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CHEMICAL CHANGES IN MEAT INDUCED BY GAMMA IRRADIATION

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Before ionizing radiations can be used extensively for food preservation, it must be conclusively established that spoilage and pathogenic microorganisms are killed or inhibited, the wholesomeness and nutritional values are not adversely affected, and no extensive objectionable changes in palatability are induced by the process. Information summarized in several recent reviews on the microbiological aspects of radiation preservation of foods (1,2,3) indicates clearly that proper application of ionizing radiations can destroy or inhibit pathogenic and food spoilage microorganisms. A recent resumé of studies on the wholesomeness of irradiated foods (4) shows that irradiated foods are wholesome and nutritious. Although many foods can be subjected to large doses of ionizing radiations without objectionable effects on color, odor or flavor (5), the appearance and palatability of other foods are adverseley affected by sterilizing dosages of ionizing radiation. This is particularly true of the high protein foods such as meat and milk. To establish objective methods for following the changes that occur when these foods are irradiated for preservation, and to devise intelligent methods for preventing these undesirable changes, the chemical reactions responsible for the changes must be elucidated.

We have continued to investigate the chemical changes induced by gamma irradiation of meat, meat fats, and meat pigments. This paper summarizes results we have obtained since those reported in the Proceedings of the First International Conference on the Peaceful Uses of Atomic Energy (6).

MATERIALS AND METHODS

Irradiation of meat and other materials for study was done in an experimental Cobalt 60 source with a radiation intensity of about 300,000 roentgens per hour, or in a more intense gamma source at the Argonne National Laboratory in Lemont, Illinois. In the Cobalt 60 source, products were irradiated at approximately 5°C., and in the more intense gamma source at ambient temperatures (approximately 22°C.).

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The meat used was raw ground beef muscle, unless otherwise indicated. Details on the fats and pigment preparations irradiated, and on the chemical methods used, are given with the results or have been presented in other reports from our laboratories.

FACTORS INFLUENCING CHEMICAL CHANGES INDUCED IN MEAT BY GAMMA IRRADIATION

Batzer and Doty (7) reported that when ground lean beef was irradiated at 5×10^5 rep or above, there was an appreciable reduction in soluble protein, an increase in non-protein nitrogen compounds, a reduction in glutathione, and the formation of hydrogen sulfide and methyl mercaptan. Since then, Herk *et al.* (8), using gas partition chromatography and mass spectrometry techniques, found many other simple sulfur compounds and carbonyls in irradiated meat. Batzer *et al.* (9) found that carbonyl compounds increased in both meat and meat fat with increasing irradiation dosages. Absorption spectra of the 2,4-dinitrophenylhydrazine derivatives of carbonyls extracted by benzene, and by an acid-salt mixture from meat and meat fat, showed the presence of several different carbonyl compounds produced by irradiation. In addition to these changes and the increase in pH as the result of gamma irradiation of meat (6), the glycogen content of meat is reduced by irradiation (Table I).

Table I. The Influence of Irradiation on the Glycogen Content of Ground Beef Round

Irradiation Dosage X 10^6 rads	Glycogen[a] mg./g. meat	
	Sample A	Sample B
0.00	0.59	0.57
0.25	--	0.53
0.50	0.47	0.47
0.75	--	0.44
1.00	--	0.44
1.50	--	0.38
2.00	0.26	0.33
4.00	0.17	--
8.00	0.32	--

[a]Determined by the method of Carroll *et al.* (10).

To determine the effect of fatness of the meat, and various pre- and post-irradiation treatments of the meat, on these changes, we have made a rather extensive study of the influence of pre-irradiation storage temperature, irradiation dosage, and post-irradiation storage temperature on beef from carcasses of different grades. The data (Table II) show that increase in pH, hydrogen sulfide, methyl mercaptan and acid-salt soluble carbonyls, and decrease in glutathione and glycogen in fresh raw muscle varies directly with irradiation dosage. However, the extent of these changes varied somewhat from sample to sample, depending in part on carcass grade and/or amount of intramuscular fat present.

Table II. Influence of Gamma Irradiation on Some Compounds in Ground Raw Longissimus dorsi Muscle from Carcasses of Different Grades 72 Hours After Slaughter

Carcass Grade	Carcass No.	Fat %	Irrad. Dosage	pH	H_2S[a]	CH_3SH[b]	Glutathione (total)	Acid-Salt Soluble Carbonyls[c]	Glycogen
			megarad		γ/g.	γ/g.	mg./100 g.	10^{-5}M/g.	mg./g.
U.S. Prime	1	9.3	0	5.42	0.05	0.08	16.50	2.62	0.59
			0.5	5.42	0.47	0.38	12.25	4.45	0.47
			2	5.48	1.49	0.69	6.25	9.43	0.26
			4	5.56	2.12	1.18	5.38	15.64	0.17
			8	5.80	2.84	2.04	5.75	23.30	0.32
	2	10.2	0	5.45	0.00	0.00	17.13	5.02	0.16
			0.5	5.45	0.16	0.30	15.25	6.10	0.13
			2	5.58	0.86	0.27	11.25	9.84	0.00
			4	5.60	1.75	1.18	8.75	14.49	0.00
			8	5.72	1.79	1.34	8.13	20.93	0.00
U.S. Good	3	4.2	0	5.48	0.15	0.49	17.25	3.54	0.42
			0.5	5.51	0.22	0.66	16.13	6.04	0.31
			2	5.58	1.07	1.25	10.00	13.12	0.22
			4	5.61	2.28	2.11	8.25	16.62	0.11
			8	5.70	3.21	3.19	6.25	21.10	0.09
	4	4.5	0	5.50	0.12	0.00	25.63	1.76	0.00
			0.5	5.43	0.08	0.10	19.00	4.46	0.00
			2	5.50	1.00	0.25	13.50	9.26	0.00
			4	5.59	1.79	0.68	12.00	12.88	0.00
			8	5.70	2.58	1.52	10.25	20.18	0.00
U.S. Utility	5	2.8	0	5.50	0.15	0.06	10.75	4.72	0.55
			0.5	5.52	0.50	0.21	9.13	7.13	0.44
			2	5.55	1.07	0.77	6.25	14.22	0.28
			4	5.62	1.56	1.84	4.38	14.51	0.17
			8	5.80	3.81	2.74	2.88	20.88	0.15
	6	6.4	0	5.45	0.00	0.33	16.13	3.88	0.44
			0.5	5.45	0.19	0.48	13.13	5.45	0.42
			2	5.50	0.65	0.88	9.50	10.73	0.26
			4	5.55	1.20	1.41	8.75	16.97	0.17
			8	5.65	2.05	2.08	7.50	23.46	0.13

[a]Determined as described by Marbach and Doty (11).

[b]Determined as described by Sliwinski and Doty (12).

[c]Extracted and determined as described by Batzer *et al.* (9).

The influence of pre-irradiation storage of beef (aging) at different temperatures on the chemical changes induced by irradiation was related to carcass grade and aging temperature (Table III).

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Table III. Influence of Pre-Irradiation Aging for Three Weeks at Different Temperatures on Chemical Changes Induced by Irradiation of Ground Raw *Longissimus dorsi* Muscle from Carcasses of Different Grades

Carcass Grade and No.	Aging Temp. °F.	Irradiation Dosage		pH	H_2S[a]	CH_3SH[b]	Acid-Salt Soluble Carbonyls[c]	Glutathione	Glycogen
		megarad			γ/g.	γ/g.	10^{-5}M/g.	mg./100 g.	mg./g.
Prime (1)	0	4	Unaged	5.56	2.12	1.18	15.64	5.38	.17
			Aged	5.60	2.36	0.71	14.51	6.25	.08
	35	8	Unaged	5.80	2.84	2.04	23.30	5.75	.32
			Aged	5.85	2.74	1.47	20.88	5.38	.15
	45	2	Unaged	5.48	1.49	0.69	9.43	6.25	.26
			Aged	5.80	0.86	0.55	14.22	11.25	.04
Good (3)	0	8	Unaged	5.70	3.21	3.19	21.10	6.25	.09
			Aged	5.72	3.36	4.09	21.86	8.25	.04
	35	2	Unaged	5.58	1.07	1.25	13.12	10.00	.22
			Aged	5.70	1.05	1.56	15.64	11.00	.05
	45	4	Unaged	5.61	2.28	2.11	16.62	8.25	.11
			Aged	5.70	1.43	2.44	16.10	8.25	.04
Utility (5)	0	2	Unaged	5.55	1.07	0.77	14.22	6.25	.28
			Aged	5.66	1.40	1.03	10.76	9.38	.08
	35	4	Unaged	5.62	1.56	1.84	14.51	4.38	.17
			Aged	5.87	2.66	2.21	17.26	7.50	.24
	45	8	Unaged	5.80	3.81	2.74	20.88	2.88	.15
			Aged	5.95	3.36	3.34	22.42	6.88	.10

[a]Determined as described by Marbach and Doty (11).

[b]Determined as described by Sliwinski and Doty (12).

[c]Extracted and determined as described by Batzer *et al.* (9).

Aged irradiated beef had a slightly higher pH and less glycogen than unaged beef, regardless of grade or irradiation dosage. Aged irradiated U.S. Prime grade beef contained less hydrogen sulfide, methyl mercaptan, glutathione and carbonyls than unaged irradiated beef; aged irradiated U.S. Good grade beef contained more methyl mercaptan, less hydrogen sulfide, and about the same percentage of carbonyls and glutathione as unaged irradiated beef; and aged irradiated U.S. Utility grade beef contained more of all the compounds determined than unaged irradiated beef of the same grade. In general, the effects of aging were more pronounced at higher aging temperatures. When unaged irradiated ground beef was stored at 35°F., 60°F., and 90°F. for three months, it was found that the higher the storage temperature the higher the pH, the lower the hydrogen sulfide, and the lower the carbonyls in all grades at all irradiation dosage levels. At high irradiation dosage levels, smaller amounts of methyl mercaptan were present after three months' post-irradiation storage at 90°F. than at lower storage temperatures for all grades of meat. Glycogen was absent from all stored irradiated samples. The effect of post-irradiation storage temperature on glutathione content was inconsistent and apparently dependent on grade and irradiation dosage. In general, aged irradiated samples exhibited these same qualitative changes during post-irradiation storage at different temperatures.

If the development of hydrogen sulfide, methyl mercaptan, and acid-salt soluble carbonyls in irradiated meat is directly related to the undesirable odor; then one could postulate from the data presented above that raw beef from high grade carcasses, aged at 45°F., irradiated, and then stored at high temperature should be more acceptable than irradiated meat of lower grades of beef treated less drastically. However, other odors and flavors resulting from protein degradation induced by the high storage temperature might well be more objectionable than the odors and flavors normally induced by irradiation.

CARBONYLS PRODUCED BY IRRADIATION OF MEAT AND MEAT FATS

Although it is unlikely that carbonyls and other compounds produced by irradiation of fat contribute directly to the off-odors of irradiated meat, they may be related indirectly to the odor of irradiated meat. Since a high proportion of the volatile carbonyl compounds produced by irradiation of meat are α-β unsaturated (13), condensation products of these compounds with mercaptans could produce sulfur-containing compounds, such as methional, with very distinctive odors. Witting and Batzer (14) reported that crude reaction mixtures of methional had an odor typical of ground beef that had received 2-4 megarad of gamma irradiation, while the reaction mixture containing 3-methylthiobutyraldehyde had an odor typical of meat irradiated at higher dosages. Although freshly distilled thioaldehydes were odorless, offensive odors developed in a few minutes, presumably as the result of air oxidation, possibly to the sulfoxide. Patton and Keeney (15), and Day, Keeney and Stahl (16) have been unable to confirm the odorless nature of methional, and have questioned the validity of the results reported by Witting and Batzer.

In addition to three unsaturated aldehydes, Witting and Schweigert (13) characterized seven saturated aldehydes from the volatile fraction from gamma irradiated meat. At least three dicarbonyl compounds were isolated but not completely characterized. The presence of amino acids, especially cystine and

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cysteine, in lard during irradiation affected the quantity and type of carbonyls formed. This, together with the results reported in other studies (9,14), supports the suggestion that carbonyl compounds may indirectly influence the odor of irradiated meat through possible condensation reactions with sulfur compounds.

MYOGLOBIN CHANGES INDUCED BY IRRADIATION

Since the color of raw meat is one of the important factors influencing its acceptability, any undesirable color changes induced by gamma irradiation must be kept at a minimum. Ginger et al. (17) found that gamma irradiation of crude myoglobin extracts resulted in the formation of a green compound with a light absorption maximum at 610-620 mμ. The actual visual appearance of irradiated raw beef depended somewhat on the presence of oxygen during irradiation and was apparently dependent upon the relative amounts of metmyoglobin, oxymyoglobin, and green pigment present. More recent work by Fox et al. (18) has shown that the the/green pigment with a light absorption maximum at 616 mμ is sulfmyoglobin. This compound is apparently an intermediate in the destruction of the pigment of muscle or muscle extracts during irradiation. It may be decomposed slowly by either reducing or mild oxidizing conditions to compounds which resemble either myoglobin or metmyoglobin, respectively. Spectrophotometric evidence indicates that the red pigment produced by the gamma irradiation of raw meat or meat extracts is separate and distinct from oxymyoglobin (19), although a previous report by Tappel (20) suggested that the bright red pigment in irradiated meat is oxymyoglobin.

SUMMARY

During the gamma irradiation of raw beef, the pH, hydrogen sulfide, methyl mercaptan, and carbonyls increase; glutathione and glycogen decrease; and changes occur in the chemical nature of the pigments present. The nature and extent of most of these chemical changes are influenced by the fat content of the meat, the time and temperature of storage prior to irradiation, the irradiation dosage, and the storage time and temperature after irradiation.

The exact relationship of these chemical changes to changes in odor and flavour of cooked meat has not been established. However, these chemical changes can be measured objectively, and would in many cases logically be expected to influence odor and flavor.

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