EFFECT OF PRE-IRRADIATION HEATING ON THE FLAVOR AND NITROGENOUS CONSTITUENTS OF BEEF DURING STORAGE

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EFFECT OF PRE-IRRADIATION HEATING ON THE FLAVOR AND NITROGENOUS CONSTITUENTS OF BEEF DURING STORAGE

INTRODUCTION

With the recognition of the potentialities of ionizing radiations as a means of effecting preservation in foods, a new era was opened in food research. It has been proven that these radiations were highly efficient in the inactivation of microorganisms and other forms of life. Their ability to penetrate considerable thicknesses of material promised the possibility of treating foods in metal, glass, and other types of containers. The fact that they produced only a negligible rise in temperature has attracted attention to the possibility of preserving in raw state many foods, the preservation of which depended on severe heat treatments. The method also promised a decreased requirement for refrigeration equipment which was very attractive from a logistical point of view.

As research went on, however, it was found that these radiations brought about very undesirable side effects. Unpleasant flavors and odors were produced in foods subjected to these radiations. It was shown that the intensity of these off-flavors and odors was dose-dependent. It was also observed that irradiation to levels ordinarily used for sterilization did not result in the complete inactivation of enzymes. This was particularly true for meats irradiated in a raw state. This suggested that enzymatic breakdown would continue during storage. Consequently, cooking prior to irradiation would be a logical approach in preventing enzymatic deterioration during storage. A tendency towards more intensified off-flavors upon irradiation of partially or completely cooked meat was reported (46, p. 238), with the effect of storage not being investigated. In another report (7, p. 605), beneficial effects of long term storage on flavor of pre-cooked meat was observed, the pre-cooked irradiated meat stored at 72° F. being not distinguishable from unirradiated control at the end of 250 days of storage while the reverse was true at the beginning of storage. Would this be true for lower pre-heating temperatures, where the final product would partly retain the qualities of uncooked meat? This was an attractive question.

Consequently, the present investigation was undertaken to determine the pattern of changes in flavor and in nitrogenous constituents, to serve as an index of proteolytic activity, of beef during prolonged storage at room temperature, with a view towards establishing the minimum pre-heating temperature and/or the maximum storage period to yield a product the flavor of which is not different from beef heated to cooking temperatures before irradiation.

REVIEW OF LITERATURE

Ionizing Radiations

Ionizing radiations have the characteristic property of being able to energize individual atoms or molecules of a medium to such an extent that an orbital electron is ejected from the molecule and a positively charged ion remains. Hannan (18, p. 4) classified some of the more important ionizing radiations into two categories, one being the electromagnetic waves, the second the particulate radiations. Electromagnetic waves include x-rays and gamma rays which are characterized by their deep penetration. Particulate radiations consist of negatively or positively charged or neutral particles. For detailed discussions of these radiations the reader is referred to the reviews by Hannan (18, p. 4-27) and by The United States Army Quartermaster Corps (50, p. 43-80).

Effect of Ionizing Radiations on Microorganisms. The importance of these radiations from food preservation point of view lies in their effects on the biological systems. The most important of these is the capability of these radiations to produce bactericidal or bacteriostatic effects on microorganisms. Dunn, *et al*. (14, p. 605) showed that spoilage microorganisms in many foods could be killed by ionizing radiations at dosage levels of 10^5 to 2×10^6 reps. Morgan and Reed (34, p. 357) investigated the resistance of bacterial spores to gamma irradiation and concluded that spores of *Cl. botulinum* appeared to be more resistant to radiation than spores of several other food spoilage organisms. Morgan (32, p. 24) reported that a dosage level of 4.8 megarads was required to kill the spores of Cl. botulinum. However, Pratt et al. (40, p. 60) inoculated different food products with spores of Cl. botulinum and concluded that no single dose appeared to be best for all products since resistance of spores was different in different foods. Denny and Bohrer (11, p. 49) reported that Cl. botulinum was more resistant to radiation in foods than in neutral phosphate when irradiated at freezing temperatures. In a review, Shea (47, p. 10) stated that the requirements for sterilization has been between 3 to 6 megarads.

Microorganisms resistant to gamma irradiation have been encountered. Anderson et al. (1, p. 577) reported that a micrococcus isolated from ground beef and pork was resistant to extreme dosages of 6 megarads of gamma irradiation.

Effects of Ionizing Radiations on Flavor. Along with the desirable effects on the microflora of foods, however, offensive odors and flavors were also produced when foods were subjected to ionizing radiations. Cain et al. (7, p. 605) reported that tasters were able to differentiate between the non-irradiated and 1.9 megarad levels in beef steak, pork roasts and chops and in bacon prior to storage. Schultz et al. (46, p. 238) concluded that a dosage of only 124,200 rep might produce a flavor different from that of non-irradiated ground beef. Batzer and Doty (3, p. 66) showed that sulfur containing compounds contributed to some of the off-odors that develop in meat during irradiation. The results obtained by these authors indicated that hydrogen sulfide was one of the components and methyl mercaptan another. Batzer et al. (4, p. 701) reported that an increase in carbonyl compounds occurred on irradiation and this increase was directly proportional to the irradiation dosages employed. The same authors also stated that these carbonyl compounds probably did not directly contribute to the off-odors in irradiated beef and possibly had a role in decreasing the apparent off-odors by reacting with the sulfhydryl compounds and amines that did contribute. Pearson et al. (36, p. 234) showed that hydrogen sulfide, methyl mercaptan and carbonyls were responsible for a considerable part of the poor acceptability of irradiated beef, pork, and veal. Marlbach and Doty (27, p. 884) made quantitive determinations of hydrogen sulfide produced and found that 1 to 4 gamma of hydrogen sulfide per gram of meat was formed from the volatile components of ground beef that had been irradiated at a dosage level of 2 million to 4 million reps whereas only half of this amount was released after irradiation at 10 million rep. The same authors also found that, in beef frozen and stored at 0° F. before irradiation, the amount of hydrogen sulfide decreased with storage time for at least two weeks and that less hydrogen sulfide was released at the same dosage level from ground beef of high fat content than from beef containing less fat. LaFuente et al. (24, p. 275) compared different radiations with regard to their production of off-flavors in milk and concluded that the intensity of off-flavors produced by ultraviolet radiation, gamma rays, and cathode rays was the same at the radiation levels required to successfully destroy the micro-organisms present in raw whole milk.

Not all foods reacted in the same way in the production of off-odors and flavors upon irradiation. Pearson et al. (37, p. 619) showed that in pre-cooked meat irradiated at 2.79 megarads, pork showed little or no adverse effects from radiation whereas beef, chicken and veal were markedly less acceptable.

It was also observed that different dosage levels produced different intensities of off-flavors. Cain et al. (7, p. 605) found that significant differences in flavor between the radiation levels of 1.9 and 2.8 megarads were detected by tasters in beef and pork roasts and in pork chops prior to storage. McGill et al. (30, p. 80) reported that the rate of rancidity development appeared to be proportional to the radiation dose and that the taste panel was unable to distinguish between radiated and unirradiated samples of chicken meat when dosage levels of 1×10^5 and 5×10^5 rep were used. Schultz et al. (46, p. 238) reported that irradiation dosages from 124,200 to 993,600 rep showed a linear relationship between irradiation dosage and intensity of irradiation flavor of ground beef.

Several methods were tested to minimize the off-flavors produced in meats on irradiation. Goldblith and Proctor (17, p. 254) pointed out the potentiality of three methods, individually or in combination, in reducing undesirable side reactions. These were (1) irradiation in the frozen state, (2) removal of oxygen, and (3) addition of free radical acceptors. Tausig and Drake (49, p. 226) reported that activated carbon was effective in reducing the intensity of irradiation odor and flavor. Pearson et al. (37, p. 619) showed that presence or absence of oxygen scavenger did not significantly influence acceptability, although there was a trend for added oxygen scavenger to result in higher mean scores. Cain and Anderson (6, p. 584) combined tetracycline antibiotic treatment and pasteurization dosages of irradiation and found that the antibiotic retained sufficient activity to offer protection to meats during storage, inferring that lower levels of radiation could be used and this would produce fewer adverse organaleptic changes than sterilizing levels. Schnautz (45, p. 36) investigated the effectiveness of certain spices in protecting beef from gaining flavors from gamma radiations and found that pepper was effective in masking irradiation flavor. Cain, Bubl, and Anderson (8, p. 540) found that ground beef exposed to 8 intermittent exposures of gamma radiations produced significantly less irradiation flavor than beef exposed to 1, 2, or 4 exposures. Schultz, Cain, and Nordan (46, p. 238) showed that there was no significant difference in the intensity of radiation flavor produced in meat canned in atmospheres of air, vacuum, or nitrogen, with frankfurters constituting a possible exception. They also reported that there was no significant difference in the intensity of radiation flavor between meats irradiated while frozen or unfrozen.

The effect of post-irradiation storage was also investigated although not very extensively. Pratt and Ecklund (39, p. 497) reported that in irradiated ground beef which had been in storage at 70° or 98° F. for nine months, the taste panel detected additional off-flavors and some liquid formation was noted. Both this additional flavor and liquid formation were reduced in ground beef pasteurized to 160° F. Drake et al. (13, p. 23) recorded the formation of tyrosine crystals in the storage of irradiation sterilized raw meat. Cain et al. (7, p. 609) reported the occurrence of extensive degradative changes during the nine month storage at 72° F. of irradiated fresh meats. Formation of tyrosine crystals and excessive fluid loss was also observed.

All of the above observations indicated action of the proteolytic enzymes during storage. Doty and Wachter (12, p. 63) observed that there was little, if any, inactivation of the proteinase in beef by irradiation at a dosage of 0.5×10^6 rep. At a higher dosage level of 1.6×10^6, there was approximately 50 percent loss in apparent activity in some samples. Drake et al. (43, p. 23) confirmed that the principal proteolytic enzymes present in beef were cathepsins and found that the rate of enzyme activity was accelerated at the higher storage temperatures. The radiation dosages required for the inhibition of enzymes were higher than those required for killing the microorganisms.

The fact that the proteolytic activity in meats continued during post-irradiation storage led the investigators to combining heat treatment with irradiation. This was suggested by Morgan and Reed (34, p. 366). Kan, Goldblith, and Proctor (23, p. 518) suggested that a combination of heat and radiation might reduce off-flavor production due to radiation doses, and serve, additionally, in the inactivation of radio resistant enzymes. Schultz et al. (46, p. 238), however, showed that combining partial or complete pre-cooking with the irradiation did not reduce the production of radiation flavor and it might actually intensity it. Other investigators (7, p. 609) showed that beef cooked to 160° F. and pork cooked to 170° F. prior to irradiation would not undergo degradative changes during 250 days storage at 72° F.

Effect of Heat on Meats

Morgan and Kern (33, p. 373) reported that heating lowered the biological value of beef proteins. It was found by these investigators that the biological value of beef was inversely proportional to the severity of heating. This was also true in the case of fish meals. Daniel and McCollum (10, p. 18) concluded that the differences in heat treatments were responsible for the differences found in fish meals. Newton et al. (35, p. 589) reported a loss of water-soluble proteins which ranged from 50 to 70 percent when meat was heated from 185° to 248° F. This was confirmed by Ginger et al. (16, p. 410-416) who reported a 4- to 30-fold decrease in the soluble nitrogen of raw beef when heated.

This loss in biological value in proteins as a result of heating has been attributed to the destruction of vitamins and some essential amino acids (28, p. 602) in proteins during heating as well as to the loss of soluble proteins and decreased digestability (29, p. 1170-1171).

Effect of Irradiation on Proteins

The most evident effect of irradiation on proteins has been in the production of undesirable flavor and odor compounds which have been shown to contain sulfur (3, p. 66; 4, p. 701). This gives evidence to the sulfur linkage as the site of attack. Deamination of amino acids upon irradiation in pure aqueous solutions has been reported by Proctor and Bhatia (42, p. 537-538; 43, p. 2; 5, p. 552). These investigators showed that histidine was most highly effected, followed by cystine, phenylalanine, tyrosine, and tryptophan. Earlier investigations (41, p. 359) by the same authors had indicated that, at sterilizing doses of 2.25 megarads, the ten essential amino acids were not significantly destroyed in haddock fillets. It has been shown by Scheffner et al. (44, p. 460) that no significant destruction of the essential amino acids in milk, turkey, or beef protein occurred following gamma irradiation of 1.86 megarads.

This difference between the action of ionizing radiations on intact proteins and on pure amino acids was attributed to the proportionately larger amount of free radicals in a dilute solution (15, p. 16).

For extensive discussions of the effects on proteins, the reader is referred to the reviews of Hannan (18, p. 71-78) and by the United States Army Quartermaster Corps (50, p. 133-158).

Effect of Storage on Animal Proteins

An important change, as evidenced by the contraction of fibrils in the muscle fibers, takes place during the first few hours after the death of an animal. Moran and Smith (31, p. 43) explained this phenomenon as the stiffening and shortening of the muscle fibers by a post-mortem coagulation; that is, by a decreased solubility of the proteins within them. According to these authors, rigor disappears as the coagulated proteins are changed again into soluble forms. According to Szent-Gyorgyi (48, p. 72-82), the rigidity characteristic of rigor mortis is conferred on dead muscle as a result of formation of actomyosin by the combination of actin and myosin A upon post-mortem removal of potassium ions and ATP from myosin A. Huxley (22, p. 66) also stated that, as ATP is exhausted in the muscle region, the protein myosin seems to combine chemically with the protein actin. Bate-Smith (2, p. 10) stressed the importance of interfilamentary reactions in rigor, rather than intimate molecular processes, stating that the process of rigor mortis involves the association of the ultimate contractile filaments by weak cross-linkages which account for the decreased extensibility of the muscle. For detailed discussions of muscle contraction and the process of rigor mortis, the reader is referred to the references cited in this paragraph.

For the first 24-72 hours after the death of an animal, the muscle continues to take on a firmer characteristic. After rigor has fully set in, a tenderizing of the muscle is observed with time. Chen and Bradley (9, p. 164) and Hertzman and Bradley (19, p. 240) attributed the increase in tenderness after rigor primarily to autolysis. However, more recent work by Wierbicki et al. (51, p. 511) and Husaini et al. (21, p. 316) showed that the amount of autolysis in fresh beef during the first 12-15 days of storage at 3.5° C. appeared to be extremely small.

The effect on autolysis of higher storage temperatures and longer storage periods should be of interest to the food technologists of our time, because sterilizing doses of ionizing radiations do not effect complete inactivation of the proteolytic enzymes in meat (13, p. 23). From economical and military viewpoints, radio-sterilized meats would be preferred to be stored at temperatures higher than that of refrigeration, and storage periods would be expected to be long. Proteolytic activity in radio-sterilized meats has not been investigated to a great depth. There have been observations, however, that, during prolonged storage at room temperature and above, a fluid was exuded from irradiated fresh meats and from those pre-heated to temperatures at which enzyme inactivation had not been accomplished (39, p. 497; 13, p. 23; 7, p. 609).

With regard to the proteolytic changes in muscle under aseptic conditions, Zender et al. (52, p. 325) recently studied the natural aseptic and anaerobic degradation of rabbit and lamb muscle for 150 days at 25° C. and for 15 days at 38° C. The authors have observed a slow rise in the level of amino acids and a parallel decrease of glycine-soluble proteins. The electrophoretic distribution of muscle proteins was modified during storage as if proteins were first split into subunits and only later into amino acids. The exudate produced during 50-days storage amounted to 15 percent of the original muscle samples and contained around 7 gr. percent of proteins and 0.4 gr. percent of potassium.

EXPERIMENTAL METHODS

Sample Preparation

Ground lean beef, purchased in a single lot from a local meat packer, was thoroughly mixed and stuffed into cellulose casings approximately three inches in diameter. These were frozen at -18° F. in moving air and then mechanically sliced in cross section into portions 3/16 inch in thickness. The slices, still in frozen state, were completely randomized and placed in polyethylene-coated saran plastic bags. Two hundred forty such bags, each containing four slices of meat, were prepared and grouped into 24 units of ten bags each.

Heat Treatments

Pre-irradiation heating of samples to internal temperatures of 130°, 140°, 150°, 160°, and 170° F. was accomplished by employing a steam-heated water bath, the temperature of which was adjusted to and maintained at that of the desired internal temperature plus 10° F. During the heating process, each group of ten bags was handled as a unit and formed a replication of the particular level of the heat treatment used. A specially built rack, from which all the ten bags comprising a replication could be suspended, permitted the handling of all ten bags in one operation. The samples were thawed before being placed in heated water bath, removed from the bath as soon as the desired internal temperature had been attained, and immediately immersed in cold water for cooling. The periods of time required for the attainment of the internal temperatures of 130°, 140°, 150°, 160°, and 170° F. were 3.5-4.3, 3.7-4.2, 4.7-5.0, 3.5-5.2, and 5.2-6.0 minutes, respectively. The internal temperatures were obtained by thermocouples from representative slices in each group. The cooled slices were randomized within each group before they were placed into half pound flat (307x202) "C" enamel cans. The fluid released from the meat and collected in bags as a result of temperature treatment was discarded. The casings were stripped from the meat before the latter was placed in cans. Each can contained four slices of meat. The cans were vacuum sealed and their contents frozen at -18° F. in moving air.

Shipment

The samples, in frozen state, were packed under dry ice and shipped in insulated containers to the Materials Testing Reactor, Idaho Falls, Idaho. Eight days elapsed between shipment to and receipt from the irradiation site. Shipment from the site was made under wet ice conditions.

Irradiation

The cans were exposed to the gamma grid to attain a dose of 3 megarads. The dose rate was 2.92 megarads per hour. On this basis, 1 hour and 2 minutes were required for the attainment of the required dose.

Storage Conditions

Upon receipt of the samples from the Materials Testing Reactor, all of the cans were placed in storage at 72° F. The cans were rested on their sides rather than on bottoms and turned periodically in a uniform manner.

Chemical Analyses and Taste Tests

The samples, in four replications, were quantitatively analyzed for moisture, total nitrogen, total water-soluble nitrogen, trichloroacetic-acid-soluble nitrogen, and amino nitrogen.

Taste tests were conducted using a trained panel of eight tasters. The same tasters were used throughout the tests.

The chemical analyses and taste tests were conducted at the end of 0, 15, 30, 45, 60, 90, 120, 150, and 180 days of storage.

Manipulation of Samples for Chemical Analyses and Procedures for Chemical Determinations

At the end of each storage period, the cans containing the samples for that period were removed from storage and opened. Any exudate that might have collected in a can was removed and saved. Two of the four slices contained in each can were taken out, wrapped in aluminum foil, and placed in 0° F. storage for use in taste testing. Half of the exudate removed from the can was returned to the can and the other half discarded. The two slices in the can were thoroughly comminuted and mixed with a spatula to obtain a uniform sample for chemical determinations.

Moisture Determination

Approximately 5 gm. of meat were placed into tared aluminum pans and accurately weighed. The pans were then placed in an oven at 70° C. under 25-28 inches vacuum for 24 hours, removed and cooled in a desiccator, and reweighed. The loss in weight after proper calculations was reported as the moisture content.

Total Nitrogen Determination

Approximately 2 grams of meat was used for this determination. The method of Hiller, Van Slyke, and Plazin (20, p. 1402-1420) was followed using the indicator developed by Ma and Zuazaga (26, p. 280-282). Instead of using the catalysts suggested by Hiller et al. (20, p. 1402-1420), "Kel-Pak" catalysts containing 10 gm. potassium sulfate and 0.3 gm. copper sulfate per package were used. Total nitrogen content was reported on a percent dry weight basis.

Further Manipulation of the Samples for Total Water-Soluble Nitrogen, Trichloroacetic-acid-Soluble Nitrogen, and Amino Nitrogen Determinations

Thirty grams of meat were blended in a macro blender for three minutes with 75 ml. of distilled water at room temperature. The mixture was filtered through Whatman #12 paper and the filtrate was used for the following determinations in the manner described.

Total Water-Soluble Nitrogen Determination

Two mls. of the filtrate, 0.6 gms. "Kel-Pak" catalyst, and two ml. concentrated sulfate acid were dispensed into a 100 ml. Kjeldahl flask and heated. One hour was allowed for digestion. After cooling, 35 ml. distilled water, 10 ml. of 50% sodium hydroxide, a few Hengar granules, and two or three small pieces of mossy zinc were added and the entire mixture was heated after being connected to a distillation apparatus. The ammonia evolved was collected in 25 ml. of 4% boric acid solution. The same indicator as was used for the total nitrogen determination was used during the titration of the distillate with approximately 0.1 N sulfuric acid. Total water-soluble nitrogen content was reported on a percent dry weight basis.

Trichloroacetic-Acid-Soluble Nitrogen Determination

Ten ml. of 25% trichloroacetic acid was added to 10 ml. of the filtrate and the mixture filtered through Whatman #12 paper. Five ml. of this filtrate was used for the determination of trichloroacetic-acid-soluble nitrogen. The procedure followed was identical with that used for total water-soluble nitrogen, described in the preceding paragraph.

Amino Nitrogen Determination

The copper method of Pope and Stevens (38, p. 1070) was used for amino nitrogen determinations. Some minor modifications in the procedure were made as a result of a preliminary work designed to test the suitability of the method to the conditions of this experiment. A water extract of fresh ground beef and an aqueous solution of a mixture of pure amino acids were used in these tests. The values obtained from the water extract of ground beef were compared against those obtained from the Van Slyke method (20, p. 385) and those obtained from the mixture of amino acids were compared against the calculated amino nitrogen values. On the basis of the results obtained from the preliminary work, the following procedure was adopted for use in the experiment.

Reagents used were prepared in the same manner as described by Pope and Stevens (37, p. 1070). Twenty-five ml. of meat extract, prepared as explained on page 17 were pipetted into a 200 ml. volumetric flask. A sufficient quantity of normal sodium hydroxide was added from a buret drop by drop to make the contents of the flask slightly alkaline as indicated by a piece of Universal pH Indicator Paper used outside the flask. One hundred ml. of the copper phosphate suspension were added and the volume made to 200 ml. with distilled water. The content of flask was well mixed and filtered through a #5 Whatman paper. Twenty-five ml. of the filtrate were acidified with 5 ml. of glacial acetic acid and 5 ml. of 50% potassium iodide added. The solution was titrated with standardized sodium thiosulfate, one ml. of starch solution being added towards the end of titration. The titration was made under an atmosphere of nitrogen gas. The results were reported as a percentage based on the dry weight of meat, taking each ml. of 0.01 N sodium thiosulfate as being equivalent to 0.28 mg. of amino nitrogen.

Taste Tests

Taste Panel. The taste panel for this experiment consisted of eight experienced tasters. The same panel was used throughout the experiment.

Ballot. The ballot used for the experiment was designed in such a manner as to reflect the magnitude of flavor differences of experimental samples from a reference sample. The experimental samples were preheated to internal temperatures of 130°, 140°, 150°, 160°, and 170° F. The reference samples were preheated to 170° F. and in all other aspects were similar to the experimental samples. The ballot was a 7-point scale ballot with the reference sample at 4.0. The flavor characteristics evaluated by the panel were "Irradiation Flavor", "Bitterness", and "Over-all Desirability". The adjectival ratings printed on the ballot were assigned numerical scores in such a manner as to permit the indication of a more desirable flavor by a higher score. Accordingly, in the case of "Irradiation Flavor" or "Bitterness", the adjectival ratings "Much Less", "Moderately Less", "Slightly Less", "Same as Reference", "Slightly More", "Moderately More", and "Much More" appeared on the ballot along a vertical line and were assigned numerical scores of 7, 6, 5, 4, 3, 2, and 1, respectively. In the case of "Overall Desirability" the adjectival ratings "Much More Desirable", "Moderately More Desirable", "Slightly More Desirable", "Same as Reference", "Slightly Less Desirable", "Moderately Less Desirable", and "Much Less Desirable" appeared on the ballot along a vertical line and were assigned numerical scores of 7, 6, 5, 4, 3, 2, and 1 respectively. These phrases indicating adjectival ratings were placed one inch apart on the vertical line to permit interpolative evaluations by the tasters.

Safety Precaution. Before the samples were delivered to the kitchen, they were tested on mice for the presence of bacterial toxins. For this purpose, ten grams of meat were mixed with four fluid ounces of .3 percent sterile physiological saline solution and blended for two minutes in a sterile blender. About 10 ml. of this blended mixture was decanted into a sterile test tube and centrifuged in a high speed centrifuge for one hour. One-half ml. of the supernatant solution was injected into mice which were observed for 48 hours for symptoms of toxin action. At no phase of the experiment was there observed any evidence of toxin development.

Preparation of Samples for Serving to Taste Panel. Gas broilers were preheated and the slices of meat were placed on the broilers three inches from the source of heat. They were broiled 5-7 minutes on one side, turned and broiled on the other side for a similar period. This produced a sample which was cooked to a well-done stage. Preparation of samples for taste testing was done by the same persons in the same kitchen throughout the experiment.

Serving and Tasting of Samples. Samples of four replications of five different pre-irradiation heat treatments were served and tasted in four sessions at the end of each storage period. Two sessions were held a day, one being at 10 o'clock in the morning, the other at 3 o'clock in the afternoon. The four replications were served on two consecutive days. At the end of each storage period, all samples to be tasted were removed from 72° F. storage, individually wrapped in aluminum foil, placed in 0° F. storage, and kept frozen until the time when they were ready to thaw before delivery to the kitchen. After cooking, the samples were individually placed in randomly coded paper cups and served along with a reference sample which was marked as such. Tasting by the members of panel was carried out in individual booths, properly equipped.

Statistical Methods

The particular statistical methods employed for the analysis of the data obtained from this experiment will be indicated by making references to the text used (25).

RESULTS AND DISCUSSION

The results will be presented and discussed under three general headings which will cover the chemical analyses, flavor evaluations, and a correlation between the two.

Terminology

The term "pre-heating" as used in this text should be construed to mean "heating prior to irradiation".

The term "beef" refers to "ground lean beef irradiated to 3 megarads after being heated to a certain internal temperature".

Repetition of the facts that the experimental material has been irradiated to a dose of 3 megarads and stored at 72° F. has been avoided in the text wherever it interfered with the continuity of thought.

Results of Chemical Analyses

Before entering upon the presentation of the results pertaining to the three chemical attributes investigated, the difference in the moisture content of the samples that received different heat treatments prior to irradiation should be brought to the attention of the reader. Although the initial experimental material was uniform, different levels of heat treatment caused a release of different quantities of fluid from the samples. The fact that this fluid was discarded, produced samples of different moisture content.

Table 1 shows the moisture content of the samples at each level of heat treatment. The figures represent the averages of four replications analyzed at each storage period.

TABLE 1.
PERCENT MOISTURE CONTENT OF PRE-HEATED IRRADIATED BEEF

Heat Treatment ° F.	Storage Period (days) 0	15	30	45	60	90	120	150	180	Heat Treatment Mean
130	74.2	74.5	73.8	74.3	74.0	74.1	74.1	73.8	74.3	74.1
140	74.1	74.4	74.1	74.4	74.1	74.5	74.2	73.7	74.2	74.2
150	73.3	73.5	73.6	73.6	73.8	73.6	73.7	73.3	73.7	73.6
160	73.2	72.9	73.4	73.3	73.3	72.9	73.0	72.6	73.2	73.1
170	70.7	70.3	71.0	70.8	70.4	70.4	70.8	70.8	70.6	70.6

Considering the uniformity of the initial experimental material and the fact that the samples were randomized, no appreciable differences in moisture content within a given temperature treatment was expected. The figures in Table 1 suggest the same. The disproportionately large drop in moisture content of samples subjected to 170° F. heat treatment suggests a rapid protein denaturation at this temperature.

Total Water-Soluble Nitrogen

The data pertaining to the total water-soluble nitrogen content of ground lean beef heated to internal temperatures of 130°, 140°, 150°, 160°, or 170° F. prior to irradiation to a dose of 3.0 megarads and stored at 72° F. for 0, 15, 30, 45, 60, 90, 120, 150, or 180 days are presented in Table 2.

TABLE 2.
TOTAL SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF
AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENT AND LENGTH
OF STORAGE AT 72° F.
(Percent dry weight basis)

Heat Treatment ° F.	Storage Period (days)									Heat Treatment Mean
	0	15	30	45	60	90	120	150	180	
130	2.19	2.44	2.49	2.60	2.79	2.99	3.03	3.20	3.33	2.78
140	2.03	2.08	2.25	2.30	2.53	2.67	2.53	2.69	2.84	2.44
150	1.66	1.55	1.68	1.63	1.82	1.99	1.96	2.07	1.92	1.81
160	1.41	1.43	1.45	1.48	1.60	1.61	1.52	1.59	1.61	1.52
170	1.17	1.18	1.24	1.20	1.23	1.31	1.30	1.29	1.32	1.25
Storage Period Mean	1.69	1.74	1.82	1.84	1.99	2.11	2.07	2.17	2.20	

Effect of Heat Treatment and Storage Period.

The trend in the change in total water-soluble nitrogen content as affected by the two variables may be seen from Table 2. Heat treatment means show a steady reduction in this constituent with increase in pre-irradiation heating temperature. Likewise, as the storage period increases, the total soluble nitrogen content increases. However, definite answers in these and other pertinent respects, such as the relative magnitude of change contributed by each variable, significance of change at each level of heat treatment, linearity and rate of change, can be obtained only through statistical analyses. The following results were obtained through the application of appropriate statistical methods (25, p. 309-324), using the values representing the total water-soluble nitrogen content of replicate samples as individual observations.

TABLE 3.
ANALYSIS OF VARIANCE FOR TOTAL SOLUBLE NITROGEN IN IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F.

Variation Due to	Degrees of Freedom	Mean Square	F	Significance At the Level of 5%	1%
Total	179				
Replication	3	.0234	2.208	NS	NS
Temperature	4	14.6066	673.115	S	S
Temp. x Repl.	12	.0217	2.047	S	NS
Time	8	.7293	53.233	S	S
Time x Repl.	24	.0137	1.292	NS	NS
Time x Temp.	32	.0873	8.236	S	S
Error	96	.0106			

It may be seen from the values in Table 3 that the effect of pre-heating temperatures as well as that of the storage time was significant. The same was also true for the temperature x storage time interaction effect. The meaning of the significant interaction effect between the levels of heat treatment and the storage time is that no prediction can be made as to the total water-soluble nitrogen content for a particular combination of the level of heat treatment and storage period on the basis of the results obtained for other combinations.

The relative magnitude of each significant effect was calculated according to Li (25, p. 200-204) and can be seen from Table 4 which shows the extent of contribution by each effect towards variations in the total soluble nitrogen content of beef.

TABLE 4.
RELATIVE MAGNITUDE OF CONTRIBUTION BY SIGNIFICANT EFFECTS TOWARDS VARIATIONS IN TOTAL SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF

Effect	Variance
Temperature	.4054
Time	.0359
Temperature x Time	.0192

It may be seen from the values in Table 4 that the temperature to which the meat was heated prior to irradiation was the primary source of variation in the total soluble nitrogen content of stored beef. The effect of storage time was not as dominating and influential as that of the pre-heating temperature. The temperature x time interaction had the least effect of the three significant factors.

Significance of Change with Time within each Temperature Treatment. In order to determine whether the total water-soluble nitrogen content of beef heated to a particular temperature prior to irradiation changed significantly with time, data for each of the five levels of heat treatment were subjected to a separate analysis of variance (25, p. 196-208) the results of which are shown in Tables 5a through e. This information is important in that it will serve to indicate the temperature at which the factors responsible for the increase in total soluble nitrogen are inhibited or destroyed.

TABLE 5.
ANALYSIS OF VARIANCE FOR TOTAL SOLUBLE NITROGEN IN IRRADIATED BEEF AS AFFECTED BY LENGTH OF STORAGE PERIOD AT 72° F.

Variation Due to	Degrees of Freedom	Mean Square	F	Significance At the level of 5%	1%
a. Beef pre-heated to 130° F.					
Total	35				
Replication	3	.0175	3.125	S	NS
Storage time	8	.5812	103.785	S	S
Error	24	.0056			
b. Beef pre-heated to 140° F.					
Total	35				
Replication	3	.0530	3.174	S	NS
Storage time	8	.3207	19.203	S	S
Error	24	.0167			
c. Beef pre-heated to 150° F.					
Total	35				
Replication	3	.0167	.739	NS	NS
Storage time	8	.1359	6.013	S	S
Error	24	.0226			
d. Beef pre-heated to 160° F.					
Total	35				
Replication	3	.0155	2.924	NS	NS
Storage time	8	.0278	5.245	S	S
Error	24	.0053			
e. Beef pre-heated to 170° F.					
Total	35				
Replication	3	.0073	1.303	NS	NS
Storage time	8	.0127	2.267	NS	NS
Error	24	.0056			

It may be seen from Tables 5a, b, c, and d that the increase in total water-soluble nitrogen with time was significant in beef heated to internal temperatures of 130°, 140°, 150°, or 160° F. prior to irradiation to 3 megarads. The increase in total soluble nitrogen in the beef heated to 170° F., however, was not significant (Table 5e). These results mean that the factors responsible for the increase in total water-soluble nitrogen were inhibited or destroyed as a result of heating to 170° F. prior to irradiation to 3 megarads, whereas they remain wholly or partially active in beef heated to 160° F. or lower temperatures.

Linearity of Increase in Total Soluble Nitrogen. In order to determine the total water-soluble nitrogen content of beef heated to a particular internal temperature prior to irradiation to 3 megarads and placed in storage at 72° F. increased in a linear manner with time in storage, the test of linearity of regression of total soluble nitrogen content on storage time was made using the appropriate statistical method (25, p. 295-298). Calculations were made using the totals of replications as individual observations. The results of this test are shown in Tables 6a through e.

It may be seen from Table 6b that a significant deviation from linearity at 5 percent significance level occurred in meat heated to 140° F. prior to irradiation. The critical region as derived from F-tables is where the F-value is larger than 2.3732 with 7 and 27 degrees of freedom and the F-value for the significant deviation from linearity at 140° F. was only 2.3940. The difference in these

TABLE 6.

TEST OF LINEARITY OF REGRESSION OF TOTAL SOLUBLE NITROGEN CONTENT ON STORAGE TIME IN IRRADIATED BEEF

Variation Due to	Degrees of Freedom		Mean Square	F	Significance At the level of 5%	1%
a. Beef pre-heated to 130° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0295	.165	NS	NS
Error	27		.1792			
b. Beef pre-heated to 140° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0498	2.394	S	NS
Error	27		.0208			
c. Beef pre-heated to 150° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0443	2.032	NS	NS
Error	27		.0218			
d. Beef pre-heated to 160° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0122	1.906	NS	NS
Error	27		.0064			
e. Beef pre-heated to 170° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0030	.536	NS	NS
Error	27		.0056			

two values is small enough to be neglected. It can be stated, therefore, that under the conditions of this experiment the increase in total water-soluble nitrogen with time was linear within all levels of pre-irradiation heat treatment including the 170° F. level where the increase was found to be not significant.

Rate of Increase in Total Water-Soluble Nitrogen. In order to determine the relation among the rates of increase with time in total water-soluble nitrogen as effected by the levels of pre-irradiation heat treatment, regression coefficients, which show the amount of increase per day, were calculated using the pertinent method of calculation (25, p. 268). The results of these calculations are given in Table 7, with the resultant regression equations which have been translated into graphical form in Figure 1.

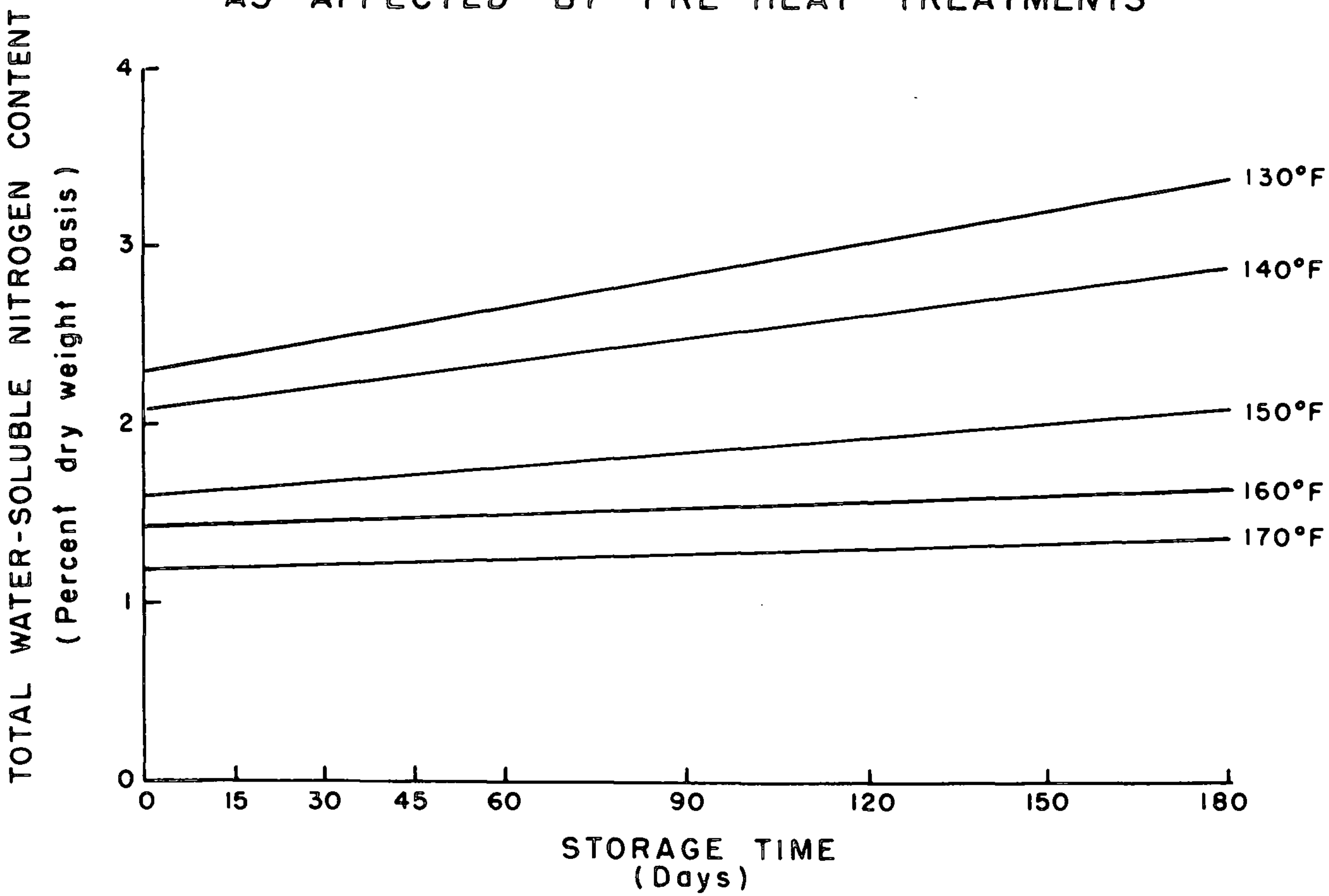
TABLE 7.
RATE OF INCREASE IN TOTAL SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF AS AFFECTED BY LEVEL OF HEAT TREATMENT

Heat Treatment ° F.	Regression Coefficient*	Regression Equation**
130	.00596	$\bar{y}_x = 2.783 + .00596\ (x-76.6)$
140	.00421	$\bar{y}_x = 2.434 + .00421\ (x-76.6)$
150	.00249	$\bar{y}_x = 1.809 + .00249\ (x-76.6)$
160	.00105	$\bar{y}_x = 1.522 + .00105\ (x-76.6)$
170	.00080	$\bar{y}_x = 1.248 + .00085\ (x-76.6)$

* Rate: gr./100 gr./day of storage.
** y = Total soluble nitrogen content.
x = Storage time (days).

FIGURE I

RATE OF CHANGE IN TOTAL WATER-SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF AS AFFECTED BY PRE-HEAT TREATMENTS



As may be derived from Table 7, the rates of increase at the pre-heating temperatures of 130°, 140°, 150°, and 160° F. are 7.0, 5.0, 2.9, and 1.2 times as that at 170° F., respectively.

More informative is a comparison among the ratios of adjacent rates of increase, as it permits an evaluation of the relative magnitude of the inhibitory or destructive effect of each particular 10° F. rise in pre-heating temperature on the factors responsible for the solubilization of nitrogenous constituents of beef during storage. Table 8 shows these ratios.

TABLE 8.
EFFECT OF EACH 10° F. RISE IN PRE-HEATING TEMPERATURE ON THE RATE OF INCREASE IN TOTAL SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF

Level of 10° F. rise In Temperature	Ratio of Adjacent Rates*
130 - 140	1.415
140 - 150	1.691
150 - 160	2.370
160 - 170	1.236

* Ratio of the rate in lower temperature to that in the higher one.

As may be seen from Table 8, the 10° F. span from 150° to 160° F. caused the highest reduction in the rate of increase in total soluble nitrogen. This serves to indicate the existeace of a "breaking point" at a temperature between 150° and 160° F. with reference to the effect of pre-heating temperatures on the inhibition or destruction of factors contributing towards the solubilization of nitrogenous constituents of pre-heated irradiated beef during storage at 72° F. This statement is further supported by the results of a test (25, p. 278-283) carried out to determine the significance of rates of change in total water-soluble nitrogen. These results are shown in Table 9.

TABLE 9.
SIGNIFICANCE OF RATE OF INCREASE IN TOTAL SOLUBLE NITROGEN AS AFFECTED BY PRE-HEATING TEMPERATURES

Heat Treatment ° F.	Regression Co-efficient*	Sum of Squares (Regression SS) w/1 d.f.	Variance (Residual SS/n-2) w/7 d.f.	$t^2=F$	Significance At the Level of 5%
130	.00596	4.4429	.0268	165.78	S
140	.00421	2.2170	.0802	27.64	S
150	.00249	.7775	.0847	9.18	S
160	.00105	.1370	.0249	5.50	NS
170	.00080	.0801	.0217	3.69	NS

*Regression coefficient or rate of increase.

Table 9 clearly shows that a significant rate of increase in total water soluble nitrogen was rendered insignificant by a 10° F. rise in temperature from 150° to 160° F. This indicates a stabilization effect of heating to a temperature between 150° and 160° F.

on the factors contributing towards solubilization of nitrogenous constituents of irradiated ground lean beef.

Trichloroacetic-Acid-Soluble Nitrogen

The data pertaining to the TCA-soluble nitrogen content of ground lean beef heated to internal temperatures of 130°, 140°, 150°, 160°, or 170° F. prior to irradiation to a dose of 3.0 megarads and stored at 72° F. for 0, 15, 30, 45, 60, 90, 120, 150, or 180 days are presented in Table 10.

TABLE 10.
TCA-SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F.
(Percent dry weight basis)

Heat Treatment ° F.	Storage Period (days)									Heat Treatment Mean
	0	15	30	45	60	90	120	150	180	
130	1.14	1.53	1.62	1.75	1.91	2.10	2.10	2.25	2.39	1.87
140	1.17	1.32	1.53	1.59	1.78	1.91	1.86	1.96	2.06	1.68
150	1.17	1.13	1.28	1.34	1.45	1.52	1.50	1.62	1.64	1.40
160	1.14	1.12	1.19	1.16	1.27	1.21	1.22	1.21	1.28	1.20
170	1.00	1.00	1.01	1.03	1.00	1.00	.95	1.05	1.07	1.01
Storage Period Mean	1.12	1.22	1.33	1.37	1.48	1.55	1.53	1.62	1.69	

Effect of the Level of Heat Treatment and Storage Period. The trend in the change in TCA-soluble nitrogen as affected by the two variables may be seen from Table 10. Heat treatment means showed a steady reduction in this constituent with increase in pre-irradiation heating temperature. Likewise, as the storage period increased the TCA-soluble nitrogen content also increased. However, definite answers in these and other pertinent respects such as the relative magnitude of change contributed by each variable, significance of change at each level of heat treatment, linearity and rate of change, can be obtained only through statistical analyses. The following results were obtained through the application of the appropriate statistical method (25, p. 309-324), using the values representing the TCA-soluble nitrogen contents of replicate samples as individual observations.

TABLE 11.
ANALYSIS OF VARIANCE FOR TCA-SOLUBLE NITROGEN IN IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F.

Variation Due to	Degrees of Freedom	Mean Square	F	Significance At the Level of 5%	1%
Total	179				
Replication	3	.0016	1.772	NS	NS
Temperature	4	4.3709	496.693	S	S
Temp. x Repl.	12	.0088	9.777	S	S
Time	8	.6962	102.355	S	S
Time x Repl.	24	.0068	7.555	S	S
Temp. x Time	32	1.1230	1247.777	S	S
Error	96	.0009			

The values in Table 11 show that there were five effects contributing toward variations in the TCA-soluble nitrogen content of beef heated to different pre-irradiation temperatures and kept in storage for nine different periods. These are the temperature effect, time effect, temperature x time interaction effect, temperature x replication interaction effect and time x replication interaction effect. The relative magnitude of each significant effect was calculated according to Li (25, p. 200-204) and can be seen from Table 12 which shows the extent of contribution by each effect towards variations in the TCA-soluble nitrogen content of beef.

TABLE 12.
RELATIVE MAGNITUDE OF CONTRIBUTION BY SIGNIFICANT EFFECTS TOWARDS VARIATIONS IN TCA-SOLUBLE NITROGEN CONTENT IN IRRADIATED BEEF

Effect	Variance
Temperature	.1214
Time	.0348
Temperature x Time	.2805
Temperature x Repl.	.0009
Time x Replication	.0012

The meaning of the values in Table 12 is that changes in the pre-irradiation heating temperatures have more pronounced effects on the TCA-soluble nitrogen content of beef than have the different storage periods. It is also evident from the significant temperature x time interaction effect that the variations in the TCA-soluble nitrogen content of beef for regularly different combinations of levels of pre-irradiation heat treatment and storage periods do not follow a similarly regular pattern and that no prediction can be made as to the TCA-soluble nitrogen content for a particular combination of the level of heat treatment and storage period on the basis of the results obtained for other such combinations. The effect of temperature x replication and time x replication is so small that they can be regarded as being negligible.

Significance of Change with Time within each Temperature Treatment. In order to arrive at conclusions as to whether TCA-soluble nitrogen content of beef heated to a particular temperature prior to irradiation to 3 megarads changed significantly with time, the data for each of the five levels of heat treatment were subjected to a separate analysis of variance (25, p. 196-208) the results of which are shown in Tables 13a through e.

It may be seen from Tables 13a, b, c, and d that the increases in TCA-soluble nitrogen with time were significant at the 5 percent significance level in beef heated to internal temperatures of 130°, 140°, 150°, or 160° F. prior to irradiation to 3 megarads. The increase of TCA-soluble nitrogen in beef heated to 160° F. was not significant at the 1 percent level. These results mean that the factors responsible for the increase in TCA-soluble nitrogen are inhibited or destroyed as a result of heating to 160° F. prior to irradiation to 3 megarads, whereas they remain wholly or partially active in beef heated to 150° F. or lower temperatures.

TABLE 13.
ANALYSIS OF VARIANCE FOR TCA-SOLUBLE NITROGEN IN IRRADIATED BEEF AS AFFECTED BY LENGTH OF STORAGE PERIOD AT 72° F.

Variation Due to	Degrees of Freedom	Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.					
Total	35				
Replication	3	.0071	.292	NS	NS
Storage time	8	.6219	25.590	S	S
Error	24	.0243			
b. Beef pre-heated to 140° F.					
Total	35				
Replication	3	.0183	2.859	NS	NS
Storage time	8	.3694	57.720	S	S
Error	24	.0064			
c. Beef pre-heated to 150° F.					
Total	35				
Replication	3	.0018	.360	NS	NS
Storage time	8	.1378	27.560	S	S
Error	24	.0050			
d. Beef pre-heated to 160° F.					
Total	35				
Replication	3	.0021	.600	NS	NS
Storage time	8	.0113	3.228	S	NS
Error	24	.0035			
e. Beef pre-heated to 170° F.					
Total	35				
Replication	3	.0077	2.556	NS	NS
Storage time	8	.0048	1.600	NS	NS
Error	24	.0030			

Linearity of Increase in TCA-Soluble Nitrogen. In order to determine if the TCA-soluble nitrogen content of beef heated to a particular internal temperature prior to irradiation to 3 megarads and placed in storage at 72° F. increased in a linear manner with time in storage, the test of linearity of regression of total soluble nitrogen content on storage time was made using the appropriate statistical method (25, p. 295-298). Calculations were made using the totals of replications as individual observations. The results of this test are shown in Tables 14a through e.

It may be seen from Table 14 that significant deviations from linearity at 5 percent significance level occurred in samples heated to 130°, 140°, or 150° F. However, the magnitude of significance in meat heated to 130° F. was so small that it disappeared when the 1 percent significance level was chosen. A high degree of importance cannot be attached to the linearity of the increase in TCA-soluble nitrogen in beef heated to 160° or 170° F., because it was shown in Tables 13d and e that the increase over time was not significant in the samples that received these levels of pre-irradiation heat treatment.

TABLE 14.
TEST OF LINEARITY OF REGRESSION OF TCA-SOLUBLE NITROGEN CONTENT ON STORAGE TIME IN IRRADIATED BEEF

Variation Due to	Degrees of Freedom		Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0785	3.505	S	NS
Error	27		.0224			
b. Beef pre-heated to 140° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0553	7.182	S	S
Error	27		.0077			
c. Beef pre-heated to 150° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0199	4.234	S	S
Error	27		.0047			
d. Beef pre-heated to 160° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0059	1.788	NS	NS
Error	27		.0033			
e. Beef pre-heated to 170° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0045	1.250	NS	NS
Error	27		.0036			

<u>Rate of Increase in TCA-Soluble Nitrogen</u>. In order to determine the relation among the rates of increase with time in TCA-soluble nitrogen as affected by the levels of pre-heat treatment, regression coefficients which show the amount of increase per day of storage were calculated on data using the pertinent method of calculation (25, p. 268). The results of these calculations are given in Table 15, with the resultant regression equations which have been translated into graphical form in Figure 2. As may be derived from Table 15, the rates of increase at the pre-heating temperatures of 130°, 140°, 150° and 160° F. are 24.8, 18.9, 11.6, and 2.6 times as that at 170° F., respectively.

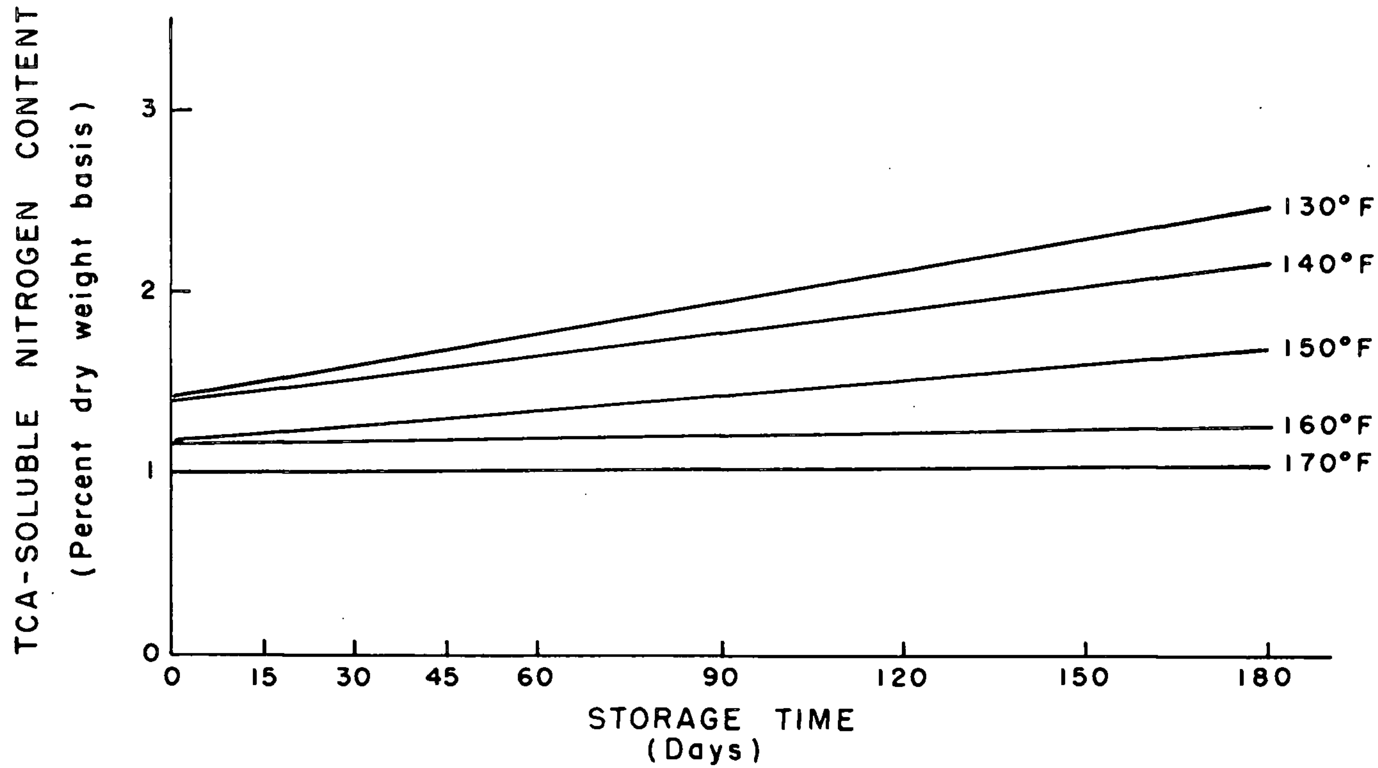
TABLE 15.
RATE OF INCREASE IN TCA-SOLUBLE NITROGEN CONTENT
OF IRRADIATED BEEF AS AFFECTED BY LEVEL OF HEAT TREATMENT

Heat Treatment ° F.	Regression Coefficient*	Regression Equation**
130	.00595	$\bar{y}_x = 1.866 + .00595\ (x-76.6)$
140	.00453	$\bar{y}_x = 1.685 + .00453\ (x-76.6)$
150	.00278	$\bar{y}_x = 1.403 + .00278\ (x-76.6)$
160	.00063	$\bar{y}_x = 1.198 + .00063\ (x-76.6)$
170	.00024	$\bar{y}_x = 1.011 + .00024\ (x-76.6)$

* Rate: gr./100 gr./day of storage.
** y = TCA-soluble nitrogen content.
x = Storage time (days).

FIGURE 2

RATE OF CHANGE IN TCA-SOLUBLE NITROGEN OF IRRADIATED BEEF AS AFFECTED BY PRE-HEAT TREATMENTS



More informative is a comparison among the ratios of adjacent rates of increase, as it permits an evaluation of the relative magnitude of the inhibitory or destructive effect of each particular 10° F. rise in pre-heating temperature on the factors responsible for protein breakdown during storage. Table 16 shows these ratios.

TABLE 16.
EFFECT OF EACH 10° F. RISE IN PRE-HEATING TEMPERATURE ON THE RATE OF INCREASE IN TCA-SOLUBLE NITROGEN CONTENT OF IRRADIATED BEEF

Level of 10° F. rise In Temperature	Ratio of Adjacent rates*
130 - 140	1.314
140 - 150	1.629
150 - 160	4.412
160 - 170	2.625

* Ratio of the rate in lower temperature to that in the higher one.

As may be seen from Table 16, the 10° F. span from 150° to 160° F. caused the highest reduction in the rate of increase in TCA-soluble nitrogen. This serves to indicate the existence of a "breaking point" at a temperature between 150° and 160° F. with reference to the effect of pre-heating temperatures on the inhibition or destruction of factors contributing toward the breakdown of protein in pre-heated irradiated beef during storage at 72° F. This statement is further supported by the results of a test (25, p. 278-

283) carried out to determine the significance of rates of change in TCA-soluble nitrogen. These results are shown in Table 17.

TABLE 17.
SIGNIFICANCE OF RATE OF INCREASE IN TCA-SOLUBLE NITROGEN AS AFFECTED BY PRE-HEATING TEMPERATURES

Heat Treatment ° F.	Regression Co-Efficient*	Sum of squares (Regression SS) w/1 d.f.	Variance (Residual SS/n-2) w/7 d.f.	$t^2=F$	Significance at the level of 5%
130	.00595	4.4257	.0862	51.342	S
140	.00453	2.5678	.0297	86.458	S
150	.00278	.9633	.0181	53.220	S
160	.00063	.0496	.0128	3.875	NS
170	.00024	.0071	.0137	.518	NS

* Regression coefficient or rate of increase.

Table 17 clearly shows that a highly significant rate of increase in TCA-soluble nitrogen was rendered insignificant by a 10° F. rise in temperature from 150° to 160° F. This also indicates a stabilization effect of heating to a temperature between 150° and 160° F. on the factors contributing towards protein breakdown during storage.

<u>Amino Nitrogen</u>

The data pertaining to amino nitrogen content of ground lean beef heated to internal temperatures of 130°, 140°, 150°, 160°, or 170° F. prior to irradiation to a dose of 3 megarads and stored at

72° F. for 0, 15, 30, 45, 60, 90, 120, 150, or 180 days, are presented in Table 18.

TABLE 18.
AMINO NITROGEN CONTENT OF IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F.
(Percent dry weight basis)

Heat Treat-ment ° F.	Storage Period (days)									Heat Treat-ment Mean
	0	15	30	45	60	90	120	150	180	
130	.284	.445	.569	.732	.811	.898	.929	.995	1.129	.755
140	.267	.389	.460	.552	.642	.714	.712	.792	.905	.604
150	.247	.284	.313	.344	.384	.446	.439	.477	.539	.386
160	.226	.250	.260	.266	.269	.258	.255	.240	.301	.258
170	.196	.203	.208	.203	.196	.184	.179	.182	.181	.192
Storage Period Mean	.244	.314	.362	.419	.461	.500	.503	.537	.611	

<u>Effect of the Level of Heat Treatment and Storage Period.</u> The trend in the change in amino nitrogen content as affected by the two variables may be seen from Table 18. Heat treatment means showed a steady reduction in this constituent with increase in pre-irradiation heating temperature. Likewise, as the storage period increased the amino nitrogen content also increased. However, definite answers in these and other pertinent respects such as the relative magnitude of change contributed by each variable, significance of change at each level of heat treatment, linearity and rate of change, can be obtained only through statistical analyses. The following results

were obtained through the application of the appropriate statistical methods (25, p. 309-324), using the values representing the amino nitrogen contents of replicate samples as individual observations.

TABLE 19.
ANALYSIS OF VARIANCE FOR AMINO NITROGEN IN IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F.

Variation Due to	Degrees of Freedom	Mean Square	F	Significance At the Level of 5%	1%
Total	179				
Replication	3	.0123	30.75	S	S
Temperature	4	2.0078	803.12	S	S
Temp. x Repl.	12	.0025	6.25	S	S
Time	8	.2684	178.93	S	S
Time x Repl.	24	.0015	3.75	S	S
Temp. x Time	32	.0601	150.25	S	S
Error	96	.0004			

The values in Table 19 show that all the effects contributed significantly toward variations in the amino nitrogen content of beef heated to different pre-irradiation temperatures and kept in storage for nine different periods. The relative magnitude of each of these significant effects was calculated according to Li (25, p. 200-204) and can be seen from Table 20, which shows the extent of contribution by each effect towards variations in the TCA-soluble nitrogen content of beef.

TABLE 20.
RELATIVE MAGNITUDE OF CONTRIBUTION BY SIGNIFICANT EFFECTS TOWARDS VARIATIONS IN AMINO NITROGEN CONTENT OF IRRADIATED BEEF

Effect	Variance
Replication	.0003
Temperature	.0558
Temperature x Replication	.0002
Time	.0134
Time x Replication	.0002
Temperature x Time	.0149

The meaning of the values in Table 20 is that changes in the pre-irradiation heating temperatures have more pronounced effects on the amino nitrogen content of beef than have the different storage periods. It is also evident from the relatively highly significant temperature x time interaction effect that the variations in the amino nitrogen content of beef for regularly different combinations of levels of pre-irradiation heat treatment and storage periods do not follow a similarly regular pattern and that no prediction can be made as to the amino nitrogen content for a particular combination of the level of heat treatment and storage period on the basis of the results obtained for other such combinations. Replication effect as well as temperature x replication and time x replication interaction effects have such a small significance that they may be considered negligible.

<u>Significance of Change with Time Within each Temperature Treatment.</u> In order to arrive at conclusions as to whether amino nitrogen content of beef heated to a particular temperature prior to irradiation at 3 megarads changed significantly with time, the data for each of the five levels of heat treatment were subjected to a separate analysis of variance (25, p. 196-208), the results of which are shown in Tables 21a through e.

TABLE 21.
ANALYSIS OF VARIANCE FOR AMINO NITROGEN IN IRRADIATED BEEF AS AFFECTED BY LENGTH OF STORAGE PERIOD AT 72° F.

Variation Due to	Degrees of Freedom	Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.					
Total	35				
Replication	3	.0038	6.333	S	S
Storage time	8	.3020	503.333	S	S
Error	24	.0006			
b. Beef pre-heated to 140° F.					
Total	35				
Replication	3	.0142	10.923	S	S
Storage time	8	.1621	128.500	S	S
Error	24	.0013			
c. Beef pre-heated to 150° F.					
Total	35	.			
Replication	3	.0021	2.625	NS	NS
Storage time	8	.0324	46.750	S	S
Error	24	.0008			
d. Beef pre-heated to 160° F.					
Total	35				
Replication	3	.0015	5.000	S	S
Storage time	8	.0017	5.600	S	S
Error	24	.0003			
e. Beef pre-heated to 170° F.					
Total	35				
Replication	3	.00050	8.333	S	S
Storage time	8	.00050	8.333	S	S
Error	24	.00006			

It may be seen from Tables 21a through e that increases in amino nitrogen with time were significant in beef heated to internal temperatures of 130°, 140°, 150°, 160°, or 170° F. prior to irradiation at 3 megarads. These results mean that the factors responsible for the increase in amino nitrogen are not inhibited or destroyed completely as a result of heating up to and including 170° F. prior to irradiation at 3 megarads.

Linearity of Increase in Amino Nitrogen. In order to see if the amino nitrogen content of beef heated to a particular internal temperature prior to irradiation at 3 megarads and placed in storage at 72° F. increased in a linear manner with time in storage, the test of linearity of regression of amino nitrogen content on storage time was made using the appropriate statistical method (25, p. 295-298). Calculations were made using the totals of replications as individual observations. The results of this test are shown in Tables 22a through e.

As may be seen from Tables 22a, b, c, and d, significant deviations from linearity occurred in amino nitrogen content of beef pre-heated to 130°, 140°, 150°, or 160° F. Considering the fact that the critical region at 1 percent level is where the F-value, as derived from the F-tables, is larger than 3.3882 with 7 and 27 degrees of freedom, deviation from linearity in beef pre-heated to 140°, 150°, or 160° F. is not of high significance. The beef pre-heated to 130° F. showed a degree of significance which can not be

TABLE 22.
TEST OF LINEARITY OF REGRESSION OF AMINO NITROGEN CONTENT ON STORAGE TIME IN IRRADIATED BEEF

Variation Due to	Degrees of Freedom		Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0391	39.100	S	S
Error	27		.0010			
b. Beef pre-heated to 140° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0160	5.714	S	S
Error	27		.0028			
c. Beef pre-heated to 150° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0036	3.600	S	S
Error	27		.0010			
d. Beef pre-heated to 160° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0014	3.500	S	S
Error	27		.0004			
e. Beef pre-heated to 170° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	.0002	2.000	NS	NS
Error	27		.0001			

neglected. However, for the discussion of rates of change, the increase in amino nitrogen will be regarded as being linear over storage time within all pre-heating temperatures.

Rate of Increase in Amino Nitrogen. In order to determine the relation among the rates of increase with time in amino nitrogen as affected by the levels of pre-heat treatment, regression coefficients which show the amount of increase per day of storage were calculated on the data using the pertinent method of calculation (25, p. 268). The results of these calculations are given in Table 23, with the resultant regression equations which have been translated into graphical form in Figure 3.

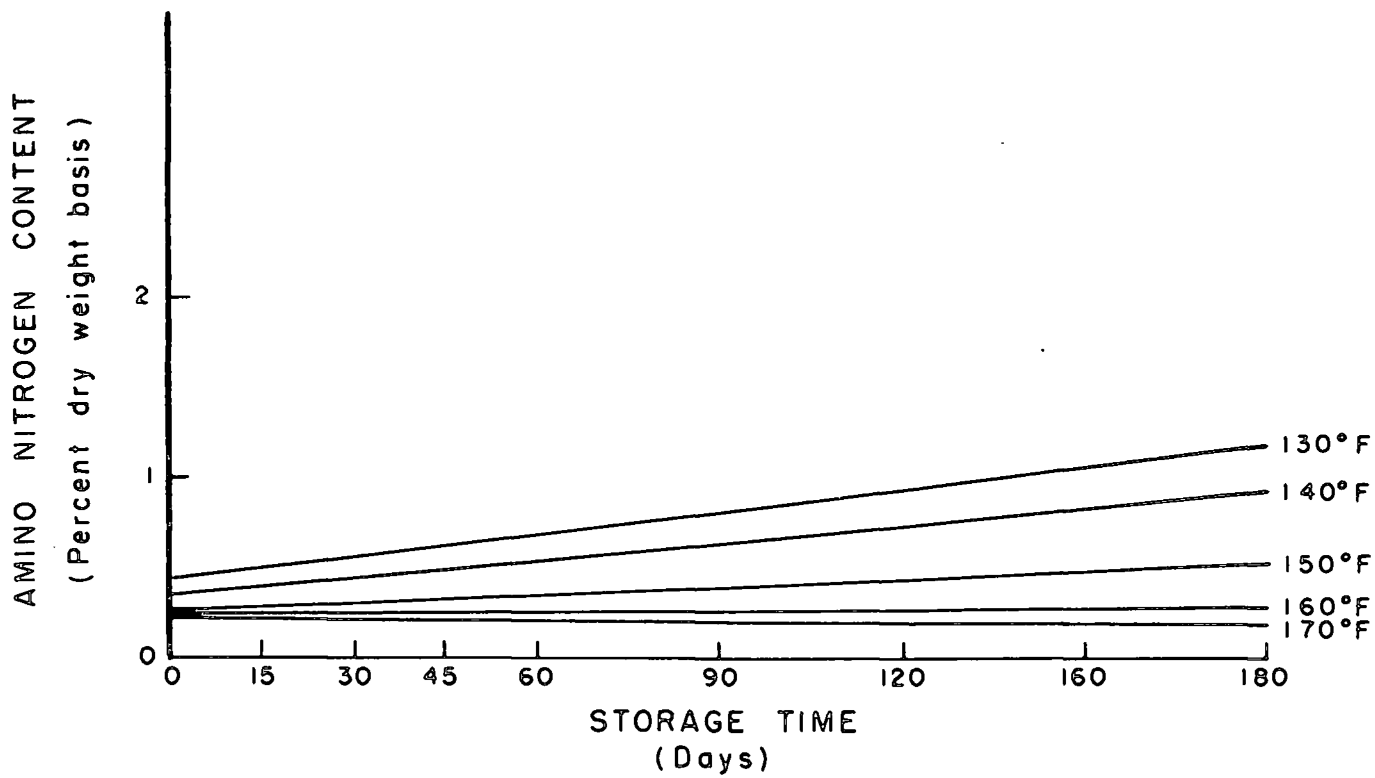
TABLE 23.
RATE OF INCREASE IN AMINO NITROGEN CONTENT
OF IRRADIATED BEEF AS AFFECTED BY LEVEL OF HEAT TREATMENT

Heat Treatment ° F.	Regression Coefficient*	Regression Equation**
130	.00414	$\bar{y}_x = .755 + .00414\ (x-76.6)$
140	.00313	$\bar{y}_x = .604 + .00313\ (x-76.6)$
150	.00148	$\bar{y}_x = .386 + .00148\ (x-76.6)$
160	.00017	$\bar{y}_x = .258 + .00017\ (x-76.6)$
170	-.00015	$\bar{y}_x = .192 - .00015\ (x-76.6)$

* Rate: gr./100 gr./day of storage.
** y = Amino nitrogen content.
x = Storage time (days).

FIGURE 3

RATE OF CHANGE IN AMINO NITROGEN CONTENT OF IRRADIATED BEEF AS AFFECTED BY PRE-HEAT TREATMENTS



As may be derived from Table 23, the rates of increase at the pre-heating temperatures of 130°, 140°, 150°, and 160° F. are 27.6, 20.9, 9.9, and 1.1 times as that at 170° F., respectively. It should also be noted that the direction of change is different in beef pre-heated to 170° F., which shows that a decrease in amino nitrogen content was noted in this beef while others showed an increase.

More informative is a comparison among the ratios of adjacent rates of increase, as it permits an evaluation of the relative magnitude of the inhibitory or destructive effect of each particular 10° F. rise in the pre-heating temperature on the factors responsible for breakdown of proteins to amino acids or lower peptides. Table 24 shows these ratios.

TABLE 24.
EFFECT OF EACH 10° F. RISE IN PRE-HEATING TEMPERATURE ON THE RATE OF INCREASE IN AMINO NITROGEN CONTENT OF IRRADIATED BEEF

Level of 10° F. rise in Temperature	Ratio of Adjacent Rates*
130 - 140	1.323
140 - 150	2.115
150 - 160	8.706
160 - 170	.566

* Ratio of the rate in lower temperature to that in the higher one.

As may be seen from Table 24, the 10° F. span from 150° to 160° F. caused the highest reduction in the rate of increase in amino nitrogen. This serves to indicate the existence of a "breaking point" at a temperature between 150° and 160° F. with reference to the effect of pre-heating temperatures on the inhibition or destruction of factors contributing toward the breakdown of proteins to amino acids and lower peptides. This statement is further supported by the results of a test (25, p. 278-283) carried out to determine the significance of rates of change in amino nitrogen. These results are shown in Table 25.

TABLE 25.
SIGNIFICANCE OF RATE OF INCREASE IN AMINO NITROGEN AS AFFECTED BY PRE-HEATING TEMPERATURES

Heat Treatment ° F.	Regression Co-efficient*	Sum of squares (Regression SS) w/1 d.f.	Variance (Residual SS/n-2) w/7 d.f.	t^2=F	Significance at the level of 5%
130	.00414	2.1418	.0038	563.63	S
140	.00313	1.2248	.0107	114.47	S
150	.00148	.2738	.0037	74.00	S
160	.00017	.0038	.0016	2.38	NS
170	-.00015	.0028	.0004	6.71	S**

* Regression coefficient or rate of change.
** Not significant at 1 percent level.

Table 25 clearly shows that a highly significant rate of increase in amino nitrogen was rendered insignificant by a 10° F. rise in temperature from 150° to 160° F. This indicates a stabilization effect of heating to a temperature between 150° and 160° F. on the factors contributing towards breakdown of proteins during storage to amino acids and lower peptides.

Results of Flavor Evaluations

Flavor attributes of ground lean beef heated to different internal temperatures prior to irradiation were evaluated with reference to a sample which received the same treatment as one of the experimental samples. Three flavor attributes were evaluated by eight tasters at the end of each of the nine different storage periods. Four replications were involved in the experiment. Considering the fact, however, that the tasters are incidental, the 32 flavor scores obtained at each storage period for each level of pre-heat treatment were treated as 32 replications of that particular combination of pre-heat treatment and storage period. The results of analyses on the data for each flavor attribute are presented and discussed in the subsequent paragraphs.

Irradiation Flavor

The data pertaining to the relative irradiation flavor of ground lean beef heated to internal temperatures of 130°, 140°, 150°, 160°, or 170° F. prior to irradiation to 3 megarads and stored at 72° F. for 0, 15, 30, 45, 60, 90, 120, 150, or 180 days, are presented in Table 26.

TABLE 26.
IRRADIATION FLAVOR SCORE OF IRRADIATED BEEF
AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS
AND LENGTH OF STORAGE AT 72° F.
(Means)

Heat Treatment ° F.	Storage Period (days) 0	15	30	45	60	90	120	150	180	Heat Treatment Mean
130	3.9	4.1	4.1	4.1	3.4	3.6	3.7	3.3	3.2	3.7
140	3.8	4.0	4.1	3.8	3.5	3.4	3.5	3.3	3.2	3.6
150	3.4	3.6	3.7	3.8	3.6	3.5	3.4	3.3	3.3	3.5
160	3.9	3.9	4.2	3.8	3.7	3.9	3.7	3.6	3.8	3.8
170	4.4	4.1	4.2	4.1	4.0	4.1	3.8	4.1	4.1	4.1
Storage Period Mean	3.9	3.9	3.9	3.9	3.6	3.7	3.6	3.5	3.5	

In order to avoid erroneous interpretations, it should be emphasized that the values appearing in Table 26 represent the evaluations made with respect to a reference sample which might have undergone changes in irradiation flavor during storage. A comparison of the values <u>within any column</u> will provide an indication of the relative intensity of irradiation flavor at a certain storage period among beef that received different levels of pre-heat treatment. The values <u>within a given row</u>, however, should be regarded as being devoid of such a capability. It may be said that, during 0-day storage, the irradiation flavor of beef heated to 130° F. was less intense than that of the beef heated to 150° F. because both of them had been evaluated against the same reference beef. It cannot be stated, however, that the irradiation flavor of beef heated to 130° F. prior to irradiation was less intense during 0-day storage than it was during 60-days of storage, because no proof can be presented that the reference beef used during 0-day storage had the same intensity of irradiation flavor as that used during 60-days of storage. A comparison among rows should indicate the relation among the rates of change in irradiation flavor over the entire storage period as affected by pre-heating temperatures. It should also show the effect of pre-heating temperatures on the degree of intensity of irradiation flavor.

Keeping the foregoing in mind it may be seen from Table 26, as indicated by pre-heating temperature means, that the irradiation flavor of the beef pre-heated to 150° F. was the poorest with respect to that of the reference sample. The beef pre-heated to 140° F. had less intense irradiation flavor and the one-pre-heated to 130° F. was still better in this respect. The beef pre-heated to 160° F. did not seem to be greatly different in irradiation flavor from the one heated to 170° F. which was rated as being practically the same as the reference beef in this attribute. Table 26 also reveals that the beef pre-heated to 130° F. did not differ appreciably from the reference beef until 60 days of storage when there was a sharp increase in the relative irradiation flavor with respect to that of the reference beef and that the increase became more pronounced at and after 150 days storage. The same can be said for the beef pre-heated to 140° F. The beef pre-heated to 150° F. changed for the better in irradiation flavor with respect to that of the reference beef up to 60 days of storage when there was an increase in this attribute with respect to that of the reference and that the rate of change followed the same trend thereafter as that of the beef pre-heated to 130° or 140° F. The beef pre-heated to 160° F. followed a parallel change in irradiation flavor as that of the reference beef. The irradiation flavor of the beef heated to 170° F. was practically the same as that of the reference beef during the entire storage period.

The above conclusions are derived from the values in Table 26 which represent averages of 32 replications and may not be as definite as the results of statistical analyses based on individual scores. The results of the statistical analyses are presented in the subsequent paragraphs.

Significance of Irradiation Flavor Difference between the Experimental and Reference Samples. In order to establish whether the panel scores for irradiation flavor differed significantly from the score assigned to the reference, the data for each combination of pre-heat treatment and storage period were subjected to an analyses using the pertinent statistical method (25, p. 309-324). The results of this analyses are shown in Tables 27a through i.

TABLE 27.
SIGNIFICANCE OF IRRADIATION FLAVOR DIFFERENCE BETWEEN THE EXPERIMENTAL AND REFERENCE BEEF

Heat Treatment ° F.	Mean Score Minus 4	Sum of Squares	Variance (s^2)	$t^2 = F$ with 1 and 31 Degrees of Freedom	Significance at 5% level	Less Or more Preferable Than Reference
a. Beef stored for 0 day at 72° F.						
130	-.091	48.13	1.5526	.1707	NS	
140	-.237	39.85	1.2855	1.3982	NS	
150	-.591	71.81	2.3165	4.8249	S	less
160	-.150	55.98	1.8058	.3987	NS	
170	+.419	15.49	.4997	11.2426	S	more
b. Beef stored for 15 days at 72° F.						
130	+.075	31.40	1.0129	.1777	NS	
140	.000	23.50	.7581	.0000	NS	
150	-.375	27.68	.8929	5.0398	S	less
160	-.125	21.78	.7026	.7116	NS	
170	+.072	12.70	.4097	.4049	NS	
c. Beef stored for 30 days at 72° F.						
130	+.128	28.90	.9323	.5624	NS	
140	+.059	26.64	.8594	.1296	NS	
150	-.309	15.97	.5152	5.9305	S	less
160	+.147	10.72	.3458	1.9997	NS	
170	+.188	11.17	.3603	3.1391	NS	
d. Beef stored for 45 days at 72° F.						
130	+.081	27.07	.8732	.2404	NS	
140	-.222	22.97	.7410	2.1283	NS	
150	-.225	16.62	.5361	3.0218	NS	
160	-.169	18.29	.5900	1.5491	NS	
170	+.109	12.51	.4035	.9422	NS	
e. Beef stored for 60 days at 72° F.						
130	-.603	31.73	1.0235	11.3683	S	less
140	-.462	32.25	1.0403	6.5656	S	less
150	-.444	26.04	.8400	7.5099	S	less
160	-.347	25.02	.8071	4.7740	S	less
170	-.009	9.01	.2906	.0089	NS	

Table 27, continued

Heat Treatment ° F.	Mean Score Minus 4	Sum of Squares	Variance (s^2)	$t^2 = F$ with 1 and 31 Degrees of Freedom	Significance at 5% level	Less Or more Preferable Than Reference
f. Beef stored for 90 days at 72° F.						
130	-.362	30.77	.9926	4.2247	S	less
140	-.566	20.33	.6558	15.6319	S	less
150	-.484	23.30	.7516	9.9736	S	less
160	-.125	14.86	.4794	1.0430	NS	
170	+.125	6.04	.1948	2.5667	NS	
g. Beef stored for 120 days at 72° F.						
130	-.334	21.71	.7003	5.0975	S	less
140	-.503	29.43	.9494	8.5278	S	less
150	-.641	26.80	.8645	15.2090	S	less
160	-.284	10.34	.3335	7.7391	S	less
170	-.178	14.57	.4700	2.1572	NS	
h. Beef stored for 150 days at 72° F.						
130	-.697	44.63	1.4397	10.7980	S	less
140	-.722	38.55	1.2435	13.4146	S	less
150	-.700	22.28	.7187	21.8172	S	less
160	-.369	22.06	.7116	6.1230	S	less
170	+.063	9.37	.3023	.4201	NS	
i. Beef stored for 180 days at 72° F.						
130	-.778	53.35	1.7210	11.2546	S	less
140	-.806	35.22	1.1361	18.2980	S	less
150	-.697	19.63	.6332	24.5513	S	less
160	-.209	9.91	.3197	4.3722	S	less
170	+.109	5.95	.1919	1.9812	NS	

TABLE 27--abridged
SIGNIFICANCE OF IRRADIATION FLAVOR DIFFERENCE
BETWEEN THE EXPERIMENTAL AND REFERENCE BEEF
(-S and +S indicates less and more preferable flavor, respectively)

Heat Treatment	Storage Period (days)								
	0	15	30	45	60	90	120	150	180
130	NS	NS	NS	NS	-S	-S	-S	-S	-S
140	NS	NS	NS	NS	-S	-S	-S	-S	-S
150	-S	-S	-S	NS	-S	-S	-S	-S	-S
160	NS	NS	NS	NS	-S	NS	-S	-S	-S
170	+S	NS	NS	NS	NS	NS	NS	NS	NS

It may be seen from Table 27--abridged that the beef pre-heated to 130° or 140° F. internal temperatures did not show a significant difference in irradiation flavor with respect to that of the reference beef until after 45 days of storage when significant differences were detected by the tasters. On this basis, it can be stated that irradiated ground lean beef heated to internal temperature of 130° or 140° F. could be stored for 45 days at 72° F. without a significant difference in irradiation flavor between these and the beef heated to 170° F. being detected by the panel.

Irradiation flavor in beef pre-heated to 150° F. was significantly more intense than that of the reference beef for 30 days of storage. This significance disappeared on the 45th day of storage.

Beef pre-heated to 160° F. did not show any significant intensification in irradiation flavor with respect to that of the reference beef until 60 days of storage, when a slight significance, as shown in Table 27e, was observed which disappeared during 90 days of storage. On this basis, it can be stated that this beef could be stored for 90 days at 72° F. without the panel being able to detect a significant difference in irradiation flavor between this and the beef pre-heated to 170° F.

The significantly high score observed on initial day of storage for beef pre-heated to 170° F., which was the same pre-heating temperature as that of the reference beef, can be explained on the basis of the tendency of a taste panel to assign an unduly high score to the better samples served with considerably poorer ones. Other than this single instance which occurred at the start of the experiment, the beef pre-heated to 170° F. was evaluated by the panel as having the same degree of irradiation flavor as the reference beef which had received identical treatment in all respects.

Linearity of Relative Irradiation Flavor Change. In order to see if the irradiation flavor of beef within individual pre-heat treatments changed in a linear manner with respect to that of the reference beef with time in storage, the test of linearity of regression of flavor scores on time was made using the appropriate statistical method (25, p. 295-298). The results of this test are shown in Tables 28a through e.

TABLE 28
TEST OF LINEARITY OF REGRESSION OF IRRADIATION FLAVOR SCORE ON STORAGE TIME IN IRRADIATED BEEF

Variation Due to	Degrees of Freedom		Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	11.0814	1.609	NS	NS
Error	27		6.8880			
b. Beef pre-heated to 140° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	5.5129	1.030	NS	NS
Error	27		5.3507			
c. Beef pre-heated to 150° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	4.4486	1.683	NS	NS
Error	27		2.6426			
d. Beef pre-heated to 160° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	5.1200	.980	NS	NS
Error	27		5.2233			
e. Beef pre-heated to 170° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	5.6130	4.483	S	S
Error	27		1.2522			

It may be seen from Table 28 that the only significant deviation from linearity occurred in beef heated to 170° F. prior to irradiation. The critical region at 1 percent significance level is where the F-value, as derived from F-tables, is larger than 3.3882 with 7 and 27 degrees of freedom and the F-value for the above deviation from linearity is only 4.4830, which shows that the magnitude of significance was not large. It can be stated, therefore, that for all practical purposes the change in irradiation flavor with respect to that of the reference was linear within all levels of pre-irradiation heat treatment.

Rate of Relative Irradiation Flavor Change. In order to determine the relation among the changes in relative irradiation flavor as affected by the levels of pre-heat treatment, regression coefficients which show the amount of change per day of storage were calculated using the pertinent method of calculation (25, p. 268). The results of these calculations are given in Table 29, with the resultant regression equations which have been translated into graphical form in Figure 4.

As may be derived from Table 29, the rates of change in relative irradiation flavor of a beef at the pre-heating temperatures of 130°, 140°, 150°, and 160° F. are 3.8, 3.5, 1.6, and 1.0 times as that at 170° F., respectively. More informative is a comparison among the ratios of adjacent rates of change in irradiation flavor, which are shown in Table 30.

TABLE 29.

RATE OF CHANGE IN RELATIVE IRRADIATION FLAVOR OF IRRADIATED BEEF AS AFFECTED BY HEAT TREATMENT

Heat Treatment ° F.	Regression Coefficient*	Regression Equation**
130	-.03763	$\bar{y}_x = 118.8 - .03763\ (x-76.6)$
140	-.03467	$\bar{y}_x = 115.7 - .03467\ (x-76.6)$
150	-.01541	$\bar{y}_x = 112.1 - .01541\ (x-76.6)$
160	-.01003	$\bar{y}_x = 122.2 - .01003\ (x-76.6)$
170	-.00992	$\bar{y}_x = 131.2 - .00992\ (x-76.6)$

* Rate: Change per day in the total of scores for 32 replications.
** y = Irradiation flavor score (total of 32 replications).
x = Storage time (days).

TABLE 30.

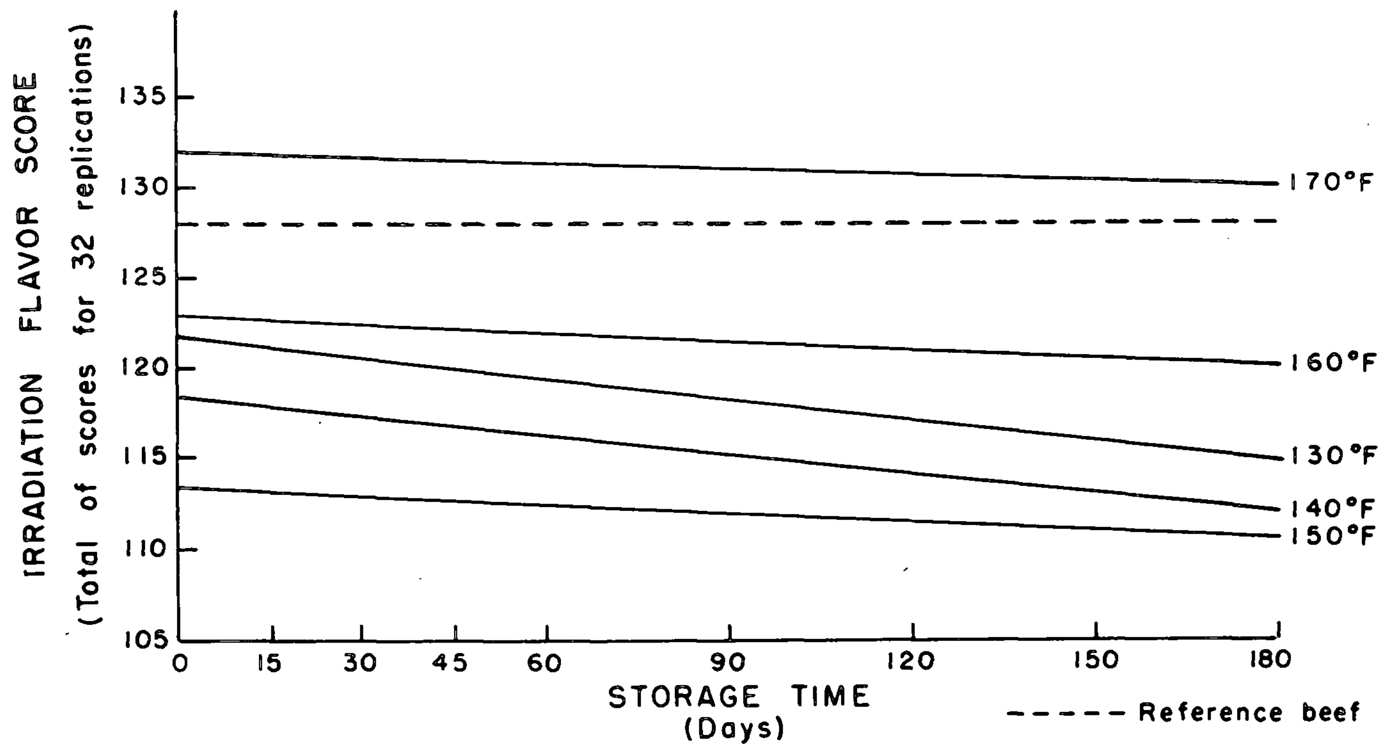
EFFECT OF EACH 10° F. RISE IN PRE-HEATING TEMPERATURE ON THE RATE OF CHANGE IN IRRADIATION FLAVOR OF IRRADIATED BEEF

Level of 10° F. rise In Temperature	Ratio of Adjacent Rates*
130 - 140	1.085
140 - 150	2.250
150 - 160	1.521
160 - 170	1.014

* Ratio of the rate in lower temperature to that in the higher one.

FIGURE 4

RATE OF CHANGE IN RELATIVE IRRADIATION FLAVOR OF IRRADIATED BEEF AS AFFECTED BY PRE-HEAT TREATMENTS



As may be seen from Table 30, the 10° F. rise from 140° to 150° F. caused the highest decrease in the rate of change in relative irradiation flavor. This suggests the presence of a "breaking point" between 140° and 150° F. with reference to the inhibition or destruction of factors contributing towards the increases in the rates of change in relative irradiation flavor. This statement is further supported by the results of a test (25, p. 278-283) carried out to determine the significance of rates of change in irradiation flavor. These results are shown in Table 31.

TABLE 31.
SIGNIFICANCE OF RATE OF CHANGE IN IRRADIATION FLAVOR
AS AFFECTED BY PRE-HEATING TEMPERATURES

Heat Treatment ° F.	Sum of squares (Regression SS) w/1 d.f.	Variance (Residual SS/n-2) w/7 d.f.	$t^2 = F$	Significance At the Level of 5%
130	177.050	26.567	6.664	S
140	150.230	20.639	7.279	S
150	29.690	10.193	2.913	NS
160	12.580	20.015	.629	NS
170	12.310	4.830	2.549	NS

As may be seen from Table 31 the rate of change in irradiation flavor was significant in meat pre-heated to 140° F. but this significance disappeared as a result of 10° F. rise in the pre-heating temperature. This also shows that there was a "breaking point" somewhere between 140° and 150° F.

Bitterness

The data pertaining to relative bitterness of ground lean beef heated to internal temperatures of 130°, 140°, 150°, 160°, or 170° F. prior to irradiation to 3 megarads and stored at 72° F. for 0, 15, 30, 45, 60, 90, 120, 150, or 180 days, are presented in Table 32.

TABLE 32.
BITTERNESS SCORE OF IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F. (Means).

Heat Treatment ° F.	Storage Period (days)									Heat Treatment Mean
	0	15	30	45	60	90	120	150	180	
130	4.0	4.3	4.0	3.9	3.7	3.7	3.5	3.1	3.1	3.7
140	3.8	4.1	4.0	3.9	3.8	3.6	3.5	3.3	3.0	3.7
150	3.7	4.0	4.0	3.9	3.7	3.5	3.3	3.3	3.2	3.6
160	3.9	4.1	4.0	3.9	4.0	4.0	3.6	3.7	3.6	3.9
170	3.9	4.1	3.9	4.0	4.0	3.9	3.8	3.9	3.9	3.9
Storage Period Mean	3.9	4.1	4.0	3.9	3.8	3.7	3.5	3.5	3.4	

In order to avoid erroneous interpretations, it should be emphasized that the values appearing in Table 32 represent the evaluations made with respect to a reference sample which might have undergone changes in bitterness during storage. A detailed discussion in this respect has been presented under irradiation flavor on page 57

Keeping the relativity of these scores in mind, it may be seen from the pre-heating temperature means in Table 32 that bitterness of beef pre-heated to 130°, 140°, or 150° F. differed from that of the reference beef practically to the same degree.

The beef pre-heated to 160° or 170° F. did not differ appreciably in bitterness from the reference beef. In the beef pre-heated to 170° F., this was true for the entire storage period whereas in the one pre-heated to 160° F. a lowering of the scores was observed at 120 days of storage, which continued until the conclusion of the experiment.

Initially and for a period of 45 days, the beef pre-heated to 130° F. was practically the same in bitterness as the one pre-heated to 170° F. Bitterness constantly increased in this sample starting at 60 days of storage. The same can be stated for the beef pre-heated to 150° F. It seems that bitterness with respect to that of the reference beef has not increased in this sample before 60 days of storage.

The above conclusions are derived from the values in Table 32 which represent averages of 32 replications and may not be as definite as the results of statistical analyses based on individual scores. The results of the statistical analyses are presented in the subsequent paragraphs.

Significance of Bitterness Difference Between Experimental and Reference Samples. In order to establish whether the panel scores for bitterness differed significantly from the score assigned to the reference, the data for each combination of pre-heat treatment and storage period were subjected to analysis using the pertinent statistical method (25, p. 196-208). The results of this analysis are shown in Tables 33a through i.

TABLE 33.
SIGNIFICANCE OF BITTERNESS DIFFERENCE BETWEEN THE EXPERIMENTAL AND REFERENCE BEEF

Heat Treat-ment ° F.	Mean Score Minus 4	Sum of Squares	Variance (s^2)	$t^2 = F$ with 1 and 31 Degrees of Freedom	Signifi-cance at 5% level	Less Or More Preferable Than Reference
a. Beef stored for 0 day at 72° F.						
130	+.003	9.35	.3016	.0010	NS	
140	-.228	22.60	.7290	2.2819	NS	
150	-.350	12.08	.3897	10.0590	S	less
160	-.116	12.18	.3929	1.0959	NS	
170	-.097	10.37	.3345	.9001	NS	
b. Beef stored for 15 days at 72° F.						
130	+.313	14.73	.4752	6.5972	S	more
140	+.147	11.20	.3613	1.9139	NS	
150	-.025	19.50	.6290	.0318	NS	
160	+.097	9.43	.3042	.9898	NS	
170	+.078	8.53	.2752	.7074	NS	
c. Beef stored for 30 days at 72° F.						
130	+.097	16.15	.5210	.5779	NS	
140	+.006	11.96	.3858	.0030	NS	
150	-.028	14.62	.4716	.0532	NS	
160	+.041	13.48	.4348	.1237	NS	
170	-.059	10.94	.3529	.3156	NS	

Table 33, continued

Heat Treatment ° F.	Mean Score Minus 4	Sum of Squares	Variance (s^2)	$t^2 = F$ with 1 and 31 Degrees of Freedom	Significance at 5% level	Less Or More Preferable Than Reference
d. Beef stored for 45 days at 72° F.						
130	-.103	19.63	.6332	.5361	NS	
140	-.062	14.97	.4829	.2547	NS	
150	-.087	8.09	.2610	.9280	NS	
160	-.109	6.73	.2171	.7512	NS	
170	-.012	8.91	.2874	.0160	NS	
e. Beef stored for 60 days at 72° F.						
130	-.312	21.03	.6784	4.5917	S	less
140	-.200	25.02	.8071	1.5859	NS	
150	-.269	13.95	.4500	5.1457	S	less
160	-.031	12.09	.3900	.0789	NS	
170	-.047	3.56	.1148	.6157	NS	
f. Beef stored for 90 days at 72° F.						
130	-.403	12.23	.3945	13.1739	S	less
140	-.450	20.70	.6677	9.7050	S	less
150	-.491	18.18	.6061	12.7282	S	less
160	-.037	12.37	.3990	.1098	NS	
170	-.100	10.22	.3297	.9706	NS	
g. Beef stored for 120 days at 72° F.						
130	-.553	22.98	.7413	13.2100	S	less
140	-.547	21.68	.6994	13.6899	S	less
150	-.725	20.62	.6652	25.2856	S	less
160	-.378	11.65	.3758	12.1668	S	less
170	-.219	12.35	.3984	3.8523	NS	
h. Beef stored for 150 days at 72° F.						
130	-.891	40.87	1.3184	19.2690	S	less
140	-.728	38.92	1.2555	13.5082	S	less
150	-.722	25.65	.8274	20.1609	S	less
160	-.278	17.27	.5571	4.4392	S	less
170	-.031	6.97	.2161	.1423	NS	
i. Beef stored for 180 days at 72° F.						
130	-.934	45.97	1.4829	18.8249	S	less
140	-.966	33.85	1.0919	27.3477	S	less
150	-.794	25.84	.8335	24.2039	S	less
160	-.378	10.75	.3468	13.1842	S	less
170	-.078	7.09	.2287	.8513	NS	

TABLE 33--abridged
SIGNIFICANCE OF BITTERNESS DIFFERENCE
BETWEEN THE EXPERIMENTAL AND REFERENCE BEEF
(-S and +S indicates less and more preferable flavor, respectively)

Heat Treatment	Storage Period (days)								
	0	15	30	45	60	90	120	150	180
130	NS	+S	NS	NS	-S	-S	-S	-S	-S
140	NS	NS	NS	NS	NS	-S	-S	-S	-S
150	-S	NS	NS	NS	-S	-S	-S	-S	-S
160	NS	NS	NS	NS	NS	NS	-S	-S	-S
170	NS	NS	NS	NS	NS	NS	NS	NS	NS

It may be seen from Table 33--abridged that the beef pre-heated to 170° F. changed in bitterness in a parallel manner with the reference beef. No significant differences between beef pre-heated to 160° F. and the reference was observed until after 90 days of storage. Initially, the beef pre-heated to 150° F. was significantly more bitter than the reference. This significance disappeared during 15 days of storage and reappeared only after 45 days of storage. Differences in bitterness between the beef pre-heated to 140° and the reference was not significant until after 60 days of storage. The beef pre-heated to 130° F. showed a significantly less bitterness than the reference beef during 15 days of storage. This significance soon disappeared and did not reappear until after 45 days of storage.

On the basis of the information contained in the foregoing table, it can be stated that, under the conditions of this experiment, beef heated to 130°, 140°, 150°, and 160° F. prior to irradiation to 3 megarads can be stored at 72° F. for 45, 60, 45, and 90 days, respectively, before the panel being able to detect a significant difference in bitterness between these and a reference beef pre-heated to 170° F.

Linearity of Relative Change in Bitterness. In order to see if the bitterness of beef within individual pre-heat treatments has changed in a linear manner with respect to that of the reference beef with time in storage, the test of linearity of regression of flavor scores on time was made using the appropriate statistical method (25, p. 295-298). The results of this test are shown in Tables 34a through e.

It may be seen from Table 34c that the only significant deviation from linearity at 5% level occurred in beef heated to 150° F. prior to irradiation. However, the magnitude of this significance was so small that it disappeared when 1% significance level was chosen. It can be stated, therefore, that the change in bitterness of experimental samples from that of the reference was linear within all levels of pre-irradiation heat treatment.

TABLE 34.
TEST OF LINEARITY OF REGRESSION OF BITTERNESS SCORE ON STORAGE TIME IN IRRADIATED BEEF

Variation Due to	Degrees of Freedom		Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	4.6929	1.060	NS	NS
Error	27		4.4270			
b. Beef pre-heated to 140° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	5.2800	1.243	NS	NS
Error	27		4.2470			
c. Beef pre-heated to 150° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	6.4286	2.681	S	NS
Error	27		2.3978			
d. Beef pre-heated to 160° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	2.8400	1.053	NS	NS
Error	27		2.6970			
e. Beef pre-heated to 170° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	1.6057	.662	NS	NS
Error	27		2.4259			

<u>Rate of Relative Bitterness Change</u>. In order to see the relation among the relative changes in bitterness as affected by the levels of pre-heat treatment, regression coefficients which show the amount of relative change per day of storage were calculated using the pertinent method of calculation (25, p. 268). The results of these calculations are given in Table 35, with the resultant regression equations which have been translated into graphical form in Figure 5.

TABLE 35.
RATE OF CHANGE IN RELATIVE BITTERNESS OF IRRADIATED BEEF AS AFFECTED BY LEVEL OF HEAT TREATMENT

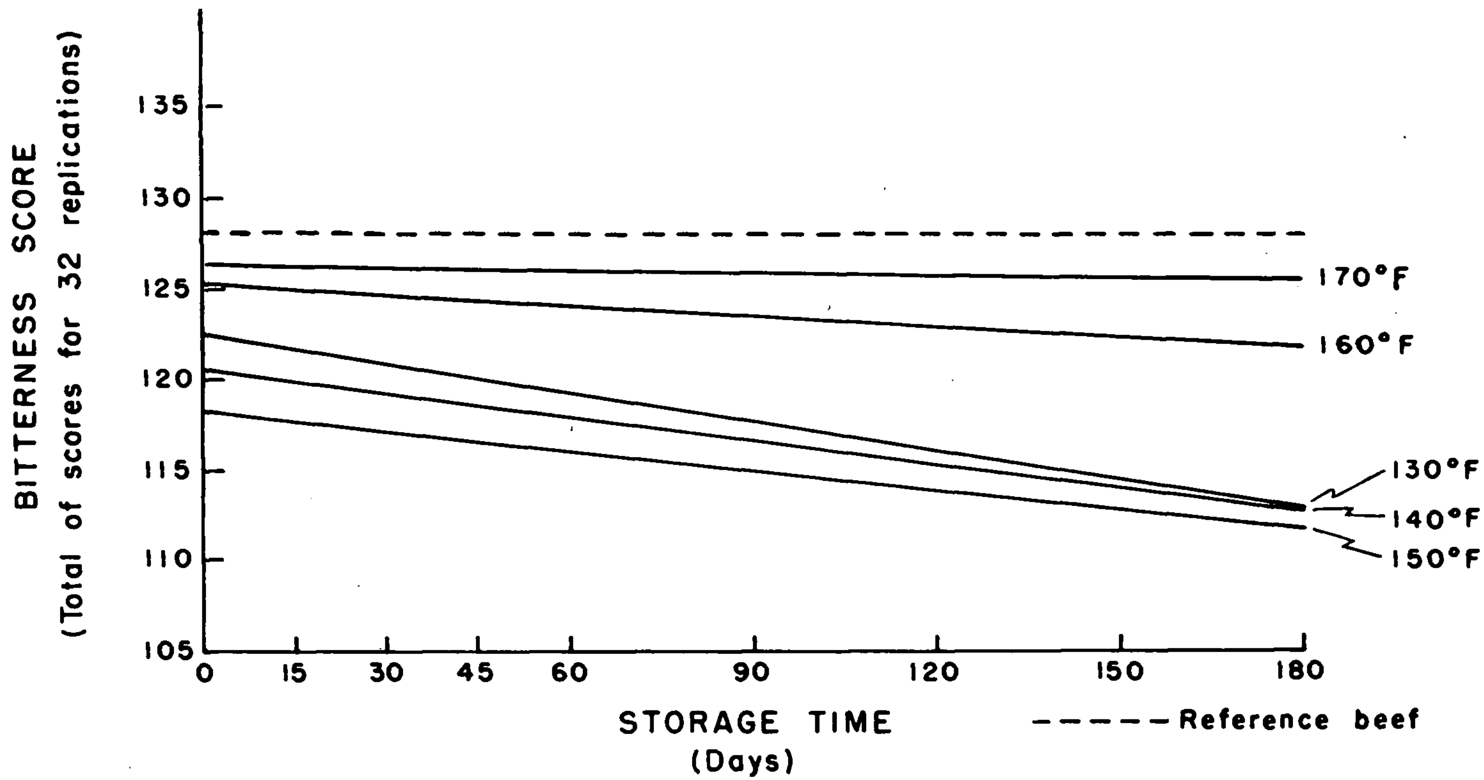
Heat Treatment ° F.	Regression Coefficient*	Regression Equation**
130	-.05259	$\bar{y}_x = 118.5 - .05259\ (x-76.6)$
140	-.04337	$\bar{y}_x = 117.2 - .04337\ (x-76.6)$
150	-.03481	$\bar{y}_x = 115.6 - .03481\ (x-76.6)$
160	-.01827	$\bar{y}_x = 123.8 - .01827\ (x-76.6)$
170	-.00374	$\bar{y}_x = 126.0 - .00374\ (x-76.6)$

* Rate: Change per day in the total of scores for 32 Replications.
** y = Bitterness score (total of 32 replications).
x = Storage time (days).

As may be derived from Table 35 the rates of change in bitterness with respect to reference at the pre-heating temperatures of 130°, 140°, 150°, and 160° F. are 14.1, 11.6, 9.3, and 4.9 times as

FIGURE 5

RATE OF CHANGE IN RELATIVE BITTERNESS OF IRRADIATED BEEF AS AFFECTED BY PRE-HEAT TREATMENTS



that at 170° F., respectively. More informative is a comparison among the ratios of adjacent rates of change in bitterness, which are shown in Table 36.

TABLE 36.
EFFECT OF EACH 10° F. RISE IN PRE-HEATING TEMPERATURE ON THE RATE OF CHANGE IN BITTERNESS OF IRRADIATED BEEF

Level of 10° F. rise In Temperature	Ratio of Adjacent rates*
130 - 140	1.213
140 - 150	1.246
150 - 160	1.905
160 - 170	4.885

* Ratio of the rate in lower temperature to that in the higher one.

As may be seen from Table 36, the 10° F. rise from 160° to 170° caused the highest decrease in the relative rate of change in bitterness. This suggests the presence of a "breaking point" between 160° and 170° F. with reference to the inhibition or destruction of factors contributing towards the increases in the rate of change in relative bitterness. This statement is further supported by the results of a test (25, p. 278-283) carried out to determine the significance of the rates of change in bitterness. These results are shown in Table 37.

TABLE 37.
SIGNIFICANCE OF RATE OF CHANGE IN BITTERNESS
AS AFFECTED BY PRE-HEATING TEMPERATURES

Heat Treat-ment ° F.	Sum of squares (Regression SS) w/1 d.f.	Variance (Residual SS/n-2) w/7 d.f.	$t^2 = F$	Significance At the Level of 5%
130	345.72	17.07	20.25	S
140	235.14	16.38	14.35	S
150	151.47	9.25	16.38	S
160	41.73	10.41	4.01	NS
170	1.75	9.36	.19	NS

The critical region is where the F-value, as derived from F-tables, is larger than 5.5914 with 1 and 7 degrees of freedom. Considering the fact that the F-value for beef pre-heated to 160° F. is much larger than that for the beef pre-heated to 170° F. and also the fact that it is close to the borderline of significance, it can be assumed to be significant for the purpose of comparing these two rates of change in bitterness. On the basis that the 10° F. rise in temperature from 160° to 170° F. rendered a nearly significant rate of change extremely insignificant and, also, considering the fact that the ratio of rate for 160° F. to the rate for 170° F. is the highest of the ratios of any two adjacent rates, it can be stated that a "breaking point" is present between 160° and 170° F. with reference to inhibition or destruction of factors contributing towards the increases in relative rates of change in bitterness.

Overall Desirability

The data pertaining to relative overall desirability of ground lean beef heated to internal temperatures of 130°, 140°, 150°, 160° or 170° F. prior to irradiation to 3 megarads and stored at 72° F. for 0, 15, 30, 45, 60, 90, 120, 150, or 180 days are presented in Table 38.

TABLE 38.
OVERALL DESIRABILITY SCORE OF IRRADIATED BEEF AS AFFECTED BY PRE-IRRADIATION HEAT TREATMENTS AND LENGTH OF STORAGE AT 72° F.
(Means)

Heat Treatment ° F.	Storage Period (days) 0	15	30	45	60	90	120	150	180	Heat Treatment Mean
130	3.8	4.1	4.2	3.9	3.5	3.5	3.6	3.0	3.1	3.7
140	3.7	4.1	4.3	3.9	3.6	3.5	3.3	3.1	2.9	3.6
150	3.4	3.7	3.9	3.9	3.5	3.5	3.2	3.2	3.2	3.5
160	3.8	3.9	4.0	3.9	3.8	3.9	3.7	3.7	3.5	3.8
170	4.3	4.1	4.0	4.0	3.9	4.0	3.8	4.0	3.9	4.0
Storage Period Mean	3.8	4.0	4.1	3.9	3.7	3.7	3.5	3.4	3.3	

The detailed discussion presented under irradiation flavor on page 57 to point out the relativity of the irradiation flavor scores is also applicable to the overall desirability scores shown in Table 38.

Keeping the relativity of these scores in mind, it may be seen from the pre-heating temperature means in Table 38 that the beef pre-heated to 150° F. was the least desirable with respect to the reference beef. The beef pre-heated to 140° F. was more desirable and the one pre-heated to 130° F. was still better in this respect. The beef pre-heated to 160° F. did not seem to be greatly different in overall desirability from the one heated to 170° F. which was rated as being practically the same in this attribute as the reference beef. The table also reveals that the beef pre-heated to 130° F. did not differ appreciably from the reference beef until 60 days of storage when there was a decrease in overall desirability with respect to that of the reference beef and that the decrease became more pronounced at and after 150 days of storage. The same is essentially true for the meat pre-heated to 140° F. The meat pre-heated to 150° F. changed for the better in overall desirability with respect to that of the reference beef up to 60 days of storage when there was a decrease in this attribute with respect to that of the reference and that a further decrease occurred at the 120 days of storage. The beef pre-heated to 160° F. did not show a high degree of difference in overall desirability from that of the reference. The overall desirability of the beef heated to 170° F. was practically the same as the reference beef throughout the storage period.

The above conclusions are derived from the values in Table 38 which represent averages of 32 replications and may not be as definite as the results of statistical analyses based on the individual scores. The results of the statistical analyses are presented in the subsequent paragraphs.

Significance of Difference in Overall Desirability Between the Experimental and Reference Samples. In order to establish whether the panel scores for overall desirability differed significantly from the score assigned to the reference, the data for each combination of pre-heat treatment and storage period were subjected to an analysis using the pertinent statistical method (25, p. 196-208). The results of this analysis are shown in Tables 39a through i.

It may be seen from Table 39--abridged, that the beef pre-heated to 130° F. did not show any significant difference in overall desirability with respect to that of the reference beef until after 45 days of storage when a significant difference was detected by the tasters. Overall desirability of beef pre-heated to 140° or 150° F. was significantly less than that of the reference at the start of the experiment; however, this significance disappeared during 15 days of storage and did not reappear until after 45 days of storage. On this basis, it can be stated that irradiated ground lean beef heated to internal temperatures of 130°, 140°, or 150° F. could be stored for 45 days at 72° F. without a significant difference in overall desirability being detected by the panel between these and the beef heated to 170° F.

TABLE 39.
SIGNIFICANCE OF DIFFERENCE IN OVERALL DESIRABILITY
BETWEEN THE EXPERIMENTAL AND REFERENCE BEEF

Heat Treatment ° F.	Mean Score Minus 4	Sum of Squares	Variance (s^2)	$t^2 = F$ with 1 and 31 Degrees of Freedom	Significance at 5% level	Less Or more Preferable Than Reference
a. Beef stored for 0 day at 72° F.						
130	-.159	30.52	.9845	.8217	NS	
140	-.316	21.70	.7000	4.5648	S	less
150	-.603	47.01	1.5165	7.6726	S	less
160	-.197	45.29	1.4610	.8500	NS	
170	+.300	16.92	.5458	5.2767	S	more
b. Beef stored for 15 days at 72° F.						
130	+.300	25.70	.8290	3.4741	NS	
140	+.069	22.81	.7358	.2071	NS	
150	-.262	30.41	.9810	2.2392	NS	
160	-.087	17.09	.5513	.4393	NS	
170	+.091	16.19	.5223	.5074	NS	
c. Beef stored for 30 days at 72° F.						
130	+.153	19.32	.6232	1.2020	NS	
140	+.309	17.11	.5347	5.7142	S	more
150	-.094	13.50	.4355	.6493	NS	
160	+.022	9.51	.3068	.0505	NS	
170	+.044	10.10	.3258	.1902	NS	
d. Beef stored for 45 days at 72° F.						
130	-.062	31.25	1.0081	.1220	NS	
140	-.062	20.51	.6616	.1859	NS	
150	-.109	12.11	.3906	.9734	NS	
160	-.084	13.76	.4439	.5087	NS	
170	+.019	11.25	.3629	.0318	NS	
e. Beef stored for 60 days at 72° F.						
130	-.519	19.57	.6313	13.6537	S	less
140	-.384	29.22	.9426	5.0059	S	less
150	-.506	18.08	.5832	14.0486	S	less
160	-.234	15.83	.5106	3.4316	NS	
170	-.100	7.30	.2355	1.3588	NS	

Table 39--continued

Heat Treat-ment ° F.	Mean Score Minus 4	Sum of Squares	Variance (s^2)	$t^2 = F$ with 1 and 31 Degrees of Freedom	Signifi-cance at 5% level	Less Or more Preferable Than Reference
f. Beef stored for 90 days at 72° F.						
130	-.481	15.73	.5074	14.5912	S	less
140	-.525	16.20	.5226	16.8772	S	less
150	-.459	15.62	.5039	13.3792	S	less
160	-.131	11.97	.3861	1.4223	NS	
170	+.031	8.59	.2771	.1110	NS	
g. Beef stored for 120 days at 72° F.						
130	-.419	22.71	.7326	7.6685	S	less
140	-.666	23.19	.7481	18.9731	S	less
150	-.844	19.46	.6277	36.3147	S	less
160	-.262	8.55	.2758	7.9645	S	less
170	-.166	13.43	.4332	2.0355	NS	
h. Beef stored for 150 days at 72° F.						
130	-.987	38.11	1.2294	25.3566	S	less
140	-.856	29.78	.9606	24.4093	S	less
150	-.844	18.70	.6032	37.7897	S	less
160	-.291	16.77	.5410	5.0089	S	less
170	-.053	4.86	.1568	.5733	NS	
i. Beef stored for 180 days at 72° F.						
130	-.941	39.64	1.2787	21.5953	S	less
140	-1.073	34.76	1.1213	32.8570	S	less
150	-.841	14.56	.4697	48.1861	S	less
160	-.500	10.88	.3510	22.7920	S	less
170	-.075	10.02	.3232	.5569	NS	

TABLE 39--abridged
SIGNIFICANCE OF DIFFERENCE IN OVERALL DESIRABILITY
BETWEEN THE EXPERIMENTAL AND REFERENCE BEEF
(-S and +S indicates less and more preferable flavor, respectively)

Heat Treatment	Storage Period (days)								
	0	15	30	45	60	90	120	150	180
130	NS	NS	NS	NS	-S	-S	-S	-S	-S
140	-S	NS	+S	NS	-S	-S	-S	-S	-S
150	-S	NS	NS	NS	-S	-S	-S	-S	-S
160	NS	NS	NS	NS	NS	NS	-S	-S	-S
170	+S	NS	NS	NS	NS	NS	NS	NS	NS

The beef pre-heated to 160° F. did not show any decrease in overall desirability with respect to that of the reference beef until after 90 days of storage. On this basis, it can be stated that the beef could be stored for 90 days at 72° F. without the panel being able to detect a significant decrease in overall desirability.

The beef pre-heated to 170° F. was evaluated as being the same as the reference beef which was identical to this experimental beef in all respects.

Linearity of Change in Overall Desirability. In order to determine if the overall desirability of beef within individual pre-heat treatments changed in a linear manner with respect to that of the reference beef with time in storage, the test of linearity of regression of overall desirability scores on time was carried out using the appropriate statistical methods (25, p. 295-298). The results of this test are shown in Tables 40a through e.

TABLE 40.
TEST OF LINEARITY OF REGRESSION OF OVERALL DESIRABILITY SCORE ON STORAGE TIME IN IRRADIATED BEEF

Variation Due to	Degrees of Freedom		Mean Square	F	Significance At the Level of 5%	1%
a. Beef pre-heated to 130° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	13.1143	2.426	S	NS
Error	27		5.4063			
b. Beef pre-heated to 140° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	13.3800	3.263	S	NS
Error	27		4.1000			
c. Beef pre-heated to 150° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	12.1300	4.493	S	S
Error	27		2.7000			
d. Beef pre-heated to 160° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	2.4086	.613	NS	NS
Error	27		3.9270			
e. Beef pre-heated to 170° F.						
Total	35					
Storage time	8					
Linear regression		1				
Deviation from linearity		7	2.6629	1.114	NS	NS
Error	27		2.3907			

It may be seen from Table 40 that the only significant deviation from linearity at 1 percent level occurred in beef heated to 150° F. prior to irradiation. The critical region at 1 percent significance level is where the F-value, as derived from F-tables, is larger than 3.3882 with 7 and 27 degrees of freedom and the F-value for the above deviation from linearity is only 4.493, which shows that the magnitude of significance was not large. It can be stated, therefore, that for all practical purposes the change in overall desirability with respect to that of the reference was linear within all levels of pre-irradiation heat treatment.

Rate of Change in Relative Overall Desirability. In order to determine the relation among the changes in relative overall desirability as affected by the levels of pre-heat treatment, regression coefficients which show the amount of change per day of storage were calculated using the pertinent method of calculation (25, p. 268). The results of these calculations are given in Table 41 with the resultant regression equations which have been translated into graphical form in Figure 6.

As may be derived from Table 41, the rates of change in overall desirability with respect to reference at the pre-heating temperatures of 130°, 140°, 150°, and 160° F. are 4.1, 4.1, 2.4, and 1.2 times as that at 170° F. respectively.

TABLE 41.
RATE OF CHANGE IN RELATIVE OVERALL DESIRABILITY OF IRRADIATED BEEF AS AFFECTED BY LEVEL OF HEAT TREATMENT

Heat Treatment ° F.	Regression Coefficient*	Regression Equation**
130	-.05043	$\bar{y}_x = 116.9 - .05043\ (x-76.6)$
140	-.05005	$\bar{y}_x = 115.5 - .05005\ (x-76.6)$
150	-.02886	$\bar{y}_x = 111.8 - .02886\ (x-76.6)$
160	-.01513	$\bar{y}_x = 121.7 - .01513\ (x-76.6)$
170	-.01228	$\bar{y}_x = 128.3 - .01228\ (x-76.6)$

* Rate: Change per day in the total of scores for 32 replications.
** y = Overall desirability score 9 total of 32 replications.
x = Storage time (days).

More informative is a comparison among the ratios of adjacent rates of change in overall desirability, which are shown in Table 42.

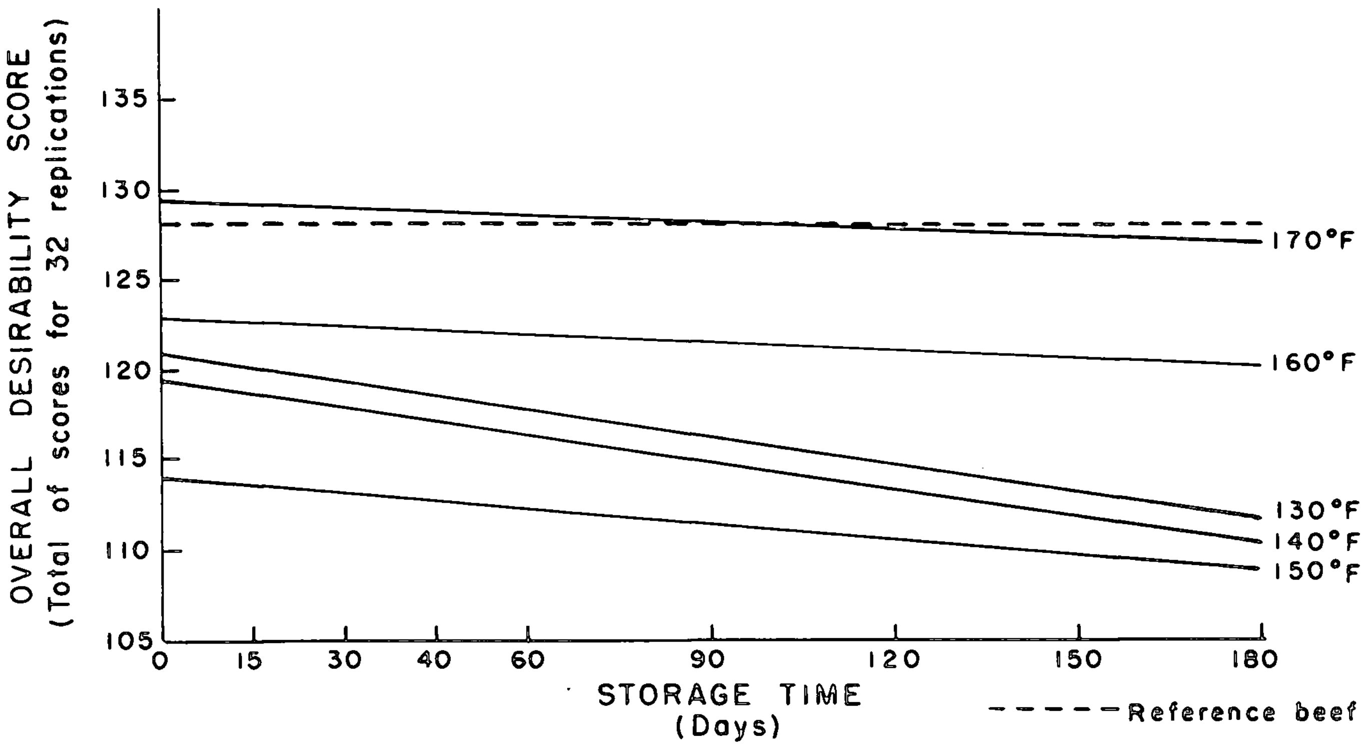
TABLE 42.
EFFECT OF EACH 10° F. RISE IN PRE-HEATING TEMPERATURE ON THE RATE OF CHANGE IN OVERALL DESIRABILITY IN IRRADIATED BEEF

Level of 10° F. rise In Temperature	Ratio of Adjacent rates*
130 - 140	1.008
140 - 150	1.734
150 - 160	1.907
160 - 170	1.232

* Ratio of the rate in lower temperature to that in the higher one.

FIGURE 6

RATE OF CHANGE IN RELATIVE OVERALL DESIRABILITY
OF IRRADIATED BEEF AS AFFECTED
BY PRE-HEAT TREATMENTS



As may be seen from Table 42, the 10° F. rise from 150° to 160° F. caused the highest decrease in the rate of change in relative overall desirability. This suggests the presence of a "breaking point" between 150° and 160° F. with reference to the inhibition or destruction of factors contributing towards the increases in the rates of change in relative overall desirability. This statement is further supported by the results of a test (25, p. 278-283) carried out to determine the significance of rates of change in overall desirability. These results are shown in Table 43.

TABLE 43.
SIGNIFICANCE OF RATE OF CHANGE IN OVERALL DESIRABILITY AS AFFECTED BY PRE-HEATING TEMPERATURES

Heat Treatment ° F.	Sum of squares (Regression SS) w/1 d.f.	Variance (Residual SS/n-2) w/7 d.f.	$t^2 = F$	Significance At the level of 5%
130	317.85	20.85	15.24	S
140	313.07	15.80	19.81	S
150	104.09	10.43	9.98	S
160	28.60	15.15	1.89	NS
170	18.85	9.22	2.04	NS

As may be seen from Table 43, the rate of change in overall desirability was significant in meat pre-heated to 150° F. but this significance disappeared as a result of 10° F. rise in pre-heating temperature. This also shows that there was a "breaking point" somewhere between 150° and 160° F.

Correlation Between Changes in Nitrogenous Constituents and Flavor Attributes.

At the start of the experiment, the flavor attributes of the beef pre-heated to 150° F. were evaluated as being inferior to the beef pre-heated to 140° F., and the latter was rated as being inferior to the one pre-heated to 130° F. (Tables 26, 32, and 38). This relation was generally maintained throughout the storage period. Since there was no storage effect involved at the beginning of the experiment, these differences in the flavor attributes has to be explained in terms of the different effects of the same irradiation dosage on the samples which had been rendered different by the varying levels of pre-heat treatments.

The more heavily emphasized aspect of the experiment, however, was the effect of storage on the beef that received different pre-heat treatments. Therefore, the relation between the chemical and flavor attributes will be discussed in terms of the rates of change over the storage period.

In order to determine the relation between the rates of change in the chemical and flavor attributes, correlation coefficients were calculated using the pertinent statistical method (25, p. 265-266). The results of these calculations are shown in Tables 44a, b, and c.

TABLE 44.
CORRELATION BETWEEN THE CHANGES IN NITROGENOUS CONSTITUENTS AND FLAVOR ATTRIBUTES OF PRE-HEATED IRRADIATED BEEF STORED FOR 180 DAYS AT 72° F.

Pre-heating Temperature	Irradiation Flavor	Bitterness	Overall Desirability
a. Correlation between total soluble nitrogen and flavor attributes*			
130	-.83	-.91	-.85
140	-.90	-.85	-.81
150	-.73	-.87	-.76
160	-.54	-.43	-.60
170	-.52	-.55	-.71
b. Correlation between TCA-soluble nitrogen and flavor attributes*			
130	-.78	-.86	-.79
140	-.84	-.79	-.74
150	-.62	-.83	-.67
160	-.39	-.53	-.66
170	+.32	+.45	+.01
c. Correlation between amino nitrogen and flavor attributes*			
130	-.79	-.88	-.82
140	-.87	-.83	-.79
150	-.60	-.83	-.66
160	-.00	-.03	-.43
170	+.40	+.60	+.49

* Correlation coefficients significant at 5% level where the absolute value is larger than .67.

As may be seen from Table 44, significant negative correlations existed between the changes in the flavor attributes and nitrogenous constituents of beef pre-heated to 130°, 140°, and 150° F. The negativity of the correlations indicate that the flavor became less preferable as the amount of the nitrogenous constituents increased.

The poor correlations observed in the samples pre-heated to 160° or 170° F. can be explained on the basis of the insignificance of the increases in the nitrogenous constituents and/or the small magnitude of differences in flavor with respect to the reference beef. It will be remembered that the rate of increase in nitrogenous constituents in beef pre-heated to these temperatures were found to be not significant (Tables 9, 17, and 25). Neither were the rates of change in flavor attributes significant in beef pre-heated to these temperatures (Tables 31, 37, and 43). It was the irregular fluctuations, although of very small magnitude, displayed by the results of the chemical and flavor analyses in these samples that caused these poor correlations. It may be concluded, therefore, that the nitrogenous constituents of the beef pre-heated to 160° or 170° F. had been more or less stabilized and the flavor attributes of the same followed a parallel change with respect to that of the reference.

<u>The Relation Between the Changes in Nitrogenous Constituents and Irradiation Flavor in Beef Pre-heated to 130°, 140°, and 150° F.</u> Tables 44a, b, and c show a strikingly regular pattern with regard to the relation between the changes in the nitrogenous constituents and the relative flavor attributes of the beef pre-heated to 130°, 140°, and 150° F. In all cases, the closest relation between the changes in the nitrogenous constituents and those in the irradiation flavor occurred in beef pre-heated to 140° F. This was followed by beef pre-heated to 130° and 150° F. The relatively poor correlation in beef pre-heated to 150° F. can be explained on the basis of the

fact that the irradiation flavor of this beef with respect to that of the reference was more or less stabilized (Figure 4), while the nitrogenous constituents still showed a relatively high increase (Figures 1, 2, and 3). The rates of change in irradiation flavor were almost equal in beef pre-heated to 130° and 140° F. (Figure 4). The difference between the two correlation coefficients was caused by the difference in the rates of increase in nitrogenous constituents in the samples pre-heated to these temperatures (Figures 1, 2, and 3).

The implication of the preceding paragraph is that, although there was a high degree of correlation between the rates of change in the chemical and flavor attributes <u>within</u> temperatures, such a relation did not exist <u>among</u> temperatures. It can be concluded, therefore, that the irradiation flavor of beef was not a function of the nitrogenous constituents <u>alone</u> but other factors also contributed to the changes in this attribute. As long as the pre-heating temperatures of 130°, 140°, and 150° F. are concerned, regularly different pre-heating temperatures caused correspondingly regular changes in the nitrogenous constituents during storage, whereas this regularity was not observed in irradiation flavor. A comparison of Figure 4 against Figures 1, 2, and 3 provides the best illustration of this statement.

<u>The Relation Between the Changes in the Nitrogenous Constituents and Bitterness in Beef Pre-heated to 130°, 140°, and 150° F.</u> As may be seen from Tables 44a, b, and c, the highest correlations between the changes in the chemical attributes and the bitterness occurred

in beef pre-heated to 130° F. The correlations in beef pre-heated to 140° and 150° F. were not highly different from those for the beef pre-heated to 130° F. On this basis it can be stated that the changes in bitterness in pre-heated irradiated beef followed a more or less regular pattern as those in the nitrogenous constituents. A comparison between Figure 5 and Figures 1, 2, and 3 illustrates this statement.

The Relation Between the Changes in Nitrogenous Constituents and Overall Desirability in Beef Pre-heated to 130°, 140°, and 150° F. Tables 44a, b, and c show that the highest correlations between the changes in the nitrogenous constituents and those in overall desirability occurred in beef pre-heated to 130° F. Although the beef pre-heated to 130° F. did not differ greatly from the one heated to 140° F., there was a marked loss of correlation in beef pre-heated to 150° F. This wide difference observed in the correlation coefficient for beef pre-heated to 150° F. is indicative of the effect of irradiation flavor in the judgment of the overall desirability. A comparison of Figures 4, 5, and 6 shows very clearly that in the judgment of overall desirability, irradiation flavor exerted a more dominant effect on the tasters.

In summary, it can be stated that there was a relatively regular relation between the changes in bitterness and those in the total water-soluble, TCA-soluble, and amino nitrogen in beef pre-heated to 130°, 140°, and 150° F. In the case of irradiation flavor and overall desirability this relation among pre-heating temperatures diminished.

Irradiation flavor exerted a more dominant effect on tasters than bitterness in their evaluation of overall desirability.

In the great majority of cases, total soluble nitrogen correlated better than amino nitrogen and the latter better than the TCA-soluble nitrogen with the flavor attributes.

In beef pre-heated to 160° or 170° F., poor correlations between the changes in the nitrogenous constituents and those of the relative flavor attributes were observed which can be explained on the basis of the insignificant but irregular changes in the nitrogenous constituents and/or the small but irregular differences in flavor attributes with respect to reference beef.

SUMMARY AND CONCLUSIONS

The total water-soluble, TCA-soluble, and amino nitrogen contents and the relative irradiation flavor, bitterness and overall desirability of ground lean beef pre-heated to internal temperatures of 130°, 140°, 150°, 160°, and 170° F. prior to irradiation to 3 megarads, showed the following changes and relations during storage at 72° F. for periods up to 180 days:

(1) The nitrogenous constituents in beef heated to 130°, 140°, or 150° F. before irradiation increased at significant rates with time in storage. The rates of increase at the lower pre-heating temperatures were higher than those at the higher ones. Although changes of small magnitude were observed in beef pre-heated to 160° or 170° F., the rates of change were found to be insignificant.

On the basis of these findings it was concluded that, for all practical purposes, the factors contributing toward increases in the nitrogenous constituents in irradiated ground lean beef in storage are inhibited or destroyed as a result of heating to an internal temperature of 160° F. or above prior to irradiation.

(2) Initially, the relative flavor characteristics of beef pre-heated to 140° or 150° F. were inferior from those pre-heated to 130°, 160°, or 170° F., the one pre-heated to 150° F. being more inferior. However, an improvement in flavor was observed in these samples during the first 45 days of storage so that the relative flavor characteristics of all the variously heat-treated beef were

evaluated as being the same as those of the reference at the 45th day of storage. Thereafter, a consistent decrease was observed in the relative flavor characteristics of beef pre-heated to 130°, 140°, or 150° F. In beef pre-heated to 160° F., this decrease in flavor was not observed until after 60 days of storage and the one pre-heated to 170° F. had the same flavor as the reference throughout the storage period.

An evaluation of the data to present a general picture for the entire storage period showed that the pre-heating temperatures less than 160° F. appeared to give way to the development of different degrees of inferiority in the relative flavor, and this inferiority followed the order of 150°, 140° and 130° F., the last being the least inferior. It appeared also that the relative irradiation flavor of the beef pre-heated to 150° F. was more or less stabilized at the same relative level over 180 days of storage at 72° F.

On the basis of these findings it was concluded that:

(a) The beef pre-heated to 130° F. or 160° F. could be stored for 45 and 60 days, respectively, without the panel being able to detect a flavor different from that of the beef pre-heated to 170° F.

(b) Initially, the beef pre-heated to 140° or 150° F. would have an inferior flavor with respect to that of the beef pre-heated to 170° F., but that the flavor would improve until the 45th day of storage when the panel would not be able to detect a flavor different from that of the beef pre-heated to 170° F.

(c) The pre-heating temperatures of 150° and 140° F. produced more intensive adverse relative flavor changes than either 130° or 160° F.

(3) An evaluation of the relation between the changes in nitrogenous constituents and those in flavor attributes provided the following conclusions:

(a) Significant correlations existed between the increases in the nitrogenous constituents and the decreases in the relative flavor attributes in beef pre-heated to 130°, 140°, and 150° F.

(b) The highest correlations between the changes in nitrogenous constituents and those in relative irradiation flavor existed in beef pre-heated to 140° F. In the case of bitterness and overall desirability, the highest correlations were obtained in beef pre-heated to 130° F.

(c) A comparison among the correlations between the changes in nitrogenous constituents and those in the relative irradiation flavor suggested that the changes in the relative irradiation flavor was not a function of the changes in nitrogenous constituents alone. A similar comparison for relative bitterness suggested that the changes in the nitrogenous constituents and bitterness followed a more or less regular pattern.

(d) Irradiation flavor seemed to exert a more dominant effect on the tasters in their judgment of overall desirability.

(e) In beef pre-heated to 160° or 170° F., poor correlations between the changes in the nitrogenous constituents and the relative flavor attributes were observed which was explained on the basis of the insignificant but irregular changes in the nitrogenous constituents and/or the small but irregular differences in flavor attributes.

(f) In the great majority of cases, total soluble nitrogen correlated better than amino nitrogen and the latter better than TCA-soluble nitrogen with the flavor attributes.

RECOMMENDATIONS

It is recommended that:

(1) In order to prevent increases in total water-soluble, TCA-soluble, or amino nitrogen for 180 days of storage at room temperature, ground lean beef should be heated to an internal temperature of not less than 160° F., preferably not less than 170° F., prior to irradiation to 3 megarads.

(2) In order that no differences in flavor attributes between ground lean beef heated to 170° F. prior to irradiation to 3 megarads and the beef pre-heated to lower temperatures should be observed, the latter should not be stored at room temperature for more than 45 days if pre-heated to 130°, 140°, or 150° F., not more than 60 days if pre-heated to 160° F., not less than 45 days if pre-heated to 150° F., and not less than 15 days if pre-heated to 140° F.

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