Some Effects of Ionizing Radiation on the Metabolism and Growth of Sugar Beets

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Introduction

Since the discovery of X rays in 1895, plants have been used extensively in studies of the effects of ionizing radiation on living material. Almost every conceivable type of study has been made using seeds, pollen, embryos, isolated tissues of various kinds, whole plants, and isolated subcellular structures such as chloroplasts and mitochondria. Studies have been made using all kinds of ionizing radiation such as X rays; alpha, beta and gamma radiation; neutrons; etc. Studies have been made from almost every point of view; i.e., in terms of radiation effects on the physiology, biochemistry, cytology, genetics, histology, morphology and embryology of plants. The general field has been reviewed recently by Sparrow (9)[4], Gunckel and Sparrow (3), and Spikes (11). The present studies were undertaken primarily to determine whether treatment with ionizing radiation would improve storage life of harvested sugar beets. At the same time, several other aspects of the radiation biology of sugar beets were surveyed.

Radiation Effects on Root Respiration

The effect of ionizing radiation on plant storage tissue depends on plant species, radiation dosage and other factors. Sussman (14) found that dosages of only a few thousand roentgens (r) significantly extended the storage life of potatoes by preventing sprouting; radiation increased the respiratory rate, however. Gustafson, et al. (4) also found that gamma radiation increased the respiratory rate of potatoes for several weeks; after this the respiratory rate decreased and reached a low point in approximately seven weeks. Dallyn, et al. (1) showed that the sprouting of onions could also be prevented by gamma radiation with a corresponding increase in storage life. Smock and Sparrow (7) showed that 2,500 r of gamma radiation decreased the respiratory rate of Cortland apples.

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[4] Numbers in parentheses refer to literature cited.

Glegg, et al. (2) examined the softening effects of gamma radiation on red beets (Detroit dark red). There was little change in tissue properties up to a "threshold" dosage of about 300,000 r. Above this, the resistance of tissue to crushing declined rapidly. They did not make any measurements on beet respiration. Snyder and Wiant (8) found that small dosages of high energy electron radiation stimulated respiratory processes without any visible injury to sugar beet roots. Higher dosages injured surface tissues. Since sugar beets often show large respiratory sugar losses during the storage period between harvesting and processing (12), an examination of the effect of ionizing radiation on sugar beet root respiratory rates was made to determine if such treatment might offer a useful technique for reducing respiratory losses.

Large sugar beet roots were split lengthwise with a saw and each half was wrapped in moist paper toweling. One-half of each beet was exposed to gamma radiation from a cobalt^{60} source[5], while the other halves were stored in the same room but protected from irradiation by appropriate shielding. Dosages of 5,000 to 20,000 r were used. Following irradiation, all pieces were stored moist in a refrigerated room at 1° to 2° C. Periodically thereafter, matched pieces were sawed from each beet-half and sliced to 1 mm thickness; disks were then cut with a cork borer, washed, and tissue respiratory rates determined by the Warburg technique, as previously reported (13) .Results, as shown in Table 1, indicate an appreciable increase in respiratory rate following irradiation. This increased rate did not subside for about 70 days. It would appear, then, that treatment of sugar beets with gamma radiation in the dosage range indicated would not represent a useful technique for decreasing respiratory sugar losses in stored sugar beets.

Table 1.—Effect of gamma irradiation on the respiration of sugar beet root tissue. Values in percentage of matched nonirradiated check (CO^{60} source).

Days after treatment	5,000 r	10,000 r	20,000 r	Mean
	Percent	*Percent*	*Percent*	
6	112	118	120	117
13	101	123	112	112
21	114	122	125	120
29	113	126	111	117
40	121	106	129	119
69	101	98	102	100
Mean	110	115	116	

Radiation Effects on Leaf Metabolism

There appear to be very few reports in the literature concerning the effects of ionizing radiation on leaf respiration (3). In the present experiments, disks were cut from mature sugar beet leaves and samples irradiated with a cobalt^{60} source[5] for varying dosages. The disks were then stored in plastic bags in a cold room at 2° C. At intervals, samples were removed and respiratory rates determined manometrically. Irradiated leaf disks, as shown in Table 2, respired more rapidly than unirradiated checks for several days. At higher dosages, respiratory rates then decreased with time, presumably as the cells gradually died. Dosages of over a million r were required to kill sugar beet leaf tissue immediately.

Table 2.— Effect of X-irradiation on the respiration of sugar beet leaf tissue.

Time after treatment	91,000 r	178,000 r	356,000 r	713,000 r
	Percentage of untreated check			
Hours	*Percent*	*Percent*	*Percent*	*Percent*
3	123	113	109	120
25	119	116	114	112
54	120	110	109	104
76	118	111	109	98
144	110	105	94	49
341	105	88	90	5

Some immediate chlorophyll destruction occurs in irradiated sugar beet leaves; however, this is not large in the dosage range up to several hundred thousand r. In contrast, chlorophyll dissolved in organic solvents is relatively sensitive to ionizing radiation. The effects of ionizing radiation on leaf photosynthesis are complex (3). For example, a dosage of 30,000 r will decrease the rate of carbon dioxide fixation in wheat leaves by 50 percent (15). This same dosage, however, had no detectable effects on the oxygen evolving ability (Hill reaction) of chloroplasts isolated from the irradiated leaves. This probably means that some enzyme system involved in the carbon dioxide-fixing steps is relatively sensitive, while those concerned with water-splitting and oxygen evolution are rather insensitive. In the present work no measurements were made on whole-leaf photosynthesis. However, studies were made of the effects of radiation on the photochemical oxygen evolution (Hill reaction) of isolated sugar beet chloroplasts. Hill reaction activity was relatively insensitive to

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[5] These irradiations were carried out with a 5,000 curie cobalt^{60} source and with a gamma source containing appropriately aged spent reactor fuel rods. The irradiation was performed by Dugway Proving Grounds, Utah, through the cooperation of Dr. C. J. Christensen of the University of Utah.

gamma radiation. The activity of irradiated chloroplasts was measured potentiometrically (10) at a series of different light intensities. This permitted determining the effects of radiation separately on the rate-limiting photochemical process and the rate-limiting dark process of the Hill reaction (6). Dosages up to 750,000 r actually increased the rate of the photochemical process of the Hill reaction; above this inhibition occurred, but loss was very slight, even at 1,000,000 r. The dark process was more sensitive, being decreased about 50% at dosages of 1,000,000 r. Radiation damage to chloroplasts was decreased somewhat by the usual protective agents such as beta-aminoethylisothiuronium salts (AET). This work will be published in detail elsewhere.

Radiation Effects on Seed Germination and Respiration

There is a tremendous literature concerning effects of many kinds of ionizing radiation on seed germination and the early growth of seedlings (3). Generally speaking, with increased dosages, seed germination is delayed and seedling growth is decreased. Numerous types of morphological abnormalities are also produced. In a few species, however, low dosages appear to be somewhat stimulating. There are only a few reports concerning radiation effects on the germination of sugar beet seeds.

In the present work, sugar beet seeds were subjected to X rays from a 250 KVA machine with dosages ranging from 3,000 to 1,000,000 r. One group of samples was irradiated in an air-dry condition, while the other group was irradiated while moist,

Table 3.—Effect of X-irradiation of dry sugar beet seeds on percent germination and mortality of seedlings. (Decline in percent germination indicates some seedlings died.)

Time after treatment	0 r	3,000 r	10,000 r	33,000 r	100,000 r	333,000 r	1,000,000 r
Hours	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*
90	4	2	0	0	0	0	0
98	24	10	28	0	4	0	0
116	63	40	59	6	20	2	0
124	63	54	75	15	35	8	0
136	65	58	78	36	49	19	0
143	63	58	78	40	47	23	0
162	69	64	81	46	51	26	0
184	71	64	88	44	51	28	0
232	71	64	81	44	56	28	8
256	71	64	78	44	56	26	8
330	67	60	78	40	56	21	11
401	67	60	78	27	47	15	14
461	67	60	78	6	47	2	14
522	69	62	72	2	47	2	8
602	67	62	72	0	42	2	3
666				0	33	2	0

following 20 hours of washing in running tap water. The data on seeds irradiated in the dry condition are presented in Table 3. As may be seen, germination is delayed, and the percentage of seeds germinating is generally decreased with increasing dosage. Seedling growth was reduced, and a variety of leaf abnormalities were observed. Seeds irradiated in a moist condition were considerably more sensitive to ionizing radiation, as shown in Table 4. This phenomenon is well known (3).

Table 4.—Effect of X-irradiation of moist sugar beet seeds on percent germination and mortality of seedlings. (Seeds washed 20 hours in running tap water and left moist during irradiation.) (Decline in percent germination indicates some seedlings died.)

Hours after treatment	0 r control	3,000 r	10,000 r	33,000 r	100,000 r		1,000,000 r
		germination					
Hours	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*
90	16	0	0	0	0	0	0
110	46	7	2	4	0	0	0
136	58	20	10	16	0	0	0
162	63	25	12	19	0	0	0
184	65	30	22	19	0	0	0
240	67	39	21	26	4	7	0
255	67	39	25	28	4	7	0
330	67	36	27	26	4	6	0
401	65	36	28	18	4	6	0
461	60	36	28	9	4	4	0
522	63	34	38	4	15	2	0
602	65	36	40	4	17	4	0

Mikaelsen, et al. (5) found that neutron dosages of 30-3600 reps had little effect on the respiration of germinating barley seeds during the first 2 days of germination. After this, irradiated plants showed a reduced respiratory rate which was correlated with radiation-produced reduction in seedling length. In the present work, sugar beet seedballs were X-rayed with dosages of 5,000 to 50,000 r. The seeds were then soaked, allowed to germinate, and the percent germination and respiratory rates determined at intervals, as shown in Table 5. Germination was progressively inhibited by increasing X-ray dosages. Irradiation stimulated respiration for the first 48 hours after treatment, even though germination was inhibited. After 73 hours, respiration was lower in the irradiated samples, but after 138 hours respiration was about the same in the treated and untreated samples.

Radiation Effect on Steckling Growth

Steckling sugar beets were trimmed, and 16 lots of 10 beets each were irradiated at dosages from 200 to 1,000,000 r. A cobalt^{60} source was used for dosages up to 5,000 r; for higher dosages a

Table 5.—Effect of X-irradiation on the germination and respiration of sugar beet seedballs. Irradiated samples are reported in percentage of the values of controls.

Time after treatment	Actual values per 100 seedballs		Irradiation dosage—germination and respiration values in percent of untreated seedballs.					
			5,000 r	10,000 r	15,000 r	20,000 r	30,000 r	50,000 r
Hours								
						No visible germination		
0.75	% germ.	----	----	----	----	----	----	----
	ul O_2/hr.	25	80	144	148	72	160	252
						No visible germination		
24.5	% germ.	----	----	----	----	----	----	----
	ul O_2/hr.	39	100	150	125	154	146	120
48.5	% germ.	10	120	40	50	30	30	10
	ul O_2/hr.	196	----	113	131	117	103	93
73	% germ.	50	94	90	96	71	82	44
	ul O_2/hr.	735	.	94	73	65	73	58
96.5	% germ.	78	92	94	92	87	85	72
	ul O_2/hr.	----	----	----	----	----	----	----
138	% germ.	----	----	----	----	----	----	----
	ul O_2/hr.	795	.	107	102	87	99	110
166.5	% germ.	82	93	95	94	85	83	71

Figure 1.—Effect of gamma radiation on regrowth of sugar beets —23 days after replanting.

radiation source containing appropriately aged, reactor fuel-rods was used. The beets were then planted in soil in aluminum cylinders. Typical effects of radiation on beet regrowth after 23 days are shown in Figure 1. None of the beets survived dosages of 200,000 r or greater; no regrowth was observed at dosages of 100,000 r, and almost all plants were killed at this dosage. The leaves of irradiated beets showed a marked increase in pubescence; in general, new leaves were finely mottled, and a variety of leaf and petiole abnormalities were observed, including leaf-blade reduction and fusion of adjacent petioles. Some of the irradiated beets showed a loss of apical dominance. In other experiments, groups of steckling beets of three varieties were irradiated with dosages of 2,000 to 5,000 r and then replanted in a field plot. The observed effects were similar to those described above. Radiation effects of these general types have been shown with a variety of other plants (3).

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