conductivity meter (JENWAY, Model 4070).

Seed germination tests. Germination potential of the onion seeds was estimated in accordance with the International Rules for Seed Testing (ISTA, 1985). Germination percentages, using 3 replicates of 50 seeds, were determined by placing the seed samples in 90 mm Petri dishes on filter papers (Whatman No. 1) moistened with 4 mL of distilled water. Seeds were distributed evenly within each dish. Petri dishes were covered with their lids and then placed in an incubator at 20±2°C with 12 h photoperiod by fluorescent light illumination. Each Petri dish was watered daily with distilled water according to its requirement. Germination in terms of radicle emergence (at least 2 mm) was assessed each day from the second day until no further radicle emergence was noted on two successive days. Germination capacity was expressed as a percentage of all seeds with fully emerged radicle in a given batch. The germination speed of the seed lots was derived from the formula of Kotowski (1926):

$$\text{Germination speed } (G.S.) = \frac{\sum n}{\sum (n \times D_n)} \times 100$$

Where, n is the number of seedlings germinated on day D_n. D_n is the number of days from sowing, corresponding to n, and greater the G.S., higher is the germination speed.

Time to reach 50% of final germination (T_{50}) was calculated from the formula of Coolbear *et al*. (1984):

$$T_{50} = t_i + \left[\frac{(N+1)/2 - n_i}{n_j - n_i}\right] x (t_j - t_i)$$

Where, $n_i < (N+1)/2 < n_j$, N is the number of seeds germinated, and n_i and n_j are total number of seeds germinated by adjacent counts at times t_i and t_j, respectively.

Morphology of seedlings. When the seedlings were 10 days old, these were classified as normal, weak normal or abnormal, following the guidelines given in the International Rules for Seed Testing (ISTA, 1985). Seedlings are classed as abnormal when one or more of the essential structures fails to develop normally because of previous damage to the embryo, or when development as a whole is weak or out of proportion compared with that of a normal seedling germinated at the same time and in the same conditions. Percentages and types of abnormalities were also recorded.

Measurements of seedling lengths as a vigour test. After 10 days from the initiation of imbibition, average shoot lengths, root lengths, and overall seedling lengths were measured using a ruler in mm. Growth reduction in terms of the effects of radiation doses on seedlings, e.g., the percentage change in mean seedling height as compared to the non-irradiated control were recorded as calculated from the following formula:

$$\text{Percentage change} = \frac{H_c - H_r}{H_c} \times 100$$

Where, H_c is the mean seedling height of the non-irradiated (control) and H_r is the mean seedling height of the radiation treatment.

RESULTS

Seed viability (TTC test). There is a degree of variability in the results included in Table I, where the seed viability is recorded against the moisture content of the seeds. A trend can be seen that with an increase in seed moisture content up to 6.87%, the seed viability was increased. However, a further increase in seed moisture content resulted in decrease in the seed viability. The maximum seed viability was at 4.89 and 6.87% seed moisture contents and minimum at the highest moisture content (26.42%) at the time of irradiation. Maximum seed viability was recorded in un-irradiated seeds (control). The seed viability decreased with an increase in radiation dose; however, the decrease was a little. At given moisture content, viability tends to fall with increase in radiation dose.

Conductivity of seed leachate. Seeds of different moisture contents after radiation treatments were soaked in water for

24 h. Measurements of conductivity were made and the seeds showed losses of leachates. It is evident that the seeds of low moisture content suffered greater losses than the moist seeds. The detailed results are presented in Table I. The results of this test were found very variable, both in the amounts of leachates released into the soak water, and between the replicate samples used for each seed treatment. However, it does appear that the leachates from the irradiated samples had generally higher conductivities, and an especially high leakage occurred from very dry seeds when irradiated. It can be seen that seeds of very low moisture content released progressively more leachates into the soak water with increase in radiation dose compared with seeds of intermediate and high moisture contents.

Germination potential following irradiation. Cells and tissues may continue to be subject to change following irradiation, due to the activities of free radicals produced during the actual exposure period. Therefore, to stimulate the metabolic processes, seeds were subjected to germination tests. The results are presented in Table II.

Table I. Direct effect of various doses of gamma radiation on onion seeds with different moisture content, estimated through viability test and electrical conductivity (µS/cm) of their leachates. Figures are the means (± SD) of 3 replicates of 100 seeds each

Exposure dose kGy)	Seed moisture content (%) 2.92	4.89	6.87	8.88	11.63	17.98	26.42	Average
	Seed viability% (TTC test)							
0.0	84 ± 1.0	86 ± 1.5	91 ± 2.0	87 ± 1.0	92 ± 1.5	83 ± 0.5	79 ± 1.0	86.0 ± 1.2
0.1	83 ± 0.5	87 ± 2.0	91 ± 1.5	80 ± 0.5	84 ± 1.5	72 ± 2.0	63 ± 0.5	80.0 ± 1.2
0.2	78 ± 1.5	88 ± 2.5	89 ± 1.5	85 ± 0.5	83 ± 0.5	68 ± 1.5	73 ± 1.5	80.6 ± 1.4
0.4	80 ± 0.5	87 ± 1.0	88 ± 1.5	84 ± 0.5	83 ± 0.5	73 ± 2.5	69 ± 0.5	80.6 ± 1.0
0.8	84 ± 1.5	87 ± 1.0	84 ± 1.0	84 ± 1.0	80 ± 0.5	76 ± 0.5	64 ± 0.5	79.9 ± 0.9
1.0	82 ± 1.0	89 ± 1.5	80 ± 0.5	83 ± 1.0	80 ± 1.0	68 ± 1.0	63 ± 0.5	77.9 ± 0.9
Average	81.8 ± 1.0	87.3 ± 1.6	87.2 ± 1.3	83.8 ± 0.7	83.7 ± 0.9	73.3 ± 1.3	68.5 ± 0.7	
	EC of seed leachate (µS/cm)							
0.0	57.8 ± 0.4	63.8 ± 2.6	56.2 ± 0.0	55.4 ± 3.0	61.5 ± 1.5	64.8 ± 4.4	56.8 ± 1.2	59.5 ± 1.9
0.1	60.3 ± 2.7	56.2 ± 2.1	58.4 ± 0.8	55.9 ± 1.2	57.8 ± 4.1	57.3 ± 0.9	56.4 ± 2.6	57.5 ± 2.1
0.2	62.1 ± 1.0	57.8 ± 2.9	56.6 ± 0.3	57.9 ± 0.5	57.8 ± 0.2	62.0 ± 0.7	52.4 ± 0.2	58.1 ± 0.8
0.4	67.6 ± 0.1	54.6 ± 3.4	62.2 ± 0.3	58.8 ± 0.4	62.2 ± 6.7	61.4 ± 0.2	58.4 ± 1.0	60.7 ± 1.7
0.8	59.6 ± 1.8	56.1 ± 0.7	60.4 ± 1.2	61.1 ± 0.5	69.7 ± 0.8	63.5 ± 3.9	59.2 ± 6.0	61.4 ± 2.1
1.0	80.8 ± 0.5	71.0 ± 4.6	60.1 ± 1.8	60.8 ± 0.9	58.4 ± 2.8	67.8 ± 0.3	64.4 ± 0.0	66.2 ± 1.6
Average	64.7 ± 1.1	59.9 ± 2.7	59.0 ± 0.7	58.3 ± 1.1	61.2 ± 2.7	62.8 ± 1.7	57.9 ± 1.8	

Table II. Onion seeds with different moisture content, tested for their germination potential after exposing to various doses of gamma radiation. Figures are the means (± SD) of 3 replicates of 50 seeds for each test

Exposure dose kGy)	Seed moisture content (%) 2.92	4.89	6.87	8.88	11.63	17.98	26.42	Average
	Final germination (%)							
0.0	75 ± 3.4	75 ± 3.8	80 ± 4.8	80 ± 5.6	87 ± 3.4	71 ± 5.2	74 ± 0.0	77.4 ± 3.7
0.1	81 ± 3.4	80 ± 5.6	88 ± 1.6	76 ± 1.6	78 ± 1.6	69 ± 6.6	61 ± 6.6	76.1 ± 3.9
0.2	76 ± 8.0	83 ± 1.0	87 ± 5.0	81 ± 1.8	75 ± 5.0	64 ± 3.2	67 ± 2.4	76.1 ± 3.8
0.4	79 ± 1.0	79 ± 4.2	83 ± 2.4	82 ± 2.8	77 ± 1.8	71 ± 1.0	61 ± 3.4	76.0 ± 2.4
0.8	82 ± 4.4	83 ± 2.4	80 ± 5.6	81 ± 1.8	74 ± 2.8	75 ± 3.4	61 ± 4.2	76.6 ± 3.5
1.0	79 ± 3.4	87 ± 7.6	74 ± 1.6	77 ± 6.6	78 ± 5.2	65 ± 5.8	57 ± 5.8	73.9 ± 5.1
Average	78.7 ± 3.9	81.2 ± 4.1	82.0 ± 3.5	79.5 ± 3.4	78.2 ± 3.3	69.2 ± 4.2	63.5 ± 3.7	
	Germination speed							
0.0	18.60 ± 1.2	19.21 ± 1.9	19.01 ± 1.3	19.10 ± 1.6	19.60 ± 1.2	19.09 ± 1.1	18.97 ± 1.3	19.08 ± 1.4
0.1	18.73 ± 1.4	18.84 ± 1.2	19.25 ± 1.7	19.36 ± 1.3	19.47 ± 0.9	18.68 ± 1.1	18.45 ± 0.5	18.97 ± 1.2
0.2	18.57 ± 1.1	18.84 ± 1.2	19.07 ± 1.0	19.47 ± 1.5	19.17 ± 0.7	19.07 ± 1.0	19.06 ± 1.3	19.04 ± 1.1
0.4	18.38 ± 0.7	19.13 ± 1.7	18.98 ± 1.1	18.86 ± 0.6	18.96 ± 1.3	20.88 ± 1.9	18.72 ± 1.6	19.13 ± 1.3
0.8	18.69 ± 0.9	18.87 ± 1.1	18.80 ± 1.1	18.91 ± 0.8	18.59 ± 1.5	18.39 ± 1.6	18.45 ± 1.6	18.67 ± 1.2
1.0	18.90 ± 1.3	19.04 ± 1.6	18.42 ± 0.9	19.18 ± 1.0	18.67 ± 1.2	18.36 ± 1.1	18.33 ± 1.7	18.70 ± 1.3
Average	18.65 ± 1.1	18.99 ± 1.4	18.92 ± 1.2	19.15 ± 1.1	19.08 ± 1.1	19.08 ± 1.3	18.66 ± 1.3	
	T_{50} (days)							
0.0	3.74 ± 0.13	3.17 ± 0.21	3.32 ± 0.29	3.25 ± 0.27	3.15 ± 0.29	3.30 ± 0.21	3.27 ± 0.31	3.31 ± 0.24
0.1	3.43 ± 0.19	3.37 ± 0.29	2.91 ± 0.33	3.08 ± 0.24	3.86 ± 0.21	3.49 ± 0.19	3.49 ± 0.27	3.38 ± 0.25
0.2	3.49 ± 0.27	3.39 ± 0.23	3.16 ± 0.15	2.99 ± 0.19	3.20 ± 0.23	3.18 ± 0.35	3.23 ± 0.23	3.23 ± 0.24
0.4	3.70 ± 0.23	3.28 ± 0.35	3.29 ± 0.16	3.36 ± 0.17	3.32 ± 0.25	3.36 ± 0.30	3.36 ± 0.28	3.38 ± 0.25
0.8	3.52 ± 0.32	3.36 ± 0.18	3.39 ± 0.28	3.32 ± 0.23	3.49 ± 0.20	3.70 ± 0.22	3.67 ± 0.26	3.49 ± 0.24
1.0	3.46 ± 0.16	3.30 ± 0.21	3.50 ± 0.27	3.19 ± 0.25	3.53 ± 0.18	3.67 ± 0.24	3.65 ± 0.35	3.47 ± 0.24
Average	3.56 ± 0.22	3.31 ± 0.25	3.26 ± 0.25	3.20 ± 0.22	3.43 ± 0.23	3.45 ± 0.25	3.45 ± 0.28	

Although the final germination percentages are rather variable, the general trend apparent is that with seeds of the lower moisture contents, from 2.9% to about 6.87%, irradiation actually increased the final germination counts obtained, whereas irradiation of seeds having moisture contents above this moisture level progressively reduced germination, especially at higher doses. Further, control samples, with no irradiation, germinated better over the

Table III. Percentages of normal, weak normal and abnormal seedlings from onion seeds of different moisture contents exposed to various doses of gamma radiation. Figures are the means (± SD) of 3 replicates of 50 seeds

Exposure dose kGy)	Seed moisture content (%) 2.92	4.89	6.87	8.88	11.63	17.98	26.42	Average
	Normal seedlings (%)							
0.0	76.9 ± 5.6	66.9 ± 4.2	72.7 ± 3.2	65.8 ± 4.2	80.4 ± 7.4	70.0 ± 4.2	73.7 ± 5.0	72.3 ± 4.8
0.1	56.5 ± 5.0	59.7 ± 4.0	64.5 ± 4.0	63.7 ± 1.0	70.1 ± 2.0	53.2 ± 7.0	63.4 ± 3.0	61.6 ± 3.7
0.2	50.6 ± 0.0	53.0 ± 2.0	54.3 ± 4.0	53.0 ± 3.0	57.3 ± 1.0	30.5 ± 1.0	55.7 ± 1.0	50.6 ± 1.7
0.4	46.4 ± 2.0	40.7 ± 1.0	40.2 ± 3.0	32.9 ± 1.0	44.2 ± 2.0	31.5 ± 3.0	26.4± 1.0	37.5 ± 1.9
0.8	24.7 ± 0.0	23.5± 1.0	26.5 ± 1.0	21.8± 1.0	33.7 ± 1.0	26.6 ± 1.0	16.1± 0.0	24.7 ± 0.7
1.0	25.3 ± 1.0	13.2± 0.0	34.6 ± 2.0	16.7± 1.0	31.2 ± 1.0	19.7 ± 3.0	0.0 ± 0.0	20.1 ± 1.1
Average	46.7 ± 2.3	42.8 ± 2.0	48.8 ± 2.9	42.3 ± 1.9	52.8 ± 2.4	38.6 ± 3.2	39.2 ± 1.7	
	Week normal seedlings (%)							
0.0	5.0 ± 3.2	3.1± 2.6	1.6 ± 1.0	7.0 ± 0.0	0.7 ± 1.0	3.2 ± 1.8	7.1 ± 2.6	4.0 ± 1.7
0.1	15.3 ± 3.0	13.0 ± 2.0	10.8 ± 0.0	8.8 ± 1.0	11.7 ± 3.0	11.3 ± 5.0	18.3 ± 1.0	12.7 ± 2.1
0.2	22.9 ± 3.0	19.3 ± 2.0	12.0 ± 1.0	18.8 ± 0.0	14.7 ± 2.0	27.5 ± 3.0	25.7 ± 2.0	20.1 ± 1.9
0.4	26.8 ± 2.0	31.4 ± 1.0	34.5 ± 4.0	26.8 ± 2.0	34.9 ± 0.0	31.5 ± 1.0	37.5 ± 1.0	31.9 ± 1.6
0.8	29.6 ± 2.0	34.5 ± 2.0	37.9 ± 1.0	28.8 ± 2.0	32.6 ± 0.0	40.5 ± 2.0	33.9 ± 1.0	34.0 ± 1.4
1.0	35.5 ± 2.0	43.9 ± 2.0	38.3 ± 1.0	33.3 ± 2.0	35.0 ± 0.0	30.3 ± 6.0	26.6 ± 1.0	34.7 ± 2.0
Average	22.5 ± 2.5	24.2 ± 1.9	22.5 ± 1.3	20.6 ± 1.2	21.6 ± 1.0	24.1 ± 3.1	24.9 ± 1.4	
	Abnormal seedlings (%)							
0.0	18.1 ± 5.2	30.0 ± 6.2	25.7 ± 1.0	27.2 ± 3.4	18.9 ± 5.8	26.8 ± 7.2	19.2 ± 6.6	23.7 ± 5.1
0.1	28.2 ± 4.0	27.3 ± 3.0	24.7 ± 1.0	27.5 ± 0.0	18.2 ± 0.0	35.5 ± 4.0	18.3 ± 1.0	25.7 ± 1.9
0.2	26.5 ± 3.0	27.7 ± 1.0	33.7 ± 5.0	28.2 ± 4.0	28.0 ± 1.0	42.0 ± 1.0	18.6 ± 3.0	29.2 ± 2.6
0.4	26.8 ± 2.0	27.9 ± 2.0	25.3 ± 4.0	40.3 ± 1.0	20.9 ± 4.0	37.0 ± 1.0	36.1 ± 2.0	30.6 ± 2.3
0.8	45.7 ± 7.0	42.0 ± 0.0	35.6 ± 7.0	49.4 ± 3.0	33.7 ± 1.0	32.9 ± 0.0	50.0 ± 3.0	41.3 ± 3.0
1.0	39.2 ± 6.0	42.9 ± 7.0	27.1 ± 0.0	50.0 ± 3.0	33.8 ± 1.0	50.0 ± 5.0	73.4 ± 1.0	45.2 ± 3.3
Average	30.8 ± 4.5	33.0 ± 3.2	28.7 ± 3.0	37.1 ± 2.4	25.6 ± 2.1	37.4 ± 3.0	35.9 ± 2.8	

Table IV. Seedling lengths, and decrease in growth (%) of seedlings from onion seeds of different moisture contents exposed to various doses of gamma radiation. Figures are the means (± SD) of 4 replicates of 10 seedlings

Exposure dose kGy)	Seed moisture content (%) 2.92	4.89	6.87	8.88	11.63	17.98	26.42	Average
	Overall seedling length (mm)							
0.0	116.2 ± 8.5	100.0 ± 9.5	114.7 ± 8.7	122.3 ± 8.8	119.2 ± 8.0	127.2 ± 9.3	119.1 ± 8.6	117.0 ± 8.8
0.1	73.6 ± 9.9	81.3 ± 8.4	80.4 ± 7.1	98.8 ± 8.5	76.3 ± 6.8	74.7 ± 7.0	56.5 ± 7.3	77.4 ± 7.9
0.2	54.5 ± 6.1	63.3 ± 9.5	71.1 ± 7.4	66.0 ± 4.3	45.7 ± 5.7	37.4 ± 3.8	37.3 ± 6.4	53.6 ± 6.2
0.4	36.4 ± 4.7	39.3 ± 3.8	35.4 ± 4.8	35.1 ± 3.7	33.0 ± 2.5	32.1 ± 4.3	27.6 ± 2.4	34.1 ± 3.7
0.8	28.4 ± 4.1	29.6 ± 3.4	30.3 ± 4.3	28.8 ± 2.5	31.8 ± 3.7	27.2 ± 2.9	28.4 ± 5.1	29.2 ± 3.7
1.0	33.8 ± 3.3	32.1 ± 2.5	34.6 ± 4.5	29.6 ± 3.6	29.7 ± 2.9	28.5 ± 4.6	26.6 ± 2.4	30.7 ± 3.4
Average	57.2 ± 6.1	57.6 ± 6.2	61.1 ± 6.1	63.4 ± 5.2	55.9 ± 4.9	54.5 ± 5.3	49.3 ± 5.4	
	Shoot and root (in parenthesis) lengths (mm)							
0.0	77.4 (38.8)	67.1 (32.9)	73.2 (41.5)	74.2 (48.1)	75.4 (43.8)	79.5 (47.7)	75.4 (43.7)	74.6 (42.4)
0.1	43.0 (30.6)	41.8 (39.5)	50.2 (30.2)	57.8 (41.0)	50.6 (25.7)	50.4 (24.3)	43.5 (13.0)	48.2 (29.2)
0.2	33.7 (20.8)	38.4 (24.9)	47.5 (23.6)	42.1 (23.9)	30.6 (15.1)	27.0 (10.4)	29.5 (7.8)	35.5 (18.1)
0.4	24.9 (11.5)	26.1 (13.2)	26.3 (9.1)	24.7 (10.4)	24.5 (8.5)	24.2 (7.9)	22.7 (4.9)	24.8 (9.4)
0.8	22.0 (6.4)	22.4 (7.2)	23.4 (6.9)	22.5 (6.3)	26.3 (5.5)	21.5 (5.7)	24.3 (4.1)	23.2 (6.0)
1.0	27.5 (6.3)	25.3 (6.8)	29.0 (5.6)	23.6 (6.0)	24.5 (5.2)	22.3 (6.2)	23.1 (3.5)	25.0 (5.7)
Average	38.1 (19.1)	36.9 (20.8)	41.6 (19.5)	40.8 (22.6)	38.7 (17.3)	37.5 (17.0)	36.4 (12.8)	
	Growth reduction (%)							
0.0	-	-	-	-	-	-	-	-
0.1	36.7 ± 2.5	18.7 ± 1.3	29.9 ± 2.7	19.2 ± 1.5	36.0 ± 3.2	41.3 ± 3.1	52.6 ± 4.5	33.5 ± 2.7
0.2	53.1 ± 4.7	36.7 ± 2.9	38.0 ± 2.8	46.0 ± 4.1	61.7 ± 4.9	70.6 ± 5.8	68.7 ± 5.1	53.5 ± 4.3
0.4	68.7 ± 5.2	60.7 ± 5.4	69.1 ± 4.7	71.3 ± 4.9	72.3 ± 5.3	74.7 ± 6.2	76.8 ± 5.7	70.5 ± 5.3
0.8	75.6 ± 5.1	70.4 ± 6.0	73.6 ± 6.1	76.5 ± 5.8	73.3 ± 6.5	78.6 ± 4.9	76.2 ± 5.3	74.9 ± 5.7
1.0	70.9 ± 5.7	67.9 ± 6.2	69.8 ± 4.1	75.8 ± 5.3	75.1 ± 5.8	77.6 ± 6.1	77.7 ± 6.6	73.5 ± 5.7
Average	61.0 ± 4.6	50.9 ± 4.4	56.1 ± 4.1	57.8 ± 4.3	63.7 ± 5.1	68.6 ± 5.2	70.4 ± 5.4	

middle range of moisture contents. The apparent reduction in germination at higher moisture contents with the same radiation dose as compared with lower moisture content can be seen (Table II). The data show that although the radiation doses appear to affect germination to approximately the same extent, seeds of the highest moisture content showed the lowest germination levels. These results in general support the TTC test, although, as is usually found, slightly higher viabilities were shown by the biochemical test. Wide variations are again seen in the speeds of germination of the seeds from each treatment. The highest germination speed was recorded with seeds having 8.88% moisture content. However, an increase or decrease in moisture content from this level resulted in germination speed. Germination speed was also affected by irradiation doses, however, the effects were less pronounced. Time to complete 50% seed germination followed the same trend. Seeds with 8.88% moisture took minimum time to complete 50% germination. As the moisture content increased or decreased, the time to complete 50% seed germination was increased (Table II).

Seedling morphology. The seedlings were evaluated ten days after the beginning of the germination test, and the numbers of apparently normal, weak, and abnormal seedlings were recorded for each radiation dose administered to the samples of onion seeds of different moisture contents (Table III). Number of normal seedlings decreased and that of weak normal and abnormal seedlings increased with the increase in radiation dose. Although there is variability in the results presented in Table III, however, it is clear that for each level of moisture in the seeds, the numbers of abnormal seedlings increased with increase in the irradiation dose, in most cases very steeply. However, seed moisture levels above 11.63% resulted in the minimum normal number of seedlings and maximum week and abnormal seedlings. The following recognizable categories of abnormal seedlings were found, as described in the ISTA (1985) International Rules for Seed Testing: a) without primary root (Ia), b) primary root short and stunted (Ib), c) poorly developed leaf-like cotyledon without a definite bend or "knee" (IVc), and d) short and weak, or spindly, or watery seedling (VIa).

Seedling growth. The results show that irradiation at each dose administered, from 0.1 to 1.0 kGy, caused severe reductions in overall seedling length compared with normal growth. Both root and shoot lengths were affected, but the roots were affected to a greater extent. It appears that although growth of the roots of non-irradiated seedlings tended to be improved with increasing seed moisture content before germination, their growth became severely inhibited by irradiation of the seeds at the higher moisture contents (Table IV). When seedling growth is compared directly with the irradiation dose given, the severe effects of increasing irradiation dose become very obvious. When the percentage reduction in growth is compared with the seed moisture content, it shows both an increase in the inhibiting effect of radiation doses administered, and also an increase in inhibition with rise in seed moisture content. It can also be seen, especially at the lower doses of 0.1 and 0.2 kGy, that the effect on seedling growth is most severe over the range of moisture contents (Table IV).

DISCUSSION

Biological material is usually associated with considerable amounts of water, e.g., the living cell contains more than 70% water. Water present in the seed, even when air-dry, is significant because of the role of water in peroxide formation, and is probably one of the most critical variables which could affect both the optimal dose and the degree of response of the seed (Bhattachariya & Joshi, 1977). The direct effects of the various radiation doses on the batches of seeds at widely different moisture contents, from 2.92 to 26.42%.

Several authors (e.g. Singh & Kumar, 1973; Lal & Richharia, 1975; Singh *et al.*, 1976; Kumar, 1978; Mahama & Silvy, 1982; He, 1985; Israni *et al.*, 1993; Latha & Nair, 1999) have reported that the damage is greater if seeds are irradiated when very dry (3-5%). Most results were obtained using only the range of moisture contents from normal storage moistures of 10-12% down to about 3%, with the exception of Mahama and Silvy (1982), who showed that very dry seeds of *Hibiscus* were most susceptible to radiation damage and almost as sensitive when moist. In the present study, seeds of different moisture contents showed that the seeds of the lower pre-imbibition moisture contents experienced greater losses of leachates than the moist seeds. It has already been shown that when rapidly imbibed, dry seeds suffer greater damage (Ellis *et al*., 1982). When irradiated, seed leachates showed higher conductivities, and again, fairly steady increases in electrolyte loss occurred with increasing dose of radiation in the very dry seeds. Electrolyte losses were reduced in seeds irradiated at the highest moisture level, in agreement with Dasgupta *et al*. (1977), who showed that either soaking seeds in water for a few hours, or even seed equilibration in a saturated atmosphere exerted a protective effect against loss of leachates from seeds of wheat and jute. The present work examined a wide range of moisture contents, and found that the damage also increased as the seed moisture content was raised from the normal air-dry state to high moisture levels. These results are in agreement with the findings of Mahama and Silvy (1982), He (1985) and Viccini *et al.* (1997).

The un-irradiated seeds showed the highest levels of germination at the middle range of moisture contents, and the poorest germination at the lower and higher moisture contents. This was shown to be supported by the results of both the TTC viability test and the germination speed test. When irradiated, however, seeds of the lower moisture contents tended to show increased germination compared with the control seeds. This effect is probably related to what is known as radiation hormesis (Sheppard & Hawkins, 1990). There is apparently no accepted explanation for this phenomenon, although it has been suggested that low levels of stress might call into operation certain metabolic processes which could overcompensate for the damage caused by the stress, thus leading to enhanced performance. It should be noted, however, that in the present case, the hormetic effect only operated at moisture contents up to about 11.63%, and at moisture contents above this level, all the radiation doses were damaging, especially so at the high doses of radiation. The numbers of abnormal seedlings found increased with the radiation dose at all moisture contents, in some cases rising very steeply.

When the seeds were not irradiated, seedling growth in length tended to increase with increase in the moisture content of the seeds. However, seedling growth in terms of length became severely reduced by irradiation, especially at higher moisture contents, from about 11.63 to 26.42%. These results are in accordance with the findings of Staikov and Antonov (1984) and Viccini *et al.* (1997), who found that the damage caused by radiation was more evident on seeds with higher moisture contents. In the present study, the root growth of seedlings was affected to a greater extent than shoot growth in length, which has commonly been reported for other species, e.g., in rye (Savaskan & Toker, 1991) and the effect was both severe and consistent. From the results of the present study, it can be concluded that the sensitivity of the onion seeds varies according to the seed moisture content and also the radiation dose.

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