

# Effect of Irradiation on Quality Characteristics of Clipper Barley and Malt<sup>1</sup>

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## ABSTRACT

Clipper barley was subjected to three different levels of <sup>60</sup>Co irradiation to determine its effect on fungal counts and barley and malt quality. Resistance to irradiation differed among the various fungi tested. This resulted in a rapid increase in numbers during the malting process in some of the more resistant strains. Malt quality parameters were sensitive to <sup>60</sup>Co irradiation. A lowering of extract levels, total soluble nitrogen, free amino nitrogen, and  $\alpha$ - and  $\beta$ -amylase levels and an increase in color and viscosity were observed with increasing doses of radiation.

Keywords: Barley, Fungi, Irradiation, Malt, Quality

Grain harvested under normal field conditions contains microorganisms. The nature and magnitude of the microflora of barley depend on the climatic conditions under which the crop was grown as well as on the postharvest history of the grain (13). The effects of these microflora on the quality of barley, malt, and beer range from a drop in the germinative capacity of kernels (13,17) to varying changes in malt parameters (5,14) and to reduced gas stability, i.e., gushing of beer (13). Microorganisms also may produce toxins. These chemical compounds are known to be harmful to humans and animals (26).

Therefore, it is important that the levels of microorganisms and insects be controlled. Conditions for control currently in use and under investigation include altering temperature and water availability (22), adding chemicals (7,8,16), making storage areas airtight (29), controlling atmospheres (18,25), or subjecting grain to gamma radiation, also called radurization (9,28,33).

Barley for malting usually is stored to overcome dormancy and to maintain stocks. It is therefore essential that the grain be preserved to prevent infestation by insects and/or microorganisms, but it must be preserved without any deleterious effects on the malting potential of the grain. The effect of gamma radiation on the malting quality of barley has been studied to some extent but has resulted in contrasting conclusions (2,12). The objective of this investigation was to study the effect of gamma irradiation on the microflora and malting quality of Clipper barley stored over a period of 12 months.

## EXPERIMENTAL

Clipper barley (1987 crop) obtained directly after harvest was screened over a 2.20-mm screen, cleaned, and put into 1-kg containers. Two months after harvest, these samples were irradiated using 0-, 2.5-, 5.0-, and 7.5-kiloGray (kGy) doses in the <sup>60</sup>Co gamma chamber at High Energy Processing Ltd., a commercial irradiation unit at Cape Town, South Africa. To avoid possible formation of condensation, the openings of the containers were covered with double layers of cloth consisting of a random surface of continuous filament polypropylene. The samples were stored at room temperature for the duration of the experiment. Samples were malted and analyzed at three-month intervals over a period of 12 months.

One-kilogram samples of each treated sample were micromalted simultaneously in an automatic Seeger malting unit (Seeger GmbH, Plüderhausen, Germany). The malting conditions were as follows. The steep cycle consisted of 8 hr wet, 15 hr dry, 9

hr wet, and 9 hr dry at 17°C. Germination was at 17°C for 76 hr. At steep-out, samples were sprayed, if needed, with distilled water, in order for them to reach a moisture level of approximately 46.5%. The samples then were kilned for 14 hr at 65°C and 4 hr at 80°C.

Green malt samples for fungal evaluation were drawn just before kilning and dried for 16 hr at 40°C. Germination energy and all malt analyses were performed according to standard methods (11).

Samples for mycological assay were not surface-disinfested. Twenty-five kernels were aseptically transferred to petri dishes containing potato-dextrose agar (39 g of potato-dextrose agar and 1 mg of chloromycetin in 1 L of distilled water sterilized at 121°C for 15 min) and malt-salt agar (8 g of technical agar, 20 g of malt extract broth, and 75 g of sodium chloride in 1 L of distilled water sterilized at 121°C for 15 min). Incubation at 26°C for seven days followed. Fungal strains were identified to genus level under a microscope according to morphological features as described by Barnett and Hunter (4). Results were expressed as percentage of kernels contaminated with the respective genera of fungi.

Evaluation of fungi on green malt and kilned malt samples was mainly to determine the degree to which various fungi would spread to sterilized or to partially sterilized grain. It must be borne in mind that, although complete sterilization of the grain can be accomplished, components of the malting plant, such as steep tanks and germination boxes, cannot be completely sterilized.

All results were statistically compared by means of the least significant difference test. For statistical evaluation, fungal analyses performed at three monthly intervals were grouped together and further treated as repeats.

## RESULTS AND DISCUSSION

### Fungi Levels on Barley and Malt

Fungi isolated and identified in barley and malt samples included *Alternaria*, *Aspergillus*, *Epicoccum*, *Fusarium*, and *Penicillium*. The percent contamination of barley kernels

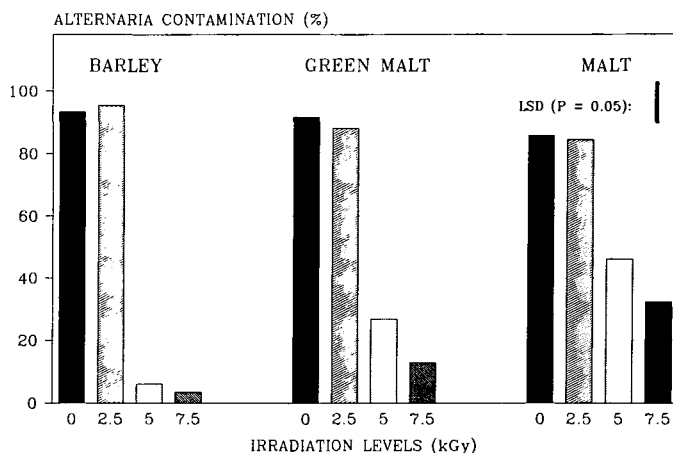


Fig. 1. Effect of <sup>60</sup>Co irradiation on the percentages of Clipper barley, green malt, and malt kernels contaminated with *Alternaria*.

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irradiated with different levels of <sup>60</sup>Co, as well as green malt and malt kernels, were determined.

The percentage of barley kernels contaminated with *Alternaria* was significantly reduced only after irradiation with 5.0-kGy (and higher) doses of <sup>60</sup>Co (Fig. 1). The resistance of *Alternaria* spp. to gamma irradiation also was found by Barkai-Golan (3) and by Mohyuddin and Skoropad (24). Green malt and malt produced from barley irradiated with 5.0- and 7.5-kGy doses of <sup>60</sup>Co showed similar contamination by *Alternaria*. These results further indicate that, given the opportunity, *Alternaria* could spread from "pockets" of mold-infected grain and proliferate in barley free from high levels of mold while steeping and germination take place. Haikara et al (15) observed small changes in *Alternaria* levels during the malting process, whereas Douglas and Flannigan (10) observed first a decrease and then an increase in contamination by *Alternaria* in two consecutive production runs.

Five- and 7.5-kGy doses of <sup>60</sup>Co irradiation virtually eliminated *Fusarium* on the barley used in this work (Fig. 2). In tests performed by Mohyuddin and Skoropad (24), 10-kGy doses of <sup>60</sup>Co irradiation were required for complete eradication of *Fusarium* spp. A significant increase in the percentage of green malt and malt kernels contaminated with *Fusarium* was seen. Green malt and malt produced from irradiated barley samples had higher percentages of *Fusarium* contamination than did the

controls. These results show the tendency of *Fusarium* to spread and proliferate during the malting process. Extensive increases in *Fusarium* contamination, especially during the early stages of malting, were previously reported (10,15).

Doses of <sup>60</sup>Co irradiation of 5.0 kGy and higher significantly reduced the percentage of barley kernels contaminated with *Epicoccum* spp. During malting, little change in the levels of *Epicoccum* contamination was detected (Fig. 3). There was minimal spread to sterilized samples during steeping and germination. Douglas and Flannigan (10) observed a decrease in levels of *E. purpurascens* on green malt as well as on the final malt.

The percentage of barley kernels contaminated with *Aspergillus* was significantly reduced after irradiation with 2.5-kGy (and higher) doses of <sup>60</sup>Co (Fig. 4). This sensitivity of *Aspergillus* to <sup>60</sup>Co irradiation also was reported by Mohyuddin and Skoropad (24). On tests performed on wheat, they found that a dose of 3 kGy was required to completely inactivate the fungi. The increase in *Aspergillus* contamination on the green malt produced from the irradiated barley indicated that cross contamination by *Aspergillus* can occur during the malting process and that sterilized kernels were especially prone to colonization by *Aspergillus*. Kilning, however, caused a significant decrease in the percentage of kernels bearing viable *Aspergillus*. Haikara et al (15) reported

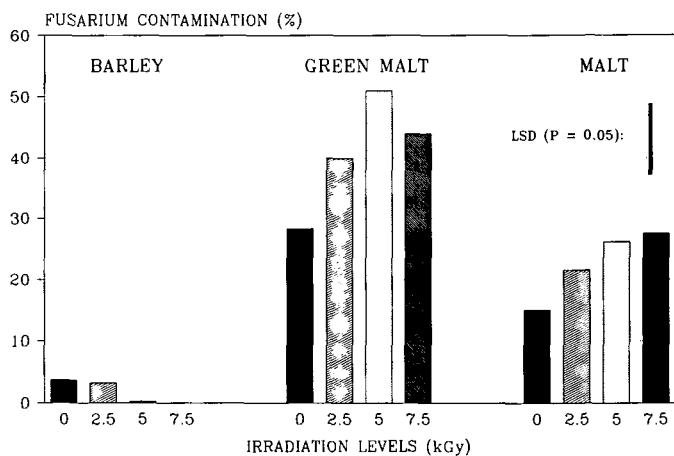


Fig. 2. Effect of <sup>60</sup>Co irradiation on the percentages of Clipper barley, green malt, and malt kernels contaminated with *Fusarium*.

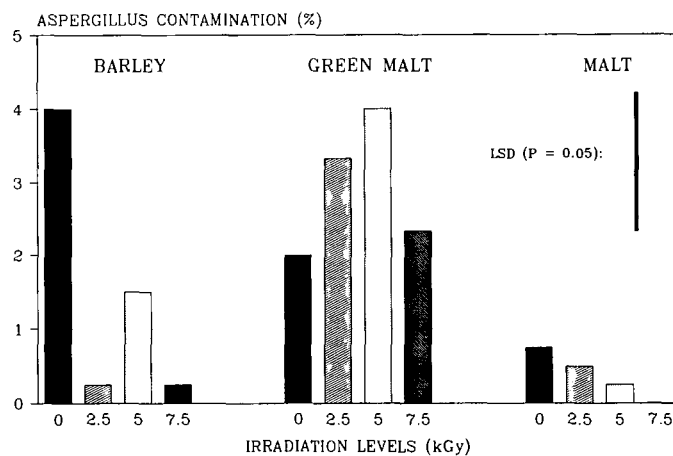


Fig. 4. Effect of <sup>60</sup>Co irradiation on the percentages of Clipper barley, green malt, and malt kernels contaminated with *Aspergillus*.

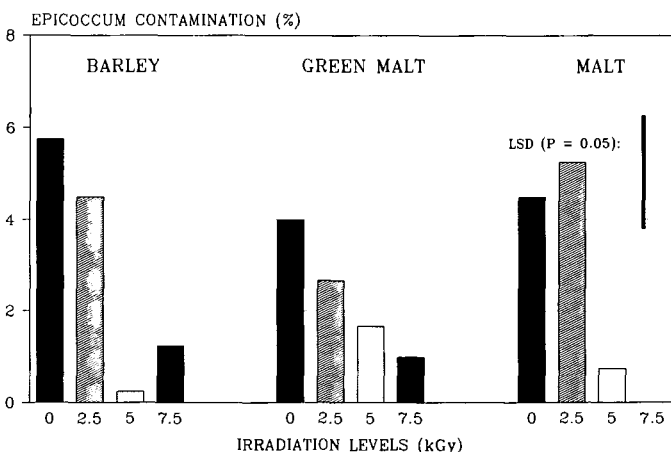


Fig. 3. Effect of <sup>60</sup>Co irradiation on the percentages of Clipper barley, green malt, and malt kernels contaminated with *Epicoccum*.

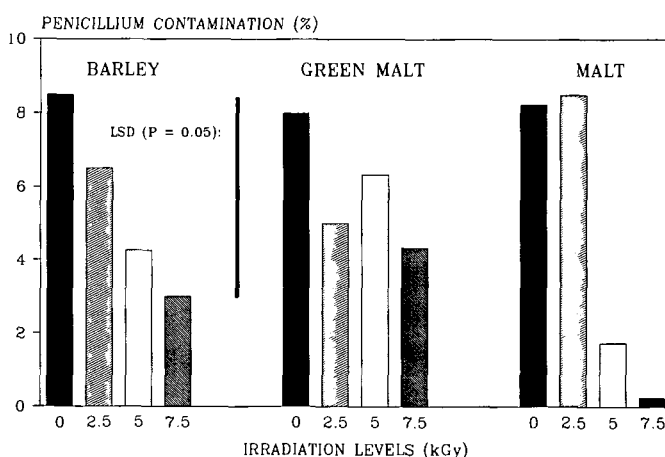


Fig. 5. Effect of <sup>60</sup>Co irradiation on the percentages of Clipper barley, green malt, and malt kernels contaminated with *Penicillium*.

an increase in *Aspergillus* during steeping, germination, and mild kilning (50°C). Douglas and Flannigan (10) noted little change in levels of *Aspergillus* during steeping and germination, whereas kilning at 68–75°C resulted in a marked decrease in the percentage of kernels contaminated with *Aspergillus*.

An almost linear decrease in the percentage of barley kernels contaminated with *Penicillium* was observed with increasing levels of <sup>60</sup>Co irradiation. No significant changes in levels of *Penicillium* were found on green malt (Fig. 5), whereas kilning resulted in a marked reduction in levels of *Penicillium* on malt irradiated with 5.0- and 7.5-kGy doses of <sup>60</sup>Co.

### Germination Studies

The effects of different levels of <sup>60</sup>Co irradiation on the germinative energy of barley samples (72-hr values), determined over a period of 12 months, are presented in Table I.

TABLE I  
Effect of Cobalt Irradiation on Clipper Barley  
and Malt Quality Parameters

Parameter	Months After Harvest	Radiation Dosage, kGy <sup>a</sup>				LSD <sup>b</sup>
		0.0	2.5	5.0	7.5	
Germination energy 4-ml test, %	3	99.5	99.5	99.0	97.0	1.96
	6	100.0	99.8	98.8	97.0	0.96
	9	99.8	99.5	99.8	96.2	2.42
	12	100.0	99.8	98.2	94.0	2.03
8-ml test, %	3	71.2	65.8	41.8	23.2	4.83
	6	90.0	94.0	79.0	57.8	8.05
	9	96.5	95.5	89.0	72.8	5.62
	12	92.8	89.5	74.2	35.5	8.53
Fine grind extract, %, w/w	3	79.2	78.8	78.0	78.1	0.33
	6	79.5	79.6	78.2	78.0	0.29
	9	...	...	78.2	78.3	0.27
	12	80.1	80.2	78.6	78.3	0.67
Color, EBC units	3	3.3	3.3	3.9	4.1	0.08
	6	3.2	3.2	3.9	4.2	0.08
	9	3.3	3.3	3.9	4.2	0.12
	12	3.6	3.4	4.0	4.2	0.18
Total soluble nitrogen, %, w/v	3	0.66	0.60	0.62	0.66	0.017
	6	0.75	0.67	0.64	0.67	0.019
	9	0.75	0.65	0.64	0.67	0.017
	12	0.82	0.70	0.68	0.69	0.023
Free amino nitrogen, mg/L	3	167.0	143.0	134.9	144.8	5.21
	6	186.1	149.6	142.9	142.4	3.92
	9	193.1	155.3	135.8	144.6	4.86
	12	218.9	167.1	151.4	154.0	4.74
α-Amylase, dextrinizing units per gram	3	40.0	17.1	13.9	7.5	3.55
	6	35.0	19.6	11.2	6.5	3.62
	9	50.2	28.8	14.9	13.5	2.59
	12	58.0	30.0	18.8	12.2	4.86
β-Amylase, Windisch- Kolbach units	3	207.1	173.1	151.4	157.0	10.94
	6	163.2	143.4	130.4	126.4	8.21
	9	208.4	178.8	176.0	160.4	6.69
	12	211.5	172.4	153.2	147.0	7.32
Viscosity, cP	3	1.51	1.57	1.84	1.91	0.109
	6	1.52	1.65	1.82	1.83	0.036
	9	1.53	1.60	1.71	1.74	0.024
	12	1.59	1.58	1.73	1.80	0.044

<sup>a</sup>Results are average of four repeats.

<sup>b</sup>P = 0.05.

Although the 7.5-kGy dose of <sup>60</sup>Co irradiation significantly lowered the germination energy values (4-ml test), the decrease was, on average, only three percentage points. The 8-ml germination energy test showed greater sensitivity to <sup>60</sup>Co irradiation. In most cases, the 5.0-kGy doses of <sup>60</sup>Co irradiation resulted in a significant reduction in 8-ml germination energy values, whereas the 7.5-kGy doses of <sup>60</sup>Co irradiation lowered these counts further by approximately 25–40 percentage points, compared with the controls.

Differences in the germination rate, as a result of different doses of <sup>60</sup>Co irradiation, also have been observed in other seeds. Progressive decreases were found in germination rates of wheat (31), as well as in other seeds treated with doses of <sup>60</sup>Co irradiation as low as 0.1 kGy (12,20). Not all researchers have found a decrease in seed germination when low levels of <sup>60</sup>Co irradiation were applied. Levels of up to 0.1 kGy have, in fact, been found to stimulate root and shoot growth and increase the number of tillers per plant (19–21,30,32).

### Malt Quality

The effect on the quality of malt produced from barley irradiated with different levels of <sup>60</sup>Co determined over a period of 12 months is presented in Table I.

No significant differences in Congress mash fine-grind extract levels were observed between the controls and the barley irradiated with a 2.5-kGy dose of <sup>60</sup>Co. On the other hand, extract levels of malt produced from barley irradiated with 5.0- and 7.5-kGy doses of <sup>60</sup>Co were significantly lower than those of the controls. These differences remained consistent throughout the test period.

No significant differences in wort color were measured between the control samples and those irradiated with a 2.5-kGy dose of <sup>60</sup>Co. Wort colors of samples irradiated with 5.0- and 7.5-kGy doses of <sup>60</sup>Co were approximately 0.6 and 0.8 EBC units higher, respectively, than the controls. In both cases, the differences were significant.

All of the doses of <sup>60</sup>Co irradiation had the effect of significantly lowering wort soluble nitrogen as well as free amino nitrogen levels. Although significant differences were observed between the treated samples, the differences were not consistent.

Similarly, even the lowest level of <sup>60</sup>Co irradiation had an effect on significantly lowering α- and β-amylase levels, compared with the controls. Higher radiation doses lowered the amylase levels further, in some cases significantly, when compared with the previous dose rate.

Wort viscosities increased significantly with increasing levels of radiation (up to 5.0 kGy). A 7.5-kGy dose of <sup>60</sup>Co resulted in a slight increase in viscosity levels when compared with samples irradiated with a 5.0-kGy dose of <sup>60</sup>Co.

For barley to be successfully converted into malt during the malting process, adequate amounts of gibberellins have to move from the embryo and scutellum to trigger production of enzymes in the aleurone layer (6). Any interference with the production and secretion of the gibberellins will, therefore, result in reduced production of enzymes necessary for the solubilization of the multicellular tissue of the endosperm. Machaiah et al (23) found a reduction in the rate of gibberellic acid biosynthesis in gamma-irradiated wheat seed during germination. Their results indicated that radiation treatment interfered with the interconversion of a less active gibberellic acid to a highly active one, by impairing the activity of mevalonate kinase and adenosinetriphosphatase.

In this study, the observed reduction of extract, soluble nitrogen, and free amino nitrogen levels, as well as increased viscosity levels, indicate that the activity of enzymes involved in cytolytic and hydrolytic activity has been reduced. This may be either by direct interference with its biosynthesis at DNA level or as a result of reduction in the rate of gibberellin biosynthesis. The decreased amylase activity found in this study agrees with the findings of Moustafa and El-Shafey (27), Avtar et al (2), and Khanna and Meharchandani (21). The addition of gibberellic acid to irradiated,

germinating barley alleviates to a certain extent the irradiation effect (21). Low levels of  $^{60}\text{Co}$  irradiation (0.08 kGy and lower) were found to increase  $\alpha$ -amylase activity in wheat and tomato seedlings (1,32).

### CONCLUSIONS

This work indicates that resistance to irradiation differs among fungi. Malting of partially or even completely sterilized grain in a contaminated environment might therefore result in the rapid increase in more resistant strains, such as *Fusarium* spp.

It also can be concluded that most malt parameters measured are sensitive to  $^{60}\text{Co}$  irradiation and that the measurement of germinative energy of irradiated barley is too insensitive to predict the effect of irradiation on the final malt produced from irradiated barley.

Further experimental work should, however, be performed to ascertain the effects of low doses of irradiation on malt quality. Reports on the stimulating effect of low doses of  $^{60}\text{Co}$  irradiation on seedling growth and enzyme activity and the sensitivity of some grain insects and fungi (such as *Aspergillus* spp.) to low doses of irradiation indicate that, under certain given circumstances, the quality of malt may be improved as a result of improved storage conditions and germination.

Finally, the costs of irradiation as well as the availability of facilities to perform irradiation on a large scale should be taken into account. Presently, depending on the dose rate, the cost of irradiation could be as high as 50% of the purchase cost of barley. The high cost of irradiation, the lack of facilities to irradiate large quantities of barley in South Africa, and the lack of sufficient data on the effect of low dose irradiation on malt quality presently rule this practice out as a viable possibility.

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