The effects of low dose irradiation, storage time, and package type on the sensory attributes and color of ground beef patties

by

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#### GENERAL INTRODUCTION

Fresh meat is a very highly perishable food product. Under normal aerobic conditions, the shelf-life of refrigerated fresh meat is limited by the growth of aerobic and psychrotropic strains of bacteria. Besides proper storage temperature, other control methods for reducing microbiological problems include modified atmosphere packaging, chemical decontamination, and ionizing radiation after packaging. Combinations of biochemical proteolysis and micro-organism growth can result in detrimental colors, odors, texture, and flavors.

When an activated orbiting electron leaves an atom, chemical changes result within the molecules called ionization. The process of ionization results in the formation of positively charged atoms known as cations (positive ions), which are formed by loosing a negatively charged electron. The lost electron is trapped by surrounding atoms, forming negatively charged ions (anions). Ionization forms highly reactive atoms and molecules called free radicals. A minimal fraction of the absorbed energy of radiation is available to be converted to thermal energy. With low dose irradiation there are minimal heat transfers allowing the typical sensory and nutritional properties of meats to be largely preserved.

There are two major sources of radiation used in the food irradiation process. The first is a gamma radiation facility or a radioisotope source. Gamma rays result from a radioactive source such as cobalt 60 ( $Co^{60}$ ) and cesium 137 ( $Cs^{137}$ ). Gamma ( $\gamma$ ) rays have a deep penetrating ability when

compared to electron beam or linear accelerator facilities. Thus, products by the pallet load can be irradiated by gamma ray facilities. While gamma ray facilities have deep penetrating abilities they have low dose rates.

Consequently, hours can be spent on products being irradiated with gamma rays. With charged particle accelerators, the second type of facility, Van der Graaff accelerators and X-ray generators may be employed. Van der Graaff accelerators produce  $\beta$  (beta) rays which have substantially less penetrating abilities (approximately 3 inches with meat products) when compared to gamma rays. X-rays have deep penetrating abilities as do gamma rays. Typically, linear accelerators (a Van der Graaff accelerator) can be configured with a stainless steal or tungsten target to produce X-rays. Unfortunately, when this process is used there is a dramatic reduction in the accelerator's power efficiency. Thus, the use of linear accelerators in producing X-rays is very inefficient. Nevertheless, both X-ray generators and Van der Graaff accelerators have very high dose rates allowing products to spend a matter of minutes being irradiated.

The unit by which absorbed levels of radiation are measured include the rad (radiation absorbed dose) and Gray (Gy). A rad is the amount of energy required for one gram of matter to absorb 100 ergs of energy. Typically, researchers today use the Gy measurement instead of a rad. The Gray is defined as the absorption of 1 Joule (1 Joule = 10 million ergs) of energy by each Kilogram of matter being irradiated. The gray is equal to 100 rad, and 1000 Gy is equal to 1 kGy (kiloGray).

The interaction of radiation energy with flexible packaging materials forms gas and volatiles. Consequently, irradiation of polymers can change the mechanical and physical properties of films. In the presence of oxygen, as irradiation occurs on a commercial basis, irradiation produces oxidative degradation reactions within the films. Ionizing radiation forms a variety of molecules known as radiolytic compounds (some of which are free radicals) as a result of chain scission within the carbon chains of the polymers involved. The properties of ionizing radiation may also generated long lived free radicals in the packaging materials which could conceivably contribute to subsequent reactions in the packaging material or presumably even in the food. Thus, migration of radiolytic compounds, the production of off-odors and off-flavors, as well as taint transfer play an important role in irradiation processing of prepackaged fresh meats.

Factors which affect the shelf life of meats include holding temperature, atmospheric oxygen, moisture, light, and micro-organisms present. Problems in the life and acceptance of fresh meats may arise from spoilage bacteria, pathogenic bacteria, molds, and yeasts. The inactivation of food spoilage micro-organisms with irradiation occurs through changes in the deoxyribonucleic acid (DNA) molecules in living cells. The DNA molecules in living cells are more sensitive to radiation than the larger molecules of food because of the small size of DNA molecules.

The major spoilage bacteria of meats are gram-negative and include aerobic, psychrotropic strains of *Pseudomonas, Moraxella, Aeromonas, Acinetobacter*, and the facultative anaerobe, *Alxeromonas putrefaciens*.

Mesophilic bacteria of significance to consumers from fresh meats include Salmonella, Staphylococcus aureus, Yersina enterocolitica, Clostridium botulinum, Clostridium perfringens, Campylobacter, Aeromonas hydrophila, and Listeria monocytogenes. Escherichia coli (E. coli) 0157:H7 is a faculatively anaerobic, gram negative bacteria which is considered an adulterant in fresh meats because it causes hemorraghic colitis in humans. While the growth of these pathogenic micro-organisms is limited at normal refrigerated storage conditions, they pose a potential public health threat if meat is temperature abused. Gram positive lactobacillus which may lead to spoilage, are also found in fresh ground beef. Even though irradiation has been proven to reduce spoilage and pathogenic bacteria there are concerns by some researchers that pathogens may increase in irradiated meats because of a lack of competing organisms. This may also allow growth of pathogenic bacteria and toxin production without normal signs of spoilage, as with non-irradiated spoiled fresh meats.

Radiation of meats has also been shown to cause sensory changes in fresh meats. Irradiation can cause discoloration of fresh meats as well as numerous off-odors and off-flavors. The degree of organoleptic changes in meats is dependent upon package type used, absorbed dose, temperature during irradiation, the presence of oxygen, and the age of the meats being irradiated. Irradiation induced oxidation, proteolysis, and free radical production all can lead to products causing consumer concerns with irradiated products. Advantages of preserving foods using irradiation have been noted by Urbain (1989) to include decontaminating foods, controlling

maturation, altering chemical composition, maintaining sensory qualities to a large extent, and no toxicological residue production.

On the whole, low dose irradiation (1 to 10 kGy) has proven to effectively eliminate microbiological sources of contamination to the consumer, while not having a serious effect on the sensory properties of food products. After extensive research in 1981 the WHO reported low dose irradiation proved no serious toxicological hazard to human beings. Nonetheless, before irradiation will be used on a large commercial scale key issues must be studied. A combination of issues like the effects of storage time, packaging, and irradiation on the sensory qualities of fresh beef and other meats need to be addressed. Also, consumer acceptance of irradiated products must be further studied. Consumer studies indicate a growing support for irradiated foods and the public willingness to buy irradiated products increases if they are properly educated (Bruhn, 1995; Lagunas-Solar, 1994; Pszczola, 1993; Resurreccion et al., 1995). Consequently, this study deals with the effects of low dose irradiation, package type, and storage times on the sensory attributes of ground beef.

## Thesis organization

This thesis is in an alternate style format consisting of a general review of literature, two papers prepared for publication, and a concluding summary. The two papers represent the work done by the first author to fulfill requirements for the degree of Masters of Science. The first two papers were prepared according to the Journal of Food Science style guide. These papers

consist of an Abstract, Introduction, Materials and Methods, Results and Discussion, Conclusions, and References.

#### **GENERAL REVIEW OF LITERATURE**

#### **Effects of Ionizing Radiation on Plastic Food Packaging Materials**

Foodstuffs to be treated with ionizing radiation are typically packaged in single or multilayer films prior to irradiation to prevent recontamination. Irradiated packaging materials are also used in aseptic processing lines to produce a sterile package in which thermal sterilized foods may be packaged to produce a shelf stable product. Ionizing radiation is also used in the sterilization of medical and pharmaceutical products as well as a final process in the production of many polymer compounds.

Irradiation of polymers has been shown to produce physical changes in polymers, such as the simultaneous scission and cross-linking of polymer chains, the formation of gases and volatile products which may migrate into foodstuffs (global radiolytic migration) and to cause off-odors and off-flavors. Factors which influence the capacity of a flexible film to be a useful product in the irradiation of meats include, radiation induced changes in the properties of plastic packaging materials should not impair the function of the packaging material, durability of the package, and the capacity of the package to withstand irradiation.

In the presence of oxygen there are additional oxidative chain scission and oxidation of the polymer, resulting in the formation of peroxides, alcohols, carbonyls, carbon dioxide, and carbon monoxide. Radiation induced changes on the polymer are also dependent upon the type of polymer, additives used in the plastic film, the processing history of the films, and irradiation conditions (Buchalla et al. 1992). Lastly, for packaging materials to be useful, radiolytic

degradation products should neither be toxic, global migration values should not increase significantly, and the packaging materials should hold the food without severely affecting the sensory qualities of the foodstuffs.

#### **Gaseous Radiolysis Products**

The production of gases during irradiation of polymers and plastic films has been well documented. The literature in this area can be grouped into two categories, the first being irradiation in the absence of oxygen and the second being in the presence of air or oxygen. The major gas products in vacuum are hydrogen (H<sub>2</sub>), methane (CH<sub>4</sub>), and hydrogen chloride (HCl) for chlorine containing films. In the presence of air irradiation produces carbon dioxide (CO<sub>2</sub>) and carbon monoxide (CO) in larger quantities than in a vacuum, as well as H<sub>2</sub> and CH<sub>4</sub> and other hydrocarbons. Typically, the amounts of gases produced during irradiation of plastics enlarges with increasing absorbed doses.

The amount of gases produced has been related to G values which have been defined by Charlesby (1960) as the quantity of chemical changes of a given kind produced per absorbed dose. G values, or radiolytic yields, have been reported by numerous researchers (Charlesby, 1960; Hegazy et al. 1981a and 1981b; and Killoran 1972) to increase with increasing doses. In studying the effects of irradiation on polypropylene Hegazy et al. (1981a) found that gas evolution or production extended with an increasing dose, but leveled off at extremely high doses (in excess of 300 kGy).

While  $H_2$ ,  $CH_4$ ,  $CO_2$ , CO, and HCl are the major gases formed by irradiation of plastic films in vacuum and air, there are several other

hydrocarbons produced. When polymer films were irradiated under vacuum  $H_2$  and  $CO_2$  were the major gases formed as well as 90 other hydrocarbons (Killoran, 1972). Hegazy et al. (1981a) reported 95 percent of the gases evolved after irradiation of polypropylene in vacuum were  $H_2$ , 3 percent methane, and several other hydrocarbons were detected. G values of more than 1000 different hydrocarbons and compounds of irradiated polymers have been noted (Charlesby, 1960).

Although oxygen and dose have marked affects on the quantity and quality of radiolytic yields (G values), other factors such as the history of the product and temperature at the time of irradiation can have effects on G values. For instance, G values of  $H_2$  decrease slightly with heavier molecular weights of polymers, yet temperature has little affect (Charlesby, 1960). Bersch (1959) found irradiation of plastic films in air and in vacuum resulted in different gaseous products being formed and the irradiation process in air resulted in a greater amount of radiolytic compounds.

It has been concluded that the production of  $H_2$  by irradiation is a result of cross-linking and increased unsaturation of the polymeric chains within the plastics (Charlesby, 1960). The interlinking of polymer chains, known as cross-linking, must result in the production of  $H_2$  to produce the chemical bond. Hegazy et al. (1981b) reported  $H_2$  and  $CH_4$  increase linearly while CO and  $CO_2$  level off at very high doses (in excess of 300 kGy) when plastercized poly(vinyl chloride) is irradiated. The authors went on to say that when poly(vinyl chloride) is irradiated under vacuum HCl is the main product, and  $H_2$ ,  $CO_2$ , CO, and  $CH_4$  are minor products.

Polymers are formed by the connection of very long carbon chains. If these chains are packed very tightly in a form where the chains are nearly

parallel in an ordered arrangement, such as the layering of bricks, the film has a higher density. For example, polyethylene follows such a make-up in its formation. To make plastics less rigid and more flexible, sometimes plasticizers are added. Sometimes polymer chains have side chains, such as polypropylene. The side chains interfere in the close packing of the molecules and result in lower density materials. Hegazy et al. (1981b) found G values for plasticized poly(vinyl chloride) irradiated under vacuum are lower than pure unplasticized poly(vinyl chloride). The production of higher G values by polymers with side chains in contrast to those without side chains at the same absorbed dose has also been noted. A greater production of hydrocarbons under all conditions by low density materials was concluded by Hegazy et al. (1981a) to be due to the polymers having a more highly branched polymer network.

When plastics are irradiated in a vacuum the oxygen remaining in the product can have an effect on gas production, degradation of the polymers, and oxidation in a process where oxygen present becomes the limiting factor. Hegazy et al. (1981a) reported the formation of CO and  $CO_2$  of irradiated polypropylene in vacuum was due to oxygen remaining in the sample. It has also been concluded the formation of  $CO_2$ , CO, and water (H<sub>2</sub>O) indicated the film contained oxygen, either absorbed or combined prior to irradiation, when samples were irradiated in vacuum (Bersch et al. 1959).

Ordinarily, researchers found total G values and G values of  $H_2$ increased when the irradiation of plastics occurs in the presence of air (oxygen). Because commercial irradiation of food products does not take place in evacuated tubes, irradiation processes in the presence of air or oxygen (O<sub>2</sub>) are more applicable to the commercial irradiation of meats in flexible plastic packaging. Hegazy et al. (1981b) noted that  $H_2$  increased in production during irradiation of poly(vinyl chloride) at the same dose when irradiation occurred in the presence of  $O_2$  instead of in a vacuum. Bersch et al. (1959) reported that the irradiation of plastic films in the presence of air and in vacuum resulted in different products with the production of  $H_2O$ ,  $CO_2$ , CO, and HCl being produced in the presence of air.

Gas evolution increases in the presence of  $O_2$  and at a linearly rate at increasing low doses and eventually levels off at extremely high doses in excess of 300 and 400 kGy (Hegazy et al. 1981b). Again the leveling off of gases may be due to  $O_2$  being a rate limiting product in the chemical reaction. Hegazy et al. (1981a and 1981b) also observed  $O_2$  increased the formation of  $CO_2$ , CO,  $CH_4$ ,  $H_2$ , and other hydrocarbons.

Other factors which play a role in the oxidative process of polymers and the formation of gases are oxygen consumption and oxygen pressure. Typically as oxygen pressure and consumption increase so does the production of radiolytic and gaseous products. Hegazy et al. (1981a and 1981b) reported that oxygen consumption increased linearly as dose increases at low doses while it levels off at higher doses. While the consumption of  $O_2$  increases so does the production of  $CH_4$  and HCl. Oxygen consumption also increases with increasing pressure (Hegazy et al. 1981a). Furthermore Hegazy et al. (1981b) noted that oxygen consumption of poly(vinyl chloride) was dependent upon oxygen pressure. Thus, the higher the  $O_2$  pressure the larger the amount of gases formed and  $O_2$  consumed.

Oxygen accelerates the degradative reactions by peroxidation of polymer chains followed by decomposition and rearrangement (Charlesby, 1960). The oxidation of polymers can have an adverse effect on the physical and mechanical properties of plastic films. During irradiation  $O_2$  is absorbed and reacts with the polymer (Bersch et al. 1959). Thus, not only does oxygen within the plastics increase degradative reactions, irradiation within the presence of  $O_2$  increases degradation of the plastics. Hegazy et al. (1981a) noted  $O_2$ consumption in films rather than powders was lower, therefore oxidation was controlled by diffusion. Moreover, most of the  $O_2$  reacted with the polypropylene powder to form polymeric oxidation products. These oxidative products can then further react with the polymers as well as meat and food products packaged within the films.

Ranking of packaging films based on the total amounts of gaseous radiolytic products can be used to show the degree of degradation in each film. Bersch et al. (1959) and Killoran (1972) ranked plastics in the orders of polystyrene, polyvinylidene chlorides, polyvinyl chloride, and polyethylenes, based on radiation stability from most to least of the films. Buchalla et al. (1993) also used radiolytic products accordingly to rank the films on radiation stability with polystyrene, polyethylene terephthalate, rubber hydrochloride the most stable materials, followed by polyamides (also known as nylon), poly(vinylidene chloride), and polycarbonate, which were less stable than the former, but considerably more stable than the polyolefines (polyethylenes and polypropylenes).

## Volatile Radiolysis Products

When packaging films are exposed to ionizing radiation volatile compounds are produced. The amount of volatile compounds produced typically increases when irradiation occurs in the presence of air and with increasing absorbed doses. The amounts of volatiles produced are predominantly hydrocarbons, alcohols, aldehydes, ketones, and carboxylic acids when irradiation takes place in air or the presence of oxygen. Rojas De Gante and Pascat (1990), in addition to Thayer (1988), noted the emission of volatile compounds such as hydrocarbons,  $CH_4$ , HCl, ketones, and aldehydes after ionization of flexible packaging was a function of the dose level, atmosphere surrounding the process, temperature of ionization, and the formulation of the plastic. It was concluded the amount of volatiles from unirradiated films was much less than that from irradiated films, and most of the volatiles from irradiated films were produced by irradiation (Azuma et al. 1983). Matsui et al. (1990) also reported the increase in the production of volatiles as the absorbed dose increased for films.

The main volatiles produced in low-density polyethylene and other films with irradiation are hydrocarbons,  $H_2$ ,  $CO_2$ , water, CO, aldehydes, ketones, carboxylic acids, and numerous other organic and inorganic compounds. Irradiation of low-density polyethylene produced over 100 different volatiles and 58 volatiles in polypropylene (Rojas De Gante and Pascat 1990; and Azuma et al., 1983). Also, Killoran (1972) found 72 different volatiles were produced in the irradiation of various films. Azuma et al. (1983) in quantify volatiles, reported aldehydes and ketones accounted for 26% of all the volatiles produced in polyethylene film irradiated with electron beam and carboxylic acids accounted for 18% of the total volatiles produced.

In general, it is estimated that the volatiles from polyethylene film are intermediate products formed in the polymerization process of ethylene or products formed by the thermal degradation of the polymer (Azuma et al. 1983). These authors went on to state a large amounts of aliphatic hydrocarbons were detected in the irradiated film, and these could be considered to be breakdown products of the polyethylene chains by irradiation with the high energy electron beams. Aliphatic carbons are contained within the branches of the polymer and not in the primary chain.

The degradation of polymers not only leads to volatile products, but can lead to adverse mechanical properties. Thus, one can use the amount of volatiles released to compare various films to produce a multilayered film based on optimal physical and chemical properties. Incidentally, the amount of volatiles evolved after ionization is higher in branched polymers than in linear structures. Also, the amounts of gas evolved are higher in polypropylene than they are in low density polyethylene (Rojas De Gante and Pascat, 1990). Keay (1968) indicated a marked increase in the amounts of volatiles in polypropylene in contrast to polyester-polyethylene and nylon.

The degradation of polymers during irradiation which leads to the production of volatile gases can be effected by a variety of factors. While dose and the presence of oxygen, which leads to oxidation, have substantial effects on the amounts of volatiles produced, temperature during irradiation, and additives in the formulation of the film can also have an effect. Azuma et al. (1984a) concluded the amounts of carboxylic acids and other volatiles produced by electron beam irradiation varied considerably depending upon the properties of the resin, temperature of the film formation, or the presence of antioxidants. Rojas De Gante and Pascat (1990) also recorded the amount of volatile products formed depends on the formulation of the film and processing history of the sample.

Lower irradiation temperatures of polymers results in lower productions of volatile gases. Azuma et al. (1984a and 1984b) reported a low irradiation temperature of -75°C in comparison to 80°C was effective in the lowering of the amounts of volatiles produced by irradiation. Krylova et al. (1979) also noted the use of plasticizers lowered the quantity of volatiles being produced while the plasticizer molecules break down and form mainly monoalkyl esters of phthalic acid. Thus, the route of degradation of polymers during irradiation can be changed to produce lower quantities of volatiles through the use of additives.

The major problem with volatile compounds being produced in flexible packaging are the off-odors attributed to volatile compounds such as ketones, aldehydes, and carboxylic acids. Carboxylic acids, for instance acetic, propionic, and n-butyric acids, released from irradiated polyethylene films, have been used as indicators of the intensity of off-odors. Antioxidants may also reduce the amounts of carboxylic acids which give the strongest off-odors.

Oxygen and the presence of air during irradiation has been shown to be the major cause from the formation of  $H_2$  in polymers during irradiation in vacuum to the production of more volatile compounds. Rojas De Gante and Pascat (1990) showed that the amounts of volatiles increased with increases in absorbed doses or oxygen concentrations during irradiation. Azuma et al. (1984a and 1984b) noted the lack of  $O_2$  during irradiation lowered the amounts of carboxylic acids and other carbonyl compounds formed during ionization processing. It has also been reported that during irradiation  $O_2$  is absorbed and reacts with poly(vinyl chloride) while inhibiting the formation of  $H_2$ , HCl, and other hydrocarbons (Bersch et al., 1959). Therefore oxygen acts to reduce the amount of non-volatile gases such as  $H_2$  and increases the amounts of volatiles and the degradation process of polymers during irradiation. Azuma et al (1984b) and Rojas De Gante and Pascat (1990) both indicated oxygen acts to inhibit the normal cross-linking reaction in polymers like polyethylene and to increase the number of main chain breaks during irradiation. This is particularly true for polymers which are easily oxidized such as polypropylene.

Azuma et al. (1984b) also reported the amount of volatiles produced by films during irradiation with gamma rays from CO<sup>60</sup> were larger than the amounts with electron beam irradiation at 20 kGy. A greater amount of oxygen molecules which form peroxidation radicals are supplied during longer irradiation times as is such with gamma irradiation. Also electron beam irradiation is carried out at a much higher dose rate where oxidation might occur with more difficulty because recombination of primary radicals is more favored than peroxidation. Krylova et al. (1979) found an initial increase in the amounts of carbonyl groups in poly(vinyl chloride) may be explained by the fact that in low dose of irradiation, polymer oxidation takes place at a higher rates than the elimination of HCl.

Degradative or oxidative reactions can occur in vacuum or in the presence of air but because of the increased  $O_2$  supply in the presence of air the amount of oxidative products and volatiles produced increases. Rojas De Gante and Pascat (1990) concluded the major volatiles identified, such as ketones, aldehydes, alcohols, and carboxylic acids, with the irradiation of flexible packaging films were final oxidation products. Azuma et al. (1983) also concluded the volatiles produced were considered to be oxidation products resulting from the reaction with  $O_2$  in air during irradiation with electron beams. Lastly, the same type of products have been observed at low doses as at high doses, proving that the degradation process in polymers does occur at low dose irradiation and volatile compounds were formed (Rojas De Gante and Pascat, 1990).

## **Mechanical and Physical Changes**

The exposure of polymers to ionizing radiation chemically causes the development of cross-linking between polymer and therefore typically increases tensile, tear, and impact strengths of films. Consequently numerous polymer film producers use irradiation to cross-link the films to form a more durable product. Wang et al. (1993) noted surface irradiation of food packaging at 30 to 120 kGy increased cross-linking of the materials.

Ionizing radiation of polymers also can form chain scission breaks within the films, and as a result gases, volatiles, and other radicals, especially in the presence of oxygen are formed. The chain scission breaks have also been associated with the degradation of polymers and alteration of the physical and mechanical properties of films. For instance, the mechanical properties of isotactic polypropylene degrade to a large extent with irradiation at a dose of 20 to 30 kGy, where other films have enhanced mechanical properties at the same dose (Hegazy et al., 1981a). The formation of stress-cracking, crystalinity of the polymers, and the gas permeability of the film can be influenced with irradiation depending upon the dose rate and properties of the polymers involved.

Chain scission involves random rupturing of the molecular bonds of the material, thus leading to the formation of short-chain polymers, evolution of gases and increase in extractables (Chuaqui-Offermanns, 1989a). As a result chemicals or radicals formed may interact with the food affecting its organoleptic characteristics as well as its toxicological safety. Chain-scission changes the actual make-up of the polymers from long chains to short chain segments. This leads to a change in molecular weights as found by Horng and Klemchuk in 1984 in which polypropylene resins had a simultaneous loss of high-molecular weight chains and the formation of lower molecular-weight chains.

Horng and Klemchuk (1984) also observed that polypropylene degraded rapidly with gamma irradiation which results in a loss of physical integrity. In fact, degradation continued following irradiation over storage time. Rojas De Gante and Pascat (1990) also found that radiation of branched polymers such as polypropylene lead to degradation and reactions that continued to develop during storage. They also mentioned internal stress due to gas evolution from irradiation of the film induces breakage of the film and chain scission. The diffusion of gas and the production of short chain final products resulted from the degradative process of irradiation.

Properties of the film such as density, branched versus single chains, film thickness in addition to the dose of irradiation play important roles in the production of chain scission breaks or cross-linking reactions. Typically the denser the product is, the less branched, and the lower the dose the more likely cross-linking reactions are to occur. If the dose rate was lowered and irradiation occurs in the presence of oxygen, a transition stage may follow, where  $O_2$  diffuses into the outer layers of the polymer. In this case, radiation induced oxidative degradation (chain scission) may occur in the outer layers, whereas the inner parts of the polymer may be non-affected or even crosslinked (Wilski, 1987). A major component in the formation of chain scission reactions instead of cross-linking are dose and the presence of oxygen. The lower the dose, the more conceivable a cross-linking reaction will occur. On the other hand, the presence of  $O_2$  leads to oxidative degradation of polymers. Thus, higher concentrations of oxygen during irradiation of polymers leads to greater chainscission products being formed. Oxygen acts to accelerate the degradative reaction by peroxidation of the polymer chain followed by decomposition and rearrangement, leading to a net result of additional chain scission (Hegazy et al. 1981a). Wilski (1987) noted if irradiation takes place in air the degradation was more severe at lower dose rates than at higher dose rates. Consequently, there may be an additive effect in using electron beam irradiation instead of gamma irradiation in the presence of  $O_2$  to reduce chain scission reactions.

Irradiation of polymers can play an important role in color formation of films. While irradiation of certain polymers such as polyurethane and polystyrene leads to improved transparency, discolorations may also develop in other polymers. The development of discoloration in polymers exposed to irradiation is typical of chlorine containing films. Hegazy et al. (1981b) found irradiated poly(vinyl chloride) changed from yellow at low doses and to brown at higher doses. They concluded discoloration was associated with conjugation of radiolytic compounds, the longer the conjugation sequences were the darker the color became. Therefore, discoloration of polymers was formed as chlorine containing chain scission products interacted within the film. Duvis et al. (1991) discovered cross-linking resulted in insolubility in poly(vinyl chloride) while degradation was primarily evidenced by discoloration effects with the use of irradiation.

One often used way to reduce degradation of polymers is the use of stabilizers, antioxidants, and other additives. Basically this group of additives functions by binding with irradiation caused radicals and volatiles within the film, thus reducing further reactions within the polymer. Hence, the radiation induced degradation of mechanical properties of plasticized poly(vinyl chloride) are well retarded by the plasticizers and stabilizers (Hegazy et al. 1981b). Horng and Klemchuk (1984) concluded the incorporation of certain stabilizers can inhibit radiation caused property deterioration, impart color stability, and provide long term protection during storage.

Cross-linking of the carbon chains of polymers by irradiation has a wide range of effects upon packaging films. Cross-linking may lead to changes in tensile strength, hardening, impact strength, bond strength, abrasion resistance, heat resistance, and elongation at break. The net effect of crosslinking reactions has been found by Chuaqui-Offermanns (1989a) to modify the mechanical properties of polymer materials such as to increase the tensile strength, increased hardening, increasing the solvent resistance, and to decrease the impact strength. Irradiation also has been concluded to increase bond strength, abrasion resistance, and heat resistance through cross-linking of polymers (Thayer, 1988).

Based upon some films configurations, properties are more prone to be a result of cross-linking than of chain scission reactions. Because they are less likely to degrade, they are seen as being more radiation resistant. Chuaqui-Offermanns (1989b) reported coextruded films and laminants are more likely to be radiation resistant. Less radiation resistant films at 10 kGy and higher doses are polypropylene, poly(vinyl chloride), cellulose, and poly(vinylidene chloride). Killoran (1983) concluded no single flexible material has all the chemical, physical and protective characteristics necessary to meet the requirements of a food container for irradiation processing. Therefore, multilayer films which combine the best properties while minimizing negative properties should be used. Also, films which provide a good moisture and oxygen barrier, protect contents during shipping, and are easily heat sealable should be used for irradiation processing of prepackaged foods.

The major effects of cross-linking of polymers are upon tensile strength and elongation at break. Tensile strength is defined as the resistance of the film to longitudinal stress without breaking. It indicates how tough the material is and how much it stretches instead of breaking. Elongation is another measure of toughness where elongation, or percent elongation at break, is a measure of the lengthwise stretch a material can withstand. Researchers found irradiation of packaging films commonly caused an increase in tensile strength and elongation at break, and both of these increase at higher doses (Ando and Uryu, 1987; Hegazy et al., 1981b; Varsányi, 1972; and Varsányi et al., 1972). To the contrary, Wilski (1987) reported elongation at break to be the most sensitive mechanical property to irradiation, which decreased with irradiation at doses in excess of 600 kGy.

Varsányi (1972) found radiation doses to decrease tensile strength and elongation at break of irradiated polyethylene films when tested in the machine direction, while the same parameters in transverse testing remained practically unchanged after the same treatments. One of the reasons the author most likely did not find a difference in the transverse tested films was the dose applied was 0.1 to 8 kGy. Normally, film producers use much higher dose rates in the production (cross-linking) of films.

Another factor which may affect the tensile strength and elongation at break of films after irradiation is the presence or absence of oxygen during irradiation. Since the presence of oxygen within a film or outside of the film during irradiation leads to oxidative degradation, tensile strength and other factors could be lowered by the increased incidence of chain scission reactions. In fact a decrease in tensile strength and elongation at break of polypropylene and poly(vinyl chloride) films during irradiation in the presence of  $O_2$  led Hegazy et al. (1981a and 1981b) to conclude oxygen accelerates degradation.

As irradiation dose increases, the proportion of cross-linked polymer increases and the material becomes more elastic (Krylova et al., 1979). This elasticity leads to improved tensile strengths, and elongations of polymers prior to breaking. Another factor of polymers which is influenced by irradiation is stress cracking. Stress cracking leads to the splitting of the package and eventual leakage of the contents. Stress cracking is an important factor in packaging films designed for meat products because stress cracking is increased by fats and free fatty acids (Dempster, 1985; and Tripp, 1959). Dempster (1985) and Tripp (1959) have also shown that irradiation decreases stress cracking of plastics. Thus, irradiation of packaged meat and food products should lead to fewer losses due of fresh meats due to stress cracking.

Abrasion resistance may also be affected by irradiation of plastics. Abrasion resistance is a measure of a films ability to withstand damage caused by friction, such as rubbing, scuffing and scratching. Killoran (1972) reported abrasion resistance of low density polyethylene films increased with radiation dose. The improvements in abrasion resistance, tensile strength, and stress cracking resistance are all due to irradiation induced cross-linking of films. Cross-linking causes the chains of polymers to become more tightly bound and linked to eachother. Matsui et al. (1990) concluded the depression of the absorption of hydrocarbons across irradiated films was a result of the increase in steric hindrance caused by cross-linking reactions. At doses above 50 kGy, Ando and Uryu (1987) found irradiation increased transparency and smoothness of polyurethane by decreasing spherulitic size (rounded crystalline body size). Irradiation also leads to a reduction in crystallinity of other polymers.

Irradiation has also been found to improve the mechanical properties of laminated films. Killoran (1974) and Killoran et al. (1979) have found no delamination among layers of irradiated films and pouches whereas delamination occurred among non-irradiated layers. Seal and bond strengths were also shown not to be significantly effected by irradiation. Two important factors in laminated products are adhesive and cohesive failure. Cohesive failure implies the original bond within the adhesive between two laminants failed rather than the adhesive and film interface, which is adhesive failure. Killoran (1974) concluded the improvement of the mechanical interlocking of layers was caused by the formation of primary chemical bonds extending across the interface due to irradiation. Thus, irradiation decreases both adhesive and cohesive failures in laminated packaging materials.

## **Global Migration**

When flexible packaging materials are exposed to ionizing radiation a number of different types of reactions may occur depending upon the conditions of irradiation and the polymers involved. One of the possible reactions that may occur is the production of low molecular compounds and

other radicals from the parent polymer. Leaching of these compounds from the container into the packaged food product (global migration) can lead to off odors, flavors, as well as further reactions with the foodstuffs. Buchalla et al. (1993) described an increase in the migration of extractives produced as a consequence of irradiation, into food simulants, particularly with fatty media.

In evaluations of the migration of additives, plasticizers, or short chain polymers from the parent polymer, one may determine either the amount leaving the polymer or the amount entering the liquid. Also, food simulating compounds such as water, aqueous acetic acid, aqueous ethanol, heptane and other compounds are typically used in research projects. Tests may involve packaging the liquids within films prior to irradiation or immersing the polymers with the liquid food simulants.

Many factors can influence the migration of additives and monomer residues of polymers. Temperature, compatibility of the migrant with the polymer, molecular size of the migrant, compatibility of the migrant with the media external to the polymer, and the interactions that may occur between the external media and the polymer all have an effect on the migration from films (Duvis et al., 1991). The form of irradiation used may also play a vital role in the global migration phenomena. Because gamma irradiation has a lower dose rate, allowing irradiation to occur over longer periods of time rather than electron beam irradiation, there may be a larger production of short chain polymers which could then migrate. Killoran (1972) reported electron and gamma radiation of plastic films in the presence of food simulating liquids produced the same chemical compounds but in slightly different amounts. The differences were attributed to the stability of the films with regard to their susceptibility to cross-linking and/or degradation at the

relatively low dose rate for gamma radiation and relatively high dose rate for electron radiation. Duvis et al. (1991) also found when cross-linking reactions predominate in the irradiation of plastics, migration was effectively reduced. Thus, higher dose rates from electron beam irradiators are effective in reducing global migration of products when compared with gamma radiation.

As the dose of radiation increases the amounts of migration from polymer films normally enlarge. Killoran (1972) reported irradiated films in comparison to non-irradiated controls had increased amounts of extractives for gamma and electron irradiated films of polyethylene, poly(vinylidene chloride), poly(vinyl chlorine), and polystyrene. Bourgés et al. (1993) went on to report the quantitative results of their study showed that the levels of the compounds lost from packaging materials after irradiation are significantly higher than those of the migrating compounds found in the food simulating liquids. To explain the difference the authors assumed there was a degradative reaction occurring which leads the decomposition of the products after migration from the packaging materials to the food simulating liquid. Killoran (1972) reported the extractive he found migrating to the parent films consisted of low molecular weight polymers of the original parent polymer, yet there was no mention of a difference in the amount of migrants found and lost from the films.

Looking at the migration of food compounds into the film during and after irradiation, Matsui et al. (1990) concluded an irradiation dose of up to 200 kGy was effective in depressing the migration of flavor compounds such as low polar compounds into ethylene vinyl acetate films. One important aspect of this study to note is the authors did not look at the migration of the film into the food. Nevertheless, it still can be concluded there is more of a problem of

polymers migrating into foodstuffs than the migration of foodstuffs into polymers (Bourgés et al., 1993; and Duvis et al., 1991).

While numerous researchers have found radiation induced migration of polymers into food stuffs or simulating liquids there were still no serious toxicological hazard present. Rojas De Gante and Pascat (1990) reported irradiation up to 25 kGy in the presence of oxygen had no significant effects on the global migration of polymers such as low density polyethylene and polypropylene, although amounts of extractive increased with dose. Payne et al. (1965) also found there were no significant differences between nonirradiated film extractives and flexible laminates irradiated with 60 kGy. Nevertheless, irradiation typically increased the production of global migration products into food simulants.

#### **Off-Odors and Taint Transfer**

The formation of gases, volatiles and radicals, as well as low molecular weight compounds may be formed during irradiation of polymers used as packaging materials for meat and other food products. These products can migrate into the food substances and taint the product forming off-odors and off-flavors. It has also been noted that irradiation of polymers produces products such as ketones, aldehydes and carboxylic acids, leading to off-odors in the food product.. Azuma et al. (1983) reported aliphatic hydrocarbons, ketones, aldehydes, carboxylic acids, and alchols were responsible for observed off-odors in irradiated polyethylene films.

While aldehydes, ketones, hydrocarbons, and alchols lead to off-odors, two other products have been implicated for causing the majority of off-odors in

irradiated packaging films. Chlorine containing films when irradiated can form chloride gas and other chlorine substances which are very detrimental to the organoleptic qualities of meat. Chloride ions have been found in water contained in poly(vinyl chloride) bags exposed to 60 kGy irradiation (Tripp, 1959). The organic gases produced by irradiation resulted in objectionable odors which could be acquired by the foods. Azuma et al. (1984a) discovered the amounts of carboxylic acids released from electron beam irradiated polyethylene film could be used to indicate the intensity of off-odor. Therefore, carboxylic acids play a major role in the development of off-odor from plastics, and chloride containing substances also are the most influential factor in the development of off-odors from chlorine containing films.

Azuma et al. (1983 and 1984b) and Keay (1968) found the production of volatiles formed during the exposure of packaging materials to irradiation were responsible for off-odors. While there are numerous factors which lead to the production of volatiles, the presence of oxygen and absorbed dose play the two major roles. In the formation of off-odors in plastics due to irradiation, increasing amounts of volatiles produced leads to greater off-odor intensities. Azuma et al. (1984b) reported a correlation between the amounts of products formed by irradiation and the off-odor intensity. While the amounts of volatiles produced from irradiation are important, the presence of oxygen and dose are the most crucial in off-odor formation because they affect the amounts of volatiles produced.

Although carboxylic acids and chlorides play essential roles in the formation of off-odor in polymers there is no one radical or volatile which can be removed to prevent off-odors. Azuma et al. (1983) suggested from their results that the off-odor of irradiated polyethylene was not composed of only a few compounds but rather many of the identified volatile products were responsible for the off-odors. Therefore, preventing the production of all volatiles, not just one single volatile, is required in reducing the amounts of offodors produced and taint transfer to food products.

Other factors influencing taint transfer to food products and off-odor from irradiated food packaging are temperature during irradiation, package type, history of the film, and oxygen concentration during irradiation. Typically, higher temperatures, greater oxygen concentrations, and older films lead to higher volatile production at the same doses which result in greater off-odor intensities. Matsui et al. (1990) indicated lower temperatures during irradiation resulted in a decrease of off-odors from films. Moreover, Azuma et al. (1984a) reported the intensity of off-odors from polyethylene films increases with oxygen concentration during irradiation.

As polymers age during storage they slowly degrade, forming volatiles and short chain polymers. This process is dramatically enhanced during irradiation, especially if irradiation occurs in the presence of oxygen, due to oxidative degradation. Another factor increasing off-odors and volatile production is the type of polymer being irradiated. Highly branched polymers have been found to be more radiation sensitive than unbranched polymers and slightly branched polymers.. For instance, in the case of polyethylenes the aliphatic side chain appears to be responsible for increased degradative products. Also, the off-odors observed after irradiation are more intense with the highly branched low density polyethylene than with the linear, high density type films (Tripp, 1959).

Many researchers have used the amounts of gases evolved or volatile production from irradiation to rank films for the use in irradiation processing.

Another way of ranking films might be to use off-odor intensity at a set dose. This would be a very similar process to using amount of volatiles produced because off-odors intensities are very highly correlated to amounts of volatiles present. In 1959 Tripp reported odor intensities were low for polystyrene, polyamide, and polyesters while odors intensities were high for polyethylene.

A few researchers have looked into the effects of irradiation transferring taint to food products from packaging films. Lynch et al. (1991) found that irradiation of packaged turkey breasts led to off-odors originating from both the package and turkey breasts. Tripp (1959) reported non-volatile radiolytic products may contribute to off-flavors to the contents of the package. Despite the production of volatiles from irradiation which may taint food products, there are minimal toxicological hazards when packaging and meat are irradiated at low doses from 0 to 10 kGy. Keay (1968) indicated that observed odor and flavor taints from packaging disappeared after cooking fish.

## Infrared Spectroscopy

One of the most sophisticated techniques used in identifying plastic films and differences in films is infrared spectroscopy. Infrared spectroscopy permits an examination of the molecular structure by means of light absorption of the film at various wavelengths, producing a curve which can be compared with charts of known materials (Hanlon, 1992). The results of infrared spectroscopy curves can be confused by additives, coatings, blending of materials used in producing films as well as the irradiation of films. Yet, once a film is identified and a known irradiated film is researched, infrared spectroscopy techniques may be used to confirm irradiation of the films. Also,

amounts of irradiation changes may be used with infrared spectra-graphs to indicate absorbed doses.

Infrared spectroscopy has been used to identify structural modifications in irradiated food packaging material, namely cross-linking and chain scission. Killoran (1972) noted infrared spectroscopic analysis of tested films showed evidence that the strong adhesion among laminate layers was not caused by mechanical interlocking of the layers, but by the formation of crosslinking extending across the interface. Varsányi et al (1972) utilized infrared spectroscopy to observe a significant change in light transmission due to structural modification of the polypropylene foil upon exposure to 8 kGy of radiation and in polyethylene films receiving a dose of 1 kGy (Varsányi, 1972).

The production of cross-linking reactions due to irradiation of plastic films has led some researchers to investigate differences in oxygen transmission caused by irradiation with infrared spectroscopy techniques. Bersch et al. (1959) found the infrared spectra of films irradiated in vacuum showed a decrease in absorption of gases due to increased cross-linking within the outer layer of the film.

Infrared spectroscopy has also been used in identifying the degree of chain scission and oxidative degradation caused by irradiation of films. Infrared spectroscopy can also be used in identifying increased unsaturation of the carbon chains and the production of volatiles and short chain polymers trapped within the irradiated film. Buchalla et al. (1993) and Rojas De Gante and Pascat (1990) found changes in the infrared spectra of low density polyethylene and polypropylene after higher dose (≥100 kGy) to show that different types of oxidation products are formed by irradiation. Charlesby (1960) along with Bersch et al. (1959) have found infrared spectroscopy a useful

technique in identifying the production of oxidative degradation materials of irradiated packaging plastics.

### Electron Spin Resonance Spectroscopy

Information on the application of electron spin resonance (ESR) spectroscopy in polymer research may be found in Rånby and Rabek (1977). The electrons in atoms and molecules form pairs. For each electron in a certain orbital with a spin quantum number, there is another electron in the same orbital with the spin quantum number. Paired electrons do not give an ESR signal, but an unpaired electron has no other electron as a partner in the same orbital and for that reason it produces an ESR signal. The interaction of ionizing radiation with matter (polymers) initiates a reaction in which the two electron chemical bond is cleaved, either symmetrically or unsymmetrically. This forms a "free radical" which is defined as an atom, or a molecule in a state containing on unpaired electron occupying an outer orbital. Thus, ESR signals are used in identifying and quantifying radical production or products in polymers.

Matsui et al. (1990) has used ESR to measure residual radicals of hydrocarbons and low polarity compounds in irradiated ethylene vinyl acetate copolymer films. They found the height of the central resonance peak of the ESR spectrum was useful as an index of residual radical concentration. Thus, ESR signals can be used instead of other gas chromatography techniques in numerating quantities of radicals formed in polymers by irradiation. Rånby and Rabek (1970) indicated ESR could also be used in identifying degradation of

polymers by irradiation forming radicals, enhanced cross-linking of polymers, and oxidation of polymers forming oxygen containing molecules.

While Rånby and Rabek (1970) provided 2,519 references to original papers reviewing the uses of ESR in the study of irradiation of polymers, Horng and Klemchuk (1984) showed no radical signal differences in irradiated polystyrene and non-irradiated controls. They did report both alkyl and peroxy radicals formation which is indicative of an abundance of degradation of polymers by irradiation.

Radicals are effectively trapped in polymers in crystalline regions where their mobility and oxygen accessibility are strongly reduced. These trapped radicals are thought to be responsible for the post-irradiation aging effects that are observed with some polymers (Buchalla et al., 1993). Decay times have been found by Onderdelinden and Strackee (1970) to depend on irradiation conditions. Signal decay in air and vacuum was dramatically different for high molecular weight polyethylene, where an ESR signal was decreased in vacuum and increased in air. From the results the authors concluded that it was not possible to deduce accurately from ESR measurements to what dose a sample was irradiated. However, the report was very promising in gaining useful information on the mechanisms of radical formation and radical diffusion in irradiated polymers.

## Additive Degradation

Plastics can be combined with low molecular weight additives such as antioxidants, plasticizers, heat and light stabilizers, lubricants, slip agents, dyes, degradation inhibitors and fillers. Vinyls are very rigid and brittle, however, with the addition of plasticizers they become soft and pliable. Polypropylene easily degrades and would have a very short life without the addition of antioxidants. Antioxidants play a main role in the removal of alkoxy and peroxy radicals which would otherwise lead to degradation of the polymer. These compounds (antioxidants) would be expected to have an important role to play in the suppression of oxidation of polymers following irradiation (Allen et al., 1987b). Such antioxidants and other additives are also capable of migrating from the plastic into the foodstuff, thereby causing a possible source of contamination, off-odors, and off-flavors.

As irradiation leads to the formation of short chain polymers through chain scission, so in the same way irradiation can lead to the degradation of additives. Allen et al. (1987b) reported two antioxidants in poly(vinyl chloride) and polyethylene polymers had been destroyed by 30 to 40 percent after a dose of 10 kGy. In the case of arylphosphite stabilizers and Irgafos 168, drastic reductions in the levels of the antioxidants occurred during gamma irradiation to such and extent that little remained after a dose of 10 kGy (Allen et al., 1988b).

While researchers have been able to easily quantify the amount of degradation which occurs to additives because of irradiation, there has been a problem in identifying the radiolytic products produced. Techniques used by Allen et al. (1987b) did not reveal the presence of detectable amounts of low molecular weight degradation products derived from antioxidants. It is possible that such products have become covalently bonded to the polymers as a result of a radical coupling process. Azuma et al. (1984a) was able to extract degradation products of hindered phenol antioxidants from irradiated polymers which were coupled to radicals. Thus, it can be easily understood

that while antioxidants easily degrade with irradiation, they join with free radicals preventing further reactions and degradation of the parent polymers.

Antioxidants within polymers act to combine with peroxy radicals and other oxygen containing compounds. The addition of antioxidants and peroxy radicals thus acts to prevent further degradative oxidation of the plastic film and at the same time stabilizes the film. Ahn et al. (1993) reported the effective use of various antioxidants and hot packaging controlled lipid oxidation in turkey patties. Allen et al. (1987a) discovered the formation of phosphate esters reflects the role of Irgafos 168 in destroying the various peroxy radicals generated during gamma irradiation.

Many factors can affect the degradation of antioxidants and other additives. Typically, as irradiation dose increases so does the amount of degradation. Lower temperatures and irradiation within a vacuum can also reduce the amount of antioxidant degradation occurring. The incorporation of antioxidants and additives can also significantly affect the quality of irradiated polymers. For instance, Azuma et al. (1984a) pointed out that without additives the total amount of carboxylic acids from films was three times the amount of carboxylic acids in the same films with additives, and the film without additives had the strongest off-odors. Antioxidants can impart color stability in irradiated poly(vinyl chloride) and polypropylene, in addition to retarding irradiation destruction of mechanical and physical properties.

Irrespective of the nature of the radiation employed, an appreciable proportion of the original antioxidant remains unchanged after a dose of 10 kGy, the maximum irradiation level likely to be permitted with foodstuffs (Allen et al. 1990 and 1987a). The exception is Irgafos 168, an antioxidant used in plastics which is easily degraded by irradiation. Consequently, one can conclude that antioxidants used in plastic production can reduce radical reactions and the amounts of antioxidants unaffected by irradiation can prevent degradation during storage. Another thing to note is that gamma sterilized polypropylene products need stabilizers to protect them during irradiation and storage (Horng and Klemchuk, 1984).

Plasticizers act to make polymers such as poly(vinyl chloride) flexible and pliable. Consequently, certain polymers without plasticizers and other stabilizers would be very brittle and degrade during storage more readily. Horng and Klemchuk (1984) studied four stabilizers and demonstrated their concentrations decreased slowly with irradiation. Hegazy et al. (1981b) stated plasticizers have a marked effect in slowly reducing the radiation chemical changes, while at the same time the plasticizer breaks down readily.

Another advantage of plasticizers is that they impart structural and mechanical stability within the polymers. When polymers containing plasticizers and stabilizers are irradiated there are commonly fewer mechanical and physical changes within the polymer. Hegazy et al (1981b) proposed that stabilizers and plasticizers retarded the degradation of mechanical properties of plastercized poly(vinyl chloride) up to a dose of 2 kGy. At the same time, stabilizers and plasticizers are readily degraded by irradiation. Krylova et al. (1979) established that during irradiation of plasticized poly(vinyl chloride), the polymer undergoes fewer structural changes in systems in which the plasticizer breaks down readily.

During irradiation of plasticized poly(vinyl chloride), both the polymer and the plasticizer undergo breakdown (Krylova et al., 1979). As a result of the composition of plasticizers, phthalic acid esters and monoalkyl esters interact with the double bonds of dehydrochlorinated poly(vinyl chloride) to form

polymer products containing C=O groups (Krylova et al., 1979). Hegazy et al. (1981b) reported stabilizers in poly(vinyl chloride) are degraded by irradiation above 2 kGy, and at the same time the evolution of hydrogen chloride gas was retarded by additives in both the presence and absence of oxygen.

## **Additive Migration**

Many factors can influence the migration of additives from polymers. Temperature, polymer type, dose, compatability of the migrant with the polymer, molecular size of the migrant and the interaction of the food and polymer can all affect the migration of additives from irradiated films. Bourgés et al. (1993) suggested that irradiation and contact with a food simulating liquid induced loss of antioxidants from polypropylene. After migrating from polypropylene into aqueous solutions the migrated compounds decompose into a number of unknown products.

Another phenomenon associated with the migration of additives from irradiated polymers is that a significantly larger proportion of additives degrade than degradation products migrate from polymers. Bourgés et al. (1993) indicated larger amounts of antioxidants are lost than migrate, thus there is a migration of compounds resulting from the antioxidants' degradation. The components of additives migrating to foods may also prove only a minor problem in irradiated prepackaged food because of the result of radical coupling process (Allen et al., 1987b).

Lastly, Allen et al. (1988a and 1988b) have reported the migration of antioxidants from polymers into fatty food simulants and other food simulants to decrease with increasing doses. Therefore, as irradiation doses increase

there becomes more of a problem with degradation of additives and less of a problem with migration of additives. Nevertheless, one must remember the doses likely to be used in prepackaged meats and other foodstuff is a low dose between 0 and 10 kGy.

### Effects of Ionizing Radiation on the Microflora of Fresh Meats

Preservation of meats is a very important issue because fresh meat and poultry provide a near perfect medium for microbial growth. Refrigeration, while the most widely used system in reducing the growth of microorganisms, is limited to a relatively short time of effectiveness. The application of ionizing radiation in the preservation of fresh meats can help to increase hygienic quality, extend shelf life, and reduce the use of chemicals and preservatives.

There are three major categories of dose ranges used in the irradiation processing of foods. The radurization of fresh meats by low dose irradiation is sufficient to delay the onset of microbial spoilage. Radurization is a similar process to food pasteurization, and thus must be used in conjunction with refrigeration. Radurization involves the use of doses less than 5 kGy, the dose range most likely to be allowed in the processing of fresh red meats. Radicidation involves applying higher doses to remove non-spore forming pathogenic organisms (e.g. Salmonella, *Escherichia coli*, and Campylobacter). This irradiation category aims at reducing microbial loads with dose levels in excess of 5 kGy. Radappertization is used for destruction of all spoilage and pathogenic micro-organisms regardless of storage conditions. It involves the use of extremely high doses (above 48 kGy) to effectively destroy spore forming organisms. One problem with the use of radappertization to commercially sterilize fresh meats is the production of off-odors, off-flavors, and discoloration of the meat products (Urbain, 1989).

The use of ionizing radiation has been shown to effectively reduce spoilage bacteria, pathogenic bacteria, molds, yeasts, viruses, and parasites which may be present in fresh meats. Consequently, irradiation is effective in extending the shelf life of fresh meat and poultry. Numerous researchers have developed  $D_{10}$  values for the required dose to effectively reduce individual micro-organisms. Factors such as package type, dose, temperature during irradiation, water activity, oxygen content, and carbon dioxide content have been shown to affect the effectiveness of ionizing radiation in reducing microorganisms. Also there have been reports of radiation induced shift in the microflora of fresh meats from gram negative to gram positive microorganisms. Lastly, certain micro-organisms have been shown to be very radiation sensitive while others have been found to be very radiation resistant.

# Irradiation Increased Shelf Life

Numerous factors affect the shelf life of fresh meats. Sanitation, storage temperature, packaging type, the use of modified atmospheres, and initial microbial contamination of meat can all affect the shelf life of meats. The major reason for a shortened shelf life in meat products is spoilage microorganism contamination. The mass breeding and fattening of livestock, mass production and processing of foods, changing food habits, and increasing environmental pollution may result in increasing food and feed contamination (Kampelmacher, 1983). A polluted environment, and the spread of disease by insects, birds, and rodents play important roles in spreading food contamination and food borne disease. Secondary or cross contamination during the production and processing of meat and poultry can lead to shortened shelf life and contamination with food borne disease microorganisms.

On the otherhand, the use of different processing techniques can have an additive effect on the shelf life of fresh meats. Low refrigerated temperatures have been known for ages to increase the shelf life of meats. Packaging meats in vacuum packaging or other modified atmosphere packaging also leads to an increased shelf life. Hand trimming of carcasses and spray washing with organic acids can also increase the shelf life of fresh meats (Reagan et al., 1996). Consequently, the use of spray washing, modified atmosphere packaging, and low temperature storage can have a synergistic result on the shelf life of fresh meat and poultry.

Irradiation has been known for decades to reduce and eliminate microorganisms in meat and poultry products. Thayer et al. (1993a) reported no surviving microflora were detected in fresh pork samples exposed to radiation doses in excess of .57 kGy even after storage at 2°C up to 35 days after irradiation. The major factor in the effectiveness of irradiation in reducing bacteria loads in meat products is the radiation dose used. As the absorbed dose increase there are greater numbers of electrons and photons released. Thus, higher doses may interact in disrupting the deoxyribonucleic acid (DNA) sequences of more micro-organisms, resulting in greater reductions in the microflora of meat products. Irradiation energy causes single and double strand breaks in the DNA (Mooseley, 1990; and Tarté et al. 1996). In addition, radiation induced radicals cause damage to the DNA molecule such as attacking the DNA bases (Mooseley, 1990). In reviewing the literature Lee et al. (1995) found 1 kGy and 3 kGy were required to extend the shelf life of pork and chicken wrapped in oxygen permeable packaging respectively, and 1.5 kGy was required to extend the shelf life of vacuum packaged beef.

Microbial spoilage of meat can be prevented or greatly reduced by treatment with ionizing radiation. Dempster et al. (1985) demonstrated that low dose irradiation in excess of 1.5 kGy, can improve the shelf life of ground beef by at least seven days at 3°C storage. This extension in shelf life is determined by the initial microbiological quality of the meat. In a further study where fresh beef rounds were irradiated with 1 kGy, Rodgríguez et al. (1993) observed that an average of 17 days more shelflife was possible in contrast to non-irradiated counterparts based on psychrophilic count status. The large increase in shelf life and a very low dose of ionizing radiation found by the authors is most likely due to very low microbial counts. Typically, deep muscles as in rounds remain practically sterile until processing, where ground beef would have a large surface area, and thus higher microbial counts. For instance, Lefebvre et al. (1992) reported that treatment of ground beef with gamma radiation at doses of 1, 2.5, and 5 kGy extended shelf life at 4°C by 4, 10, and 15 days, respectively, while the control samples already exceeded  $10^7$  colony forming units (CFU)/g on the first day of the study. Levels of  $10^7$  CFU or total plate counts are commonly noted by researchers as the point in microbial growth where adverse organoleptic qualities can be detected.

Proctor et al. (1955) also mentioned that at 7.4 and 9.3 kGy, the shelf life increased of various beef and pork fresh meat products. In a study of the effects of irradiation on the shelf life of chicken ,Mercuri et al. (1966) reported

irradiation at 1, 3, and 5 kGy extended the shelf life by 7 days to two weeks. As a result, the higher the absorbed dose, the longer the shelf life.

The quantity of microbial contamination is another major factor effecting the efficiency of radiation in reducing the microfloral of fresh meats. Ehioba et al. (1987) discovered irradiation prolonged the shelf life 2.5 to 3.5 days in uninoculated and 1.0 to 1.5 days in inoculated ground pork. Vacuum packaged pork irradiated at 1 kGy followed the same pattern of spoilage observed in non-irradiated meat but had a considerably longer shelf life.

Another important factor in the effectiveness of irradiation in reducing micro-organisms in food products is temperature at irradiation and storage temperatures. Typically higher temperatures during radiation reflect room temperature and result in higher reductions. Temperatures below freezing result in lower reductions of microbes because the freezing of certain microorganisms preserves them. Opposingly, lower temperatures during storage prior to and after irradiation results in longer shelf lives. A considerable extension in the storage life of green bacon can be achieved with pasteurizing irradiation and low temperatures. For example, Rhodes and Shepherd (1967) reported a dose of 4 kGy delayed spoilage from 4 weeks to more than 20 weeks at 5°C, and from 9 weeks to more than 40 weeks at - 1°C. Naik et al. (1993) found irradiation at 2.5 kGy increased the shelflife from 18 hours to 42 hours of buffalo meat stored at ambient temperatures.

Combining the treatments of ionizing radiation with vacuum packaging, or modified atmosphere packaging, can substantially increase the shelf life of poultry, pork, and beef (Thayer, 1993). Modified atmosphere packaging (MAP) has been reported to increase the shelf life of fresh meats by 50 to 400 percent at refrigerated temperatures (Farber, 1991). While most authors consider MAP and vacuum packaging to be different, Farber (1991) considered both packaging styles to modify the atmosphere. Modified atmospheres usually involves packaging products with a single or combination of gases such as  $CO_2$  and  $N_2$ , and vacuum packaging involves packaging products without a headspace (anaerobic).

Lambert et al. (1992a) confirmed that a substantial extension (9 to 26 days) in shelf life of fresh pork could be achieved using modified atmosphere packaging with nitrogen gas  $(N_2)$  in conjunction with low dose irradiation (1 kGy). In a further study, Lambert et al. (1992b) found at 5°C non-irradiated pork had a shelf life of 9 days if packaged with 20% oxygen, and it was extended to 14 days by packaging in 100% N<sub>2</sub>. Irradiation at 1 kGy extended the shelf life to 21 days in the absence of  $O_2$ , and to 31 days in the presence of  $O_2$ . While the presence of  $O_2$  in the package headspace enhanced the antimicrobial effects of low dose irradiation, it adversely affected the acceptability of the sensory qualities of pork.

#### Irradiation Reduction of Spoilage Microflora

The spoilage of meat by microbial contamination can take place, as well as biochemical degradation may also occur. Meat may be proteolytically degraded by enzymes, fats and heme components may be oxidized resulting in the production of free fatty acids, radical production, and discoloration. While biochemical degradation of fresh meat may play an important role in spoilage, the major factor causing spoilage and degradation of meats is still microbial contamination. The major controlling factor in the quality and quantity of microbial spoilage is storage temperature. Consequently, the longest shelf life of fresh non-frozen meat is achieved by using very low refrigeration temperatures around 0°C.

The shelf life of fresh meats stored at refrigerated temperatures is influenced by the type and numbers of spoilage bacteria. Therefore, to reduce spoilage and increase shelf life of meat at refrigerated temperature, measures should be taken to control and reduce the initial microbial load prior to chilling. To accomplish this several methods may be employed such as spray washing the carcass with solutions of organic acids, hand trimming, and packaging in vacuum or other modified atmospheres. Another measure which may be used to reduce the numbers of spoilage organisms present after packaging is the application of ionizing radiation.

The major spoilage organisms present in refrigerated fresh meats are gram negative, aerobic, Psychrotrophic micro-organisms such as *Pseudomonas sp.* and *Enterobacteriaceae sp.* (Lambert et al., 1992b and 1991d; and Rodríguez et al., 1993). *Pseudomonas sp.* constitute the largest family of bacteria which exist in fresh foods. *Pseudomonas sp.* are typically bacteria of soil and water and are widely distributed in foods. They are by far the most important of the spoilage organisms because many species are psychrotrophic and grow at refrigerated temperatures. *Enterobacteriaceae sp.* are a genera of bacteria within the coliforms and are related to *Citrobacter*, and *Escherichia*, two other coliforms.

While *Pseudomonas* and *Enterobacteriaceae* are the two major forms of micro-organisms present in meats there are numerous other spoilage organisms present in fresh meats at chilled temperatures. Lambert et al. (1991d) reported the major spoilage bacteria of meats are gram negative and include aerobic, psychrotropic strains of *Pseudomonas*, *Moraxella*,

Acinetobacter, Aeromonas and the facultative anaerobe Alteromonas putrefaciens. Lactobacillus and Brochothrix thermosphacta, other gram positive bacteria may also be found in high numbers on fresh meat. Nevertheless, psychrotrophs are important since they are the main organisms responsible for meat deterioration under an aerobic environment at chilled temperatures. They are also the most rigorous indicators of spoilage evidence (Rodríquez et al. 1993). The amount of oxygen available, temperature, and particle size of the meat can also affect the kind of growth on fresh meats. Lambert et al. (1991d) found that unlike spoilage of whole carcasses and primal cuts, ground meats undergo spoilage almost exclusively by gram negative bacteria.

Irradiation can significantly reduce and even eliminate spoilage microorganisms in addition to pathogenic bacteria. Numerous researchers have shown that low doses of irradiation reduced the spoilage of micro-organisms in refrigerated fresh meats and poultry (Ehioba et al., 1988; Lambert et al., 1991d; Lee et al., 1995; Rhodes and Shepherd, 1966; and Thayer, 1993). While the use of low dose irradiation in reducing spoilage bacteria is well documented, the proper absorbed dose for effective use seems very debatable.

Lea et al. (1960) indicated that microbial spoilage could be considerably retarded by doses of ionizing radiation between .25 and 1 kGy for beef and beef fatty tissues. In studying the effects of 2 kGy on beef top rounds, Rodríguez et al. (1993) discovered that psychrotroph counts on non-irradiated samples reached 10<sup>7</sup> CFU/cm<sup>2</sup> between 8 and 11 days of storage, while similar counts were not found until after 28 days of storage on irradiated samples. Consequently, irradiation at 2 kGy was a reliable preservation tool by reducing the naturally occurring spoilage microflora. By studying the use of numerous

doses on refrigerated fresh pork, Thayer et al. (1993a) reported an absorbed dose of 1.91 kGy or higher was effective in eliminating the spoilage microflora. Most likely the differences in the effectiveness of the doses were dependent on the type of fresh meat used and the processing history, which would be indicative of the type and numbers of micro-organisms present prior to irradiation.

Irradiation has also been used to reduce spoilage and competing bacteria on meats used for fermented products. Dickson and Maxcy (1985) noted the use of irradiation lowered the levels of competing bacteria and provided a more uniform product by allowing better control of the fermentation process. Nonetheless, the application of irradiation should be used on fresh product and not on spoiled product. When high numbers (10<sup>6</sup> to 10<sup>7</sup>) of spoilage and pathogenic bacteria were present on pork meat, Grant and Patterson (1991b) found the meat appeared spoiled. Although irradiation at 1.75 kGy significantly reduced the number of bacteria, the meat was still found unacceptable by the taste panel after treatment.

Though low density irradiation is an effective process in reducing gram negative bacteria and gram positive *Staphlococci sp.*, it is very ineffective in reducing lactic acid producing bacteria. This ineffectiveness of reducing lactic acid bacteria by irradiation is also enhanced by vacuum packaging which also reduces gram negative bacteria. Ehioba et al. (1987) and Lambert et al. (1992b) reported numbers of naturally occurring mesophiles, psychrotrophs, and anaerobes or facultative anaerobes were reduced by 1 kGy radiation, whereas lactic acid bacteria were least affected. Irradiation was also found by Grant and Patterson (1991b) to reduce the microflora of modified atmosphere packaged pork, leaving lactic acid bacteria the most dominant organism

present. Thus, psychrotrophic bacteria populations are the most radiation sensitive whereas lactic acid bacteria are some of the least affected spoilage organisms.

Even though irradiation most significantly reduces numbers of psychrotrophic bacteria, some researchers have found further effects on different micro-organisms. Varabioff et al. (1992) in studying the effects of 2.5 kGy on raw chicken packaged in vacuum and in air found the standard plate counts (SPC) were significantly reduced during the 15 days of storage at 4°C. Mattison et al. (1986) reported irradiation of pork loins at 1 kGy reduced numbers of mesophiles, aerobic bacteria and *Staphylococci*, with the greatest effect on mesophiles and psychrotrophic spoilage organisms. Dickson and Maxcy (1985) found irradiation reduced coliforms and *Staphylococci*, while Lambert et al. (1992b) discovered irradiation had the greatest effect in reducing *Enterbacteriaceae sp.* 

Although irradiation is effective in reducing spoilage organisms in fresh meats and poultry, it does not necessarily kill all bacteria. Irradiation is effective in killing a certain percentage of bacteria and damaging another percentage. This is brought about by electrons or photons damaging bacteria, but allowing conditions to exist in which the bacteria may recover over time. Dickson and Maxcy noted that samples irradiated with 2 and 3.5 kGy showed an increase in counts either through growth of surviving bacteria or by recovery of injured cells. Ehioba et al. (1987) also concluded that 1 kGy of irradiation on vacuum packaged ground pork was not always lethal to bacteria because of partial bacterial recovery during subsequent storage at 5°C.

## **Radiation Induced Microflora Shift**

Gram negative micro-organisms are often associated with the spoilage of fresh, refrigerated meats stored in the presence of oxygen. They have been shown to be greatly reduced by lowered oxygen availability. Depending on the meat pH, storage temperature and oxygen permeability of the packaging, gram positive, facultatively anaerobic, lactic acid producing bacteria such as *Lactobacillus sp., Micrococcus sp.,* and *Streptococcus sp.* become predominant over storage time. These gram positive spoilage bacteria result in discoloration and souring of the meat. Other facultatively anaerobic spoilage organisms which may grow in vacuum packaged, refrigerated meats are *Brochothrix thermosphacta* and *Enterobacteriaceae sp.* 

Packaging has been known to make a significant shift from carbon dioxide producing gram negative bacteria in fresh meats to lactic acid producing gram positive bacteria through the use of vacuum and modified atmosphere packaging. Since modified atmosphere packaging typically contains large portions of  $N_2$ ,  $CO_2$ , and slight to no amounts of oxygen it is considered similar to vacuum packaging. Farber (1991) showed that MAP and vacuum packaging reduced the growth of gram negative bacteria while increasing gram positive bacteria over the storage time of refrigerated meats. Lambert et al. (1991d) noted that storage of vacuum packaged, chilled, meat inhibits the growth of aerobic *Pseudomonas* species while aerobic tolerant *Lactobacillus* species or facultative anaerobes become the predominant spoilage micro-organism.

Lactic acid producing bacteria are known to be more radiation resistant, and faster growing than most spoilage organisms at anaerobic, refrigerated temperatures (Grant and Patterson, 1991b). Farber (1991) concluded that MAP

and vacuum packaging favor lactic acid producing bacteria. Lambert et al. (1991d) explained that Lactobacillus sp., Brochothrix thermosphacta and Enterobacteriaceae sp. are not affected by  $CO_2$ , under low  $O_2$  or  $O_2$  free conditions. But lactobacillus bacteria have faster growth than Brochothrix thermosphacta and Enterobacteriaceae sp., thus Lactobacillus sp. predominate in vacuum and modified atmosphere packaging. Their relatively high radiation resistance, coupled with the fact that they are facultative anaerobes, favor their dominance in irradiated MAP meats (Grant and Patterson, 1991b).

Irradiation can make a significant mark on the reduction and elimination of spoilage organisms increasing the shelf life of prepackaged fresh meats, as well as reducing pathogenic bacteria. Nevertheless, the use of low dose irradiation in a pasteurizing form has been shown to induce a shift in the microflora of fresh meats (Lefebvre et al., 1992). Although the microflora of non-irradiated samples shifted from gram negative to gram positive microorganisms, 76 percent were characterized as gram negative at the onset of spoilage in vacuum packaged ground pork (Ehioba et al., 1988). However, irradiated ground pork microflora in this study was mainly gram positive (66%) shortly after irradiation and increased to 97 percent when spoilage of the controls occurred.

Thayer et al. (1995) noticed the change of microflora was predominately from gram negative rods in non irradiated mechanically deboned meat to gram positive streptococci in 3 kGy irradiated samples. Grant and Patterson (1991b) also found that lactic acid bacteria were predominantly isolated in irradiated samples stored in MAP for 17 days. In general, low dose irradiation has its largest reduction in psychrotrophic and anaerobic or facultative anaerobic bacteria whereas lactic acid producing bacteria are least affected by irradiation when meat is packaged in vacuum or MAP (Ehioba et al., 1987; Lambert et al., 1992b; Lebepe, 1990).

Niemand et al. (1983) found irradiated ground pork contained 90 percent or more gram positive organisms at the end of the 12 day refrigerated storage period. Lactic acid producing bacteria were least affected while *Psuedomonas* and *Enterobacteriaceae* species were greatly affected by irradiation. In contrast, Welch and Maxcy (1975) reported the residual micro-organisms surviving a 10 kGy dose in samples were predominantly gram negative coccobacilli. At such a high dose most of the gram positive spoilage organisms were leaving only gram negative radiation resistant coccobacilli.

# D<sub>10</sub> Values for Food Bacteria with Irradiation

Many researchers have reported the  $D_{10}$  values for food pathogens. A  $D_{10}$  value represents the required absorbed dose or irradiation to get a 10 fold decrease in the viable counts. In layman's terminology a  $D_{10}$  dose is the dose as well as a temperature to eliminate 90 percent of the microbial population. Palumbo et al. (1986), Thayer (1993) and Thayer et al. (1993b) reported the  $D_{10}$  value of *Aeromonas hydrophila* at 2°C in beef to be 0.14 - 0.19 kGy. Lefebvre et al. (1992) discovered the  $D_{10}$  value of *Achromobacter sp.* in ground beef to be 0.129 kGy, 1.485 kGy for *Bacillus cereus*, and .291 kGy for *Brochotrix thermosphacta*. Clavero et al. (1993b) found the  $D_{10}$  value for *Campylobacter jejuni* to be 0.14 - 0.235 kGy on beef and turkey at 0 - 5°C. Thayer (1993) and Thayer et al. (1993b) found the  $D_{10}$  value of *Clostridium botulinum* to be 3.56 kGy at -30°C

on chicken, while Clavero et al. (1994) and Thayer (1993) reported the  $D_{10}$  value of *Escherichia coli* 0157:H7 to be 0.241 - 0.307 kGy in ground beef at 5°C. Lefebvre et al. (1992) found the  $D_{10}$  value of *Listeria monocytogenes* to be between 0.035 and 1.827 kGy and 0.053 - 0.153 kGy for *Pseudomonas sp.* in ground beef. Clavero et al. (1994), Lefebvre et al. (1992), Tarkowski (1984b) and Thayer et al. (1993b) and (1990) have reported the  $D_{10}$  value of *Salmonella sp.* in ground beef and mechanically separated chicken at 2°C to be 0.30 - 1.20 kGy and 0.36 - 1.827 for *Staphylococcus aureus* at 0°C. *Yersinia enterocolitica* had  $D_{10}$  values reported by Lefebvre et al. (1992), Radomyski et al. (1993), and Tarkowski et al. (1984b) to be 0.04 - 0.21 kGy in ground beef.

The  $D_{10}$  value is very much affected by temperature. Lower temperatures usually result in greater  $D_{10}$  values to obtain the same deathloss at a higher temperature. Clavero et al. (1994) reported  $D_{10}$  values for pathogens in frozen ground beef were generally higher than those calculated for refrigerated beef. In contrast, lower  $D_{10}$  values for pathogens exist when irradiation occurs in the presence of oxygen (Grant and Patterson, 1991a). The  $D_{10}$  value for *Clostridium botulinum* is the highest, typically at a frozen temperature, because this value is used in commercial sterilization of food products. Typically food pathogens are more radiation resistant and spoilage organisms are more radiation sensitive. For pathogenic micro-organisms *Yersinia sp.* and *Campylobacter sp.* are the most radiation sensitive while *Listeria monocytogenes* and *Staphylococcus sp.* are the most radiation resistant.

### **Radiation Resistance of Microflora**

The physical composition and growth factors of micro-organisms may allow them to be more radiation resistant than other microflora. For instance, spore formers are typically a more radiation resistant type of bacteria. Factors such as the temperature during irradiation, water activity, and reexposure to radiation may affect the radiation resistance of bacteria.

Typically, as irradiation temperature increases, resistance of microorganisms decreases, of course there are some exceptions. Anellis et al. (1977) recorded the fact that *Streptococcus faecium* are more resistant than *Clostridium botulinum* in beef and are considerably less resistant to irradiation below - 20°C and are much more resistant above this temperature.

Moraxella - Acinetobacter sp., is a gram negative coccobacillus, which is known to occur on raw beef and poultry. Certain strains are radiation resistant, while others are able to survive heat treatments. Elias (1985) noted Moraxella - Acinetobacter cells are more resistant to irradiation than Clostridium botulinum spores. Earlier thermal processing of the food redresses the balance of both Moraxella - Acinetobacter and Clostridium botulinum spores as well as contributes to the elimination of these microorganisms. In reviewing the effects of temperature on the radiation resistance of micro-organisms Anellis et al. (1977) concluded that vegetative microorganisms may experience radiation resistance equal to, if not surpassing, the resistance of some bacterial spores at low temperatures. Thus, temperature can have a wide variety of effects on the radiation resistance of many bacteria.

Radiation sensitivity of bacteria is known to be strongly influenced by the amount of water in the system (Huhtanen et al., 1989). The irradiation of water as well as meat products forms radicals which may interact with the

bacteria to cause damage. Consequently, the higher the amounts of water present in the food during irradiation, the more damage may occur to microorganisms.

While almost all of the radiation resistant strains of micro-organisms play a very minor possible role in foodborne illness there are numerous strains which are radiation resistant. For instance, doses of 47 kGy or greater are required to achieve a 12D reduction in the number of Clostridium botulinum spores in meat products (Lambert et al. 1991d). Moraxella - Acinetobacter sp. are also very radiation resistant. Enterobacteriaceae sp., Brochothrix thermosphacta, and lactic acid bacteria are all quite resistant to low dose irradiation (Lambert et al., 1992b). Lefebvre et al. (1992) found Salmonella typhimurium and spoilage organisms in ground beef, to be more radiation resistant than most food pathogens while Bacillus cereus, Micrococcus sp.. and Staphylococcus sp. were significantly more radiation resistant than other psychrotrophic bacteria. Welch and Maxcy (1975) also wrote the radiation resistance  $D_{10}$  values ranged from .273 to 2.039 kGy for the normal vegetative bacteria of ground beef.

#### Irradiation Reduction of Food Pathogens

#### <u>Aeromonas hydrophila</u>

Aeromonas hydrophila are psychrotrophic sugar fermenting gram negative rods which may grow at temperatures as low as 0°C. The fairly common occurrence of Aeromonas on red meats, poultry, and fresh produce and its ability to grow and produce cytotoxin, hemolysin, and enterotoxin under refrigerated temperatures give rise to further concerns regarding the

public health risks associated with the consumption of these foods (Radomyski, et al. 1994). Aeromonas sp. have been found in temperature abused samples (Lebepe et al., 1990), while Palumbo et al. (1986) found 1.5 kGy was sufficient to eliminate this organism from food. In contrast, Lebepe et al. (1990) discovered that Aeromonas hydrophila survived an irradiation dose of 3 kGy in low numbers. Nevertheless, Aeromonas hydrophila cells are reduced by irradiation relative to unirradiated samples (Radomyski et al., 1993).

### <u>Salmonella sp.</u>

Salmonella are gram negative enteric bacteria associated with animal fecal matter. While Salmonella are non-sporing rods there are enterotoxin and cytotoxin producing strains; Salmonella can cause a foodborne infection known as salmonellosis, a mild form of food poisoning. Salmonella has also been found as the cause for typhoid fever. As far as poultry and meat are concerned, Salmonella is presently the most important causal agent of food infections in most countries (Kampelmacher, 1983). Of gram negative pathogens Salmonella sp. may be the most resistant to radiation (Monk et al., 1995). Thus, if irradiation can eliminate Salmonella all other food pathogens should be eliminated. Numerous researchers (Monk et al, 1995; Radomyski et al., 1993 and 1994; and Satin, 1993b) have shown that ionizing radiation is an effective means of reducing Salmonella from fresh red meat and poultry. Thayer et al. (1992) showed a population of 1000 CFU/cm<sup>2</sup> would be decreased to 500 cells with 1.4 kGy, which is well below the estimated infectious dose. Thus, a very large amount of raw irradiated chicken would have to be consumed for a healthy adult to receive an infectious dose.

Many factors influence the effectiveness of radiation in reducing or elimination Salmonella from fresh meats and poultry. The Salmonella species involved, dose used, temperature during irradiation, oxygen level, fat level, and storage time can all affect the usefulness of radiation treatments. Under normal circumstances the radiation resistance of bacteria decrease with increasing temperatures (Hanis et al., 1989; Mulder et al., 1977; and Thayer et al., 1990), which allows  $D_{10}$  values to rise dramatically at frozen temperatures rather than at refrigerated temperatures (Clavero et al., 1994). Thayer and Boyd (1991b) also found that irradiation was significantly more lethal to the bacterial cells at temperatures above freezing. Thayer and Boyd (1991a) further noted that gamma irradiation was significantly more lethal for Salmonella typhimrium in the presence of air and at higher temperatures.

Fat levels appear to have less of an effect on radiation reduction of Salmonella than storage time. Clavero et al. (1994) found at any given temperature, during irradiation, the level of fat did not significantly influence  $D_{10}$  values for Salmonella. Thayer et al. (1995) reported that an initial inoculum of Salmonella enteritidis of 3.86 log<sub>10</sub> CFU/g of mechanically deboned chicken meat (MDCM) decreased during storage at 5°C and was further reduced by irradiation. Therefore, populations of Salmonella present prior to irradiation would be lowered by irradiation and possibly during refrigerated storage. In studying the effects of irradiation on Salmonella in mechanically deboned chicken meat in vacuum and aerobic packaging, Thayer and Boyd (1991a) discovered a dose of 3.0 kGy at -20°C in air reduced the numbers of Salmonella by 4.78 logs, and if irradiated in vacuum, by 4.29 logs.

European and South American countries have a delicacy known as 'filet américain' which consists of raw ground beef with a mayonnaise type sauce,

salad oil, egg yolk, vinegar, salt and other spices. Because this product is eaten raw there is a possibility of infection from food pathogens, which could be eliminated with irradiation producing a safer product for the consumer. In studying the effects of irradiation on 'filet américain', Tarkowski et al. (1984b) found that  $D_{10}$  values for four strains of *Salmonella* were higher than for raw ground beef. One kGy was effective in eliminating *Salmonella* from 119 of 120 samples of 'filet américain' when 23 percent of the samples had isolated *Salmonella* prior to irradiation (Tarkowski et al., 1984a).

The use of low dose irradiation at or under 2 kGy has proven effective in reducing Salmonella from poultry, mechanically deboned chicken meat, and fresh red meats (Thayer et al., 1995, 1992, and 1990; and Thayer and Boyd 1991b). Meanwhile, irradiation has proven effective in eliminating Salmonella typhimurium from poultry at 10 kGy (Hanis et al., 1989), Salmonella sp. from pork loins at 3.0 kGy (Lebepe et al. 1990), Salmonella sp. from broiler carcasses at 2.5 kGy (Mulder et al., 1977), Salmonella enteritidis from MDCM at 3.0 kGy (Thayer et al., 1995), and Salmonella typhimurium from chicken wings at 2.7 kGy (Thayer et al., 1992). In studying different strains of Salmonella , Thayer et al. (1990) reported that Salmonella enteritidis was significantly more resistant to ionizing radiation than the other Salmonella strains.

## Escherichia coli (E. coli 0157:H7)

*Escherichia coli* is a dominant gram negative bacteria found in the intestine of warm blooded animals. The existence of *Escherichia coli* in the environment originates from feces of livestock, animals, and humans. Consequently the occurrence of *E. coli* in water and food products is an

indication of fecal contamination. *E. coli* 0157:H7 is capable of causing hemorrhagic colitis, hemolytic uremic syndrome (HUS) and thrombocytopenic thrombotic purpura (TTP). These infections have mostly been linked in children with the consumption of undercooked ground beef. The *E. coli* gastroenteritis syndrome is caused by the ingestion of viable cells that colonize the small intestine and produce enterotoxins.

Irradiation processing of fresh meats and poultry has proven effective in reducing and eliminating *Escherichia coli* 0157:H7. *E. coli* 0157:H7 was found to be very sensitive to irradiation at doses within the range of 1.5 to 3.0 kGy, indicating that it could be very effectively controlled in poultry meat by irradiation (Thayer and Boyd, 1993). Monk et al. (1995) and Radomyski et al. (1994) also reported that low dose irradiation was an effective method of controlling *Escherichia coli* 0157:H7 in fresh meats and poultry. Dickson and Maxcy (1985) noted that 5 kGy reduced coliforms in the batter for the production of fermented sausage below detectable limits.

Neither the fat levels of meat products nor packaging types have little consequence on the effectiveness of irradiation controlling *E. coli*. Clavero et al. (1994) showed that fat levels did not have any significant effects on the  $D_{10}$  values of *Escherichia coli* 0157:H7. Thayer and Boyd (1993) found no evidence for an effect of air versus vacuum packaging of inoculated meat samples after irradiation.

The major factor affecting the effectiveness of radiation in eliminating or reducing *E. coli* 0157:H7 from meat products is temperature. *Escherichia coli* 0157:H7 was unusually sensitive to temperature during irradiation, with irradiation being significantly more lethal above 0°C than frozen temperatures (Thayer and Boyd, 1993). Clavero et al. (1994) also reported that D values for *E*. *coli* 0157:H7 were higher in frozen than in refrigerated samples. The failure to detect either viable *E. coli* 0157:H7 or toxin in meat challenged with 10<sup>4.8</sup> CFU/g and irradiated to 1.5 kGy at 0°C following 20 hours of temperature abuse at 35°C indicates that very substantial protection can be offered to the consumer by irradiation (Thayer and Boyd, 1993).

## <u>Campylobacter jejuni</u>

Campylobacter jejuni is a gram negative, microaerophilic to anaerobic rod which often is found in the gastrointestinal tract of livestock and poultry. Because these bacteria are microaerophilic they grow in vacuum packaged and modified atmospheres of packaged poultry and red meats. In fact, certain strains of Campylobacter jejuni require 10 percent of the atmosphere to consist of  $CO_2$  for good growth, leading to a possible serious problem with modified atmosphere packages. Campylobacter jejuni is a frequent contaminant of poultry and red meats and is recognized as a leading cause of acute bacterial gastroenteritis (Monk et al., 1995).

Campylobacter jejuni can produce a heat labile enterotoxin which causes diarrhea in humans. This enteritis syndrome mimics acute appendicitis. While diarrhea and a fever are normal symptoms, bloody stools may occur. The incubation period can be very long, 2 to 10 days or more with diarrhea lasting 2 to 7 days. Thus, tracing food poisoning caused by *Campylobacter jejuni* is very difficult due to the long incubation period. Nevertheless, because of the existence of the enteritis syndrome, *C. jejuni* is seen as a serious food pathogen. Campylobacter jejuni has been noted (Monk et al., 1995) to be very sensitive to low dose irradiation in meat and poultry. Of *E. coli* 0157:H7, Salmonella sp., and *C. jejuni* in ground beef, Campylobacter jejuni was the most sensitive bacterium to irradiation (Clavero et al., 1994). Tarkowski et al. (1984b) reported that Campylobacter jejuni sensitivities were greater in the filet américain at approximately 0.10 kGy than in ground beef without sauce at about 0.15 kGy. In another study, (Tarkowski et al., 1984a), concluded that 1 kGy was effective in producing product free of *C. jejuni* because the  $D_{10}$  values of 0.08 to 0.16 kGy for this bacterium indicates it is among the most irradiation sensitive micro-organisms.

Factors such as package type, temperature, and growth phase can influence the sensitivity of *Campylobacter jejuni* to radiation. Because *C. jejuni* is microaerophilic, vacuum packaging and MAP lead to higher survival rates from irradiation than aerobic packaging. Lower temperatures also lead to higher  $D_{10}$  values. Clavero et al. (1994) found significantly higher  $D_{10}$  values were calculated for *C. jejuni* in frozen rather than in refrigerated high fat beef. Radiation resistance can also be influenced by the physiological age of *C. jejuni*, with early log cells being more susceptible to irradiation than stationary cells (Lambert and Maxcy, 1984).

Because of *C. jejuni's* radiation sensitivity, researchers have discovered that low dose irradiation is effective in eliminating *Campylobacter jejuni* from vacuum packaged, refrigerated red meats and poultry (Radomyski et al., 1993 and 1994). Lebepe et al. (1990) found irradiation of pork loins at 3.0 kGy and storage at 2 to 4°C for 98 days in vacuum packaging tested negative for *Campylobacter sp.* 

# Yersinia enterocolitica

Yersinia enterocolitica is a gram negative, facultatively anaerobic bacterium which can grow at 0 to 4°C. It is found in the gastrointestinal tract of livestock and in soil. Thus, Y. enterocolitica is a health hazard that can grow at refrigerated temperatures in vacuum packaged fresh meats. Yersinia enterocolitica causes gastroenteritis syndrome which typically develops over several days after the ingestion of the infected foods and is characterized by abdominal pain and diarrhea (Jay, 1992).

While Yersinia enterocolitica is known to be somewhat radiation resistant in comparison to other food pathogens, numerous researchers have reported that low dose irradiation is effective in greatly reducing the bacterium from ground beef, pork, and other meats (Monk et al., 1995; Radomyski et al., 1993 and 1994). El-Zawahry and Rowley (1979) discovered a dose of 2 kGy at 5 to  $25^{\circ}$ C reduced Yersinia enterocolitica in meat by 10 log cycles. Furthermore, some cells surviving low dose irradiation were injured, as evidenced by their inability to form colonies in the presence of 3.0 percent sodium chloride or at an incubation temperature of  $5^{\circ}$ C.

Yersinia enterocolitica has been found to survive a 1 kGy dose in vacuum packaged ground pork samples stored at 5°C (Ehioba et al., 1988). Lebepe et al. (1990) concluded that protection against Y. enterocolitica survival and potential growth in fresh vacuum packaged pork may require higher doses than 3 kGy. Consequently, one can easily see that Yersinia enterocolitica are slightly more radiation resistant in comparison to other pathogens, sometimes requiring slightly higher doses to eliminate this organism.

Tarkowski et al. (1984b) reported  $D_{10}$  values for filet américain ranged from 0.080 to 0.043 kGy and for ground beef without a sauce 0.21 to 0.10 kGy for *Yersinia enterocolitica*. The authors concluded that a dose of 1 kGy was sufficient to eliminate *Yersinia enterocolitica* without affecting the organoleptic values of the filet américain if the meat was irradiated before the addition of the mayonnaise. In a second experiment Tarkowski et al. (1984a) found that *Y. enterocolitica* was present in fifty percent of the raw meat samples but the organism was not isolated from samples irradiated with 1.5 kGy. Thus low dose irradiation may be effective in eliminating *Yersinia enterocolitica* in low numbers from fresh meats.

#### <u>Bacillus cereus</u>

*Bacillus cereus* is a spore forming, gram positive rod which is aerobic and found in water and in the soil and can cause foodborne gastroenteritis. *Bacillus cereus* does not grow well below 4°C, but does produce a number of toxins. Symptoms of *Bacillus cereus* food poisoning occur within 16 hours after infection. Symptoms consist of abdominal pains and watery stools. Toxin production is also associated with spores. Low dose irradiation of fresh meats can reduce *Bacillus cereus* by 3 to 4 log<sub>10</sub> cycles, while a higher dose of 4 kGy may be required for D<sub>10</sub> values of spores (Monk et al., 1995). Because *Bacillus cereus* is an aerobic food pathogen, the use of vacuum packaging and irradiation should prove effective in minimizing incidents of food poisoning from this particular pathogen.

### Listeria monocytogenes

Listeria monocytogenes is a gram positive, non sporing rod which can be found in the gastrointestinal tract of livestock and poultry as well as in the soil. It grows at 1°C in the presence of oxygen, and is a known contaminant of milk, red meats and poultry. *Listeria monocytogenes* can lead to an infection known as listeriosis in humans. Symptoms can last months and include mild influenza-like symptoms. In pregnant women premature birth and stillbirth may occur. Listeriosis has a high fatality rates for the young and immunocompromised individuals.

Listeria monocytogenes is known to be fairly sensitive to radiation with  $D_{10}$  values in poultry of 0.42 to 0.55 kGy (Patterson et al., 1993). While irradiation is not very effective in eliminating Listeria monocytogenes, low dose irradiation has been proven to be very effective in greatly reducing L. monocytogenes (Monk et al., 1995; Patterson et al., 1993; and Radomyski et al., 1993 and 1994). Huhtanen et al. (1989) reported that a dose of 2 kGy was sufficient to destroy 4 log<sub>10</sub> cycles of Listeria monocytogenes in MDCM.

The phase of *Listeria monocytogenes* growth has been found to be very important factor on the destruction of bacteria by irradiation. The use of irradiation predominantly during the log phase of *L. monocytogenes* has been shown to be most affective in reducing the contamination in poultry (Huhtanen et al., 1989; and Patterson et al., 1993). Consequently, the irradiation of a refrigerated product at low temperatures around 0°C would be more effective in reducing *L. monocytogenes* from fresh meat and poultry products than at higher temperatures.

The USDA (1992) allowed irradiation of chicken and fresh poultry products packaged under air and not in vacuum, so not only to control

Clostridium but Listeria growth could also be prevented. This is most likely due to the increased lethality of radiation in the presence of oxygen. Varabioff et al. (1992) found that Listeria monocytogenes was only recovered from the vacuum packaged irradiated chickens after 7 days of storage at 4°C. This observation indicated that not all the Listeria monocytogenes were destroyed by irradiation at 2.5 kGy and the surviving cells were able to grow in the absence of air. At the same time in unirradiated chickens, L. monocytogenes proliferated similarly in both air and vacuum packaged chickens. But, following 15 days of storage the number of Listeria monocytogenes were significantly higher in aerobically packaged unirradiated chickens than in vacuum packaged unirradiated chickens.

Two strains of *Listeria monocytogenes* being studied by Tarté et al. (1996) were found to possess very effective mechanisms for the repair of their sublethal damage by irradiation. *Listeria innocua* was also discovered to possess a superior mechanism for the immediate and complete repair of damaged DNA (Tarté et al., 1996). Thus, it was concluded that irradiation doses that would eliminate *L. monocytogenes* would also be adequate for the destruction of *L. ivanovii*, but not necessarily *L. innocua*. While low dose irradiation may or may not eliminate *Listeria monocytogenes* from red meats and poultry, Huhtanen et al. (1989) reported 10 kGy would ensure complete elimination of this contaminant from meat.

#### Staphylococcus aureus

Staphylococcus aureus is a gram positive, toxin producing, mesophilic pathogenic bacterium. It can grow at temperatures as low as 7°C, while

enterotoxin production may occur between 10°C and 46°C. Enterotoxigenic bacteria may arise from gastrointestinal contamination from animal origins, while human contamination of foods with *Staphylococcus aureus* is most typical. The enterotoxin produced by *Staphylococcus aureus* which is heat stable can cause a form of food intoxication resulting in gastroenteritis if enough toxin is ingested. Symptoms occur 1 to 6 hours after ingestion of the contaminated meat and food products leading to nausea, vomiting, and diarrhea.

Radiation is known to significantly reduce or eliminate *Staphylococcus aureus* from meat and food products depending on dose and irradiation conditions. Gamma radiation doses of 0.26 and 0.36 kGy administered to MDCM vacuum packaged and stored at 0°C, destroyed 90 percent of the logphase and stationary-phase of CFU of *Staphylococcus aureus* (Thayer and Boyd, 1992). The authors went on to estimate that doses of 3.0 and 1.5 kGy should destroy 6.32 and 3.20 logs of CFU/g respectively of *Staphylococcus aureus* in MDCM. The temperature at which the product was irradiated significantly affected the destruction of *S. aureus*. The higher the temperature above 0°C during irradiation the higher the lethality of the dose.

Irradiation of fresh refrigerated meats and poultry has also been reported to eliminate *Staphylococcus aureus* (Monk et al., 1995). Lebepe et al. (1990) noted that 3 kGy eliminated *S. aureus* from vacuum packaged pork loins stored at 4°C for more than 13 weeks. Thayer and Boyd (1992) found that 1.5 kGy was effective in eliminating *Staphylococcus aureus* from MDCM prior to and after storage. Enterotoxin was never discovered in the authors irradiated samples. Monk et al. (1995) also noted that *S. aureus* enterotoxins are radiation stable.

### <u>Clostridia</u>

Clostridium perfringens is a gram positive, spore forming anaerobic rod which is commonly found in soil and water and produces an enterotoxin. Food poisoning is caused by ingestion of the enterotoxin in sufficient amounts. Typically, foods leading to this type of food poisoning result from heating the food to a point sufficient to kill off the majority of the micro-organisms present and not *Clostridium perfringens*. Thus, *C. perfringens* is allowed to grow without competitors present. Food poisoning typically results when foods are first cooked and then stored in refrigeration and reheated the next day or two allowing time for a large amount of enterotoxin to develop. Symptoms appear between 6 and 24 hours of ingesting the toxin and consist of abdominal pain, diarrhea, and nausea. Duration of symptoms are one day or less. The fatality rates in healthy adults are quite low.

Because Clostridium perfringens is a spore forming rod it is more radiation resistant than Salmonella, Listeria, E. coli, and Yersinia (Monk et al., 1995). Thus, of common food pathogens C. perfringens is one of the most irradiation tolerant. Lebepe et al. (1990) reported that 3 kGy eliminated Clostridium perfringens from vacuum packaged pork loins stored at 4°C for 98 days, while it was found in temperature abused samples. Grant and Patterson (1991a) noted that the growth of C. perfringens is not inhibited by MAP containing  $CO_2$  or  $N_2$ , while irradiated MAP pork is safer than unirradiated MAP pork especially under temperature abuse conditions.

*Clostridium botulinum* is a gram positive, anaerobic, spore forming rod which produces a neurotoxin. This micro-organism is also found in the soil

and water and the neurotoxin causes a food illness known as botulism. In a healthy adult the ingestion of a relatively small amount of neurotoxin results in symptoms within 12 to 72 hours. Symptoms include nausea, vomiting, fatigue, dizziness, paralysis, respiratory failure, blindness, and death. Symptoms have a duration of 1 to 10 days with a 30 to 60 percent mortality rate. Because of the severity of this neurotoxin the canning industry has adopted a thermal processing procedure sufficient to receive a 12 D reduction of this micro-organism.

There are fears by some officials in government agencies that the use of irradiation in vacuum packaged meat products may substantially reduce or eliminated spoilage micro-organisms allowing *Clostridium botulinum* spores to germinate and produce toxin while the product remained acceptable in sensory characteristics. There are also fears that MAP as well as vacuum packaging may enhance toxin production. Carbon dioxide may exert a bactericidal effect whereas it can act as a stimulatory effect on micro-organism spores of meat systems. Gram negative bacteria are more sensitive to  $CO_2$ , while gram positive bacteria such as *C. botulinum*, are more resistant. Nitrogen gas typically has no significant effect on micro-organisms.

Since  $CO_2$  gas has been linked to enhanced spore germination the USDA (1992) decided to allow only aerobic packaging in the irradiation processing of fresh and frozen chicken. To achieve a 12 D reduction in the number of *Clostridium botulinum* spores, 47 kGy was required (Lambert et al., 1991d). Of course temperature can change this, consequently higher temperatures require lower doses. It is also possible by using cryogenic temperatures and vacuum packaging to produce high quality sterile meats by irradiation to the 12 D dose for *Clostridium botulinum* spores (Thayer, 1993).

The major factor affecting the growth and sporulation of *Clostridium* botulinum is the temperature at which the meat substance is stored. Lambert et al. (1991d) recorded that the growth of *C. botulinum* in MAP irradiated fresh meats could be prevented by storage at proper refrigeration temperatures. Irradiation of buffalo meat at 2.5 kGy and held at ambient temperatures (~30°C) developed *Clostridium sp.* after 12 hours of storage. Using a higher dose of 20 kGy Coleby et al. (1961a) found cans of beef stored at ambient temperatures also developed *Clostridia* contamination. Nevertheless, *Clostridia* counts were found by Mattison et al. (1986) to be significantly lower for irradiated pork (1 kGy) than for non-irradiated pork with the differences growing greater over 21 days of storage. Anellis et al. (1977) also reported that vegetative micro-organisms may experience a higher rate of protection than spore formers at decreasing radiation temperatures under comparable conditions.

Clostridium botulinum spores are very heat and irradiation resistant (Monk et al., 1995). Nonetheless, numerous factors such as temperature, dose, and atmosphere or head space composition can affect the sporulation or growth of *Clostridium botulinum* spores. Typically *C. botulinum* spores are more radiation resistant at lower temperatures (below 0°C). Anellis et al. (1977) reported resistance of the spores decreased linearly with increasing temperature from -140 to 5°C.

Typically levels of oxygen in MAP irradiated meat products inoculated with *Clostridium botulinum* spores are thought to prevent sporulation and production of toxin. At the same time amounts of  $CO_2$  in MAP irradiated fresh meat products inoculated with *C. botulinum* spores are thought to increase toxin production through increased growth of spores. Lambert et al. (1991d) reported that high levels of  $CO_2$  can stimulate spore germination. In a challenge study with *C. botulinum* in pork, Lambert et al. (1991a) found that if the meat is packaged with 20 percent  $O_2$ , the level of  $O_2$  rapidly decreased and headspace  $CO_2$  increased from 20 to 40 percent due to respiratory activity of the meat and aerobic micro-organisms present. Carbon dioxide is very soluble in meat, thus increases in headspace  $CO_2$  are due to meat tissue and microbial respiration (Lambert et al., 1991b).

With respect to the levels of  $O_2$  present in the headspace of MAP fresh meats the fact that the presence of  $O_2$  is more detrimental to micro-organisms because of the greater amounts of radiolytic and bactericidal substances produced by irradiation should also be taken into account (Dickson and Maxcy, 1984). One of the main radiolytic components of irradiation in the presence of  $O_2$  is ozone. While  $CO_2$  has been found to stimulate spore germination and toxin production of *C. botulinum*, ozone has not (Lambert et al., 1991a).

The interactions of packaging atmospheres, toxin production and irradiation have been studied with fresh meat inoculated with *Clostridium botulinum* spores. Lambert et al. (1991c) found toxin production occurred faster in non-irradiated samples packaged initially with  $O_2$  than irradiated samples. While Grant and Patterson (1991a) noted that a dose of 3 kGy may allow *Clostridium botulinum* spores to produce toxin in uninoculated fresh meats, Lambert et al. (1991a) found 1 kGy was sufficient in delaying toxin detection by 22 days in inoculated pork.

Moreover, toxin was not detected in any sample stored at 5°C even after 44 days of storage (Lambert et al., 1991a). Thus proper refrigeration temperatures can prevent toxin production in irradiated fresh meats, even if samples are inoculated with *C. botulinum* spores. Thayer et al. (1995) reported none of their samples stored at 5°C developed botulinum toxin, however, if samples were abused at 28°C they became toxic within 18 hours and had obvious signs of spoilage, such as swelling of the cans. It was assumed that the swelling was due to amino acid decarboxylation. Because of the dose being less than 3 kGy, radiation should have relatively little effect on the highly radiation resistant *C. botulinum* spores. Spores were expected to survive irradiation and storage, but not to multiply or produce toxin at temperature equal to or lower than 5°C.

Modified atmosphere packaging can also affect the production of C. botulinum toxin. Lambert et al. (1991b) reported that toxin production occurred faster in inoculated samples initially packaged with 15 to 30 percent of CO<sub>2</sub> while higher levels of CO<sub>2</sub>, 45 to 75 percent, delayed toxin production. Nevertheless, 75 percent of the atmosphere consisting of CO<sub>2</sub> did not completely inhibit toxin production. In contrast, Lambert et al. (1991c) found the presence of CO<sub>2</sub> in the package head space was not a significant factor affecting time until toxin production. Also levels of CO<sub>2</sub> produced from atmospheres containing O<sub>2</sub>, appeared to enhance toxin production under temperature abuse conditions (Lambert et al., 1991a).

While extremely high doses of radiation are necessary to inactivate the botulinum toxin in foods (Monk et al., 1995). Irradiation, low temperatures for storage, and  $CO_2$  levels also lower and eliminate toxin production. It has also been concluded that fresh meat products treated with low dose irradiation levels should be spoiled prior to production of toxin (Lambert et al., 1991c; and Radomyski et al. 1994). Thayer et al. (1995) also concluded that there was no evidence that the reductions of the indigenous populations of micro-organisms in (MDCM) treated with irradiation increased the potential for the formation of

botulinum toxin. Lastly, refrigerated samples would not become toxic before there were obvious signs of spoilage.

# Irradiation Reduction of Foodborne Parasites

Trichinella spiralis is a nematode which is sometimes present as an encysted larvae in fresh pork muscle. When undercooked pork containing Trichina is eaten a disease known as trichinosis may result in which the digested larvae become free and mature producing a second generation in the thousands within 2 to 4 days. These larvae may spread and encyst within the host's muscle tissues. The severity of trichinosis may range from asymptotic to death.

While dose of 7 to 9.3 kGy are required to kill *Trichinella spiralis* (Monk et al., 1995), low doses of irradiation have proven effective in inactivating the development, growth, and reproduction of adult larvae (Lee et al., 1995; Monk et al., 1995; Taylor and Parfitt, 1959; and Thayer et al., 1993b). The USDA regulations (1985) allow a dose of 0.30 to 1.00 kGy to control *Trichinella spiralis* in fresh pork. While this process is less expensive than cold storage required to produce *Trichinella spiralis* free pork, the doses allowed merely prevent maturation of the larvae. Unfortunately this does not prevent the initial phase of trichinosis associated with the release of the ingested organisms in the intestine and therefore, may be inadequate as a public health measure (Urbain, 1978).

Other parasites which are effected by irradiation include *Toxoplasma* gondi, Cysticercus bovis, and Cysticercus cellulosae. Toxoplasma gondi is a protozoan parasite that can be transmitted to man in raw or undercooked beef,

mutton, and pork. This leads to a disease known as toxoplasmosis, which is a common infection in man that may lead to pneumonitis. The cestoda *Cysticercus bovis* and *Cysticercus cellulosae* are the larval forms of beef and pork tapeworms, respectively. The mature worms are also noted as *Taenia saginata* (beef tapeworm) and *Taenia solium* (pork tapeworm). *Toxoplasma gondi, Cysticercus bovis*, and *Cysticercus cellulosae* may all be effectively rendered incapable of development with 0.25 to 0.60 kGy, thus eliminating infections in man (Monk et al., 1995; Radomyski et al., 1993; Taylor and Parfitt, 1959; Thayer et al., 1993b; and Urbain, 1978). If pork is irradiated as permitted by the USDA for control of *Trichinella spiralis*, then *Toxoplasma gondi* and *Cysticercus cellulosae* will also be inactivated (Thayer et al., 1993b).

*Eschinococcus granulosus* are parasites which are easily seen in meat during meat inspection and thus the rejected offals are feed to dogs. The parasites then may be transmitted to humans through the dog's feces. While Taylor and Parfitt (1959) discovered *Eschinococcus granulosus* larvae were inactivated with a 0.10 kGy dose of irradiation, they concluded that offals would be unlikely to be irradiated. Thus, elimination of *Eschinococcus granulosus* with the application of irradiation seemed futile to the authors.

### Irradiation Effects on Molds, Yeasts, and Viruses

Molds are generally more resistant to irradiation than are bacteria. Conversely, irradiation has a significantly lethal effect on yeasts (Monk et. al., 1995). Doses of 3.5 to 7.0 kGy are required to inactivate molds such as *Aspergillus, Penicillium, and Rhizopus spp.* in many food products (Monk et al., 1995). While irradiation reduces the mold populations in foods, there appears to be some uncertainty about the effects of irradiation on subsequent production of myotoxins by survivors. Also, any mold surviving irradiation should be expected to grow very rapidly because of the lack of competitors allowing it to eventually dominate the microflora.

For fresh meats the dose requirements are too high (<50 kGy) to allow serious considerations of irradiation inactivation of the foot and mouth disease virus (Urbain, 1978). While the foot and mouth disease virus is mainly in livestock animals of other countries, numerous enteric viruses of customer concern include polivirus, Coxsackie virus, echovirus, hepatitis A virus, and Norwalk virus may be found in shellfish of polluted oceans and waters. A  $D_{10}$ values of 2.0 and 2.4 kGy have been observed for hepatitis A virus and rota virus, respectively (Monk et al., 1995). The authors also noted that a 100 fold reduction of poliovirus in fish fillets has been observed after a 6 kGy dose. Satin (1993b) concluded that irradiated shellfish would be the same in every way as untreated shellfish, except that the risk of hepatitis, cholera and other diseases would be minimized.

# The Effects of Ionizing Radiation on Fresh Meat

### The Regulatory Status of Irradiation in the U.S.

The use of irradiation as a food preservation technique has been researched in the U.S. as well as numerous other countries since World War II. The use of ionizing radiation in the preservation of foods gained it's greatest driving force when President Eisenhower proposed the Atoms for Peace Program to the United Nations in December of 1953 (Dempster, 1985). This lead to a great amount of research being done by and on behalf of the U.S. military to produce high quality foods with an extended shelf life. Unfortunately, most of the doses being used were at sterilizing doses where unfavorable sensory qualities developed. On the fortunate side, irradiation can preserve foods, decontaminate foods, control maturation, alter the chemical composition, provides no toxic residue in foods, and maintains most of the nutritive value of foods (Urbain, 1989).

There are many advantages to the use of irradiation in the preservation of foods. Because irradiated foods are typically packaged prior to the application of irradiation the possibility of cross-contamination is greatly reduced. Also the low costs of the process, which has been estimated at 0.5 to 1¢ per pound, and the low amount of energy required for radiation in comparison to conventional heat and freezing processes adds to the advantages of irradiation (Kampelmacher, 1983). Cost benefits by the USDA indicate benefits of irradiation would likely exceed the cost by a ratio of 2.2 - 2.8 to 1 and that the irradiation of just 10% of the U.S. poultry would produce annual savings of up to 50 million dollars (Loaharanu, 1994). Also, the incidence of foodborne disease remains largely unknown as most cases are not reported. Thus, the potential role of food irradiation in reducing those costs are not fully apparent.

The World Health Organization (WHO) in 1981 concluded that the irradiation of any food commodity up to an average dose of 10 kGy presents no toxicological hazard. Their radiation chemistry studies showed that radiolytic products of foods were identical, regardless of the origin of the food. Also, the radiolytic compounds identified from irradiated foods have been identified previously in foods which have been subjected to other accepted types of food processing. Consequently, the use of food irradiation has been endorsed by

designated experts from 57 countries, however, only 37 countries have allowed the use of this technology for treating one or more food items for consumption (Loaharanu, 1994).

The use of irradiation as a food preservation technique has been approved in the U.S. for pork and poultry. The Food and Drug Administration (FDA) amended the food additive regulations to permit the irradiation treatment of pork to control *Trichinella spiralis* (USDA, 1985) at dosages between 0.3 and 1.0 kGy. Poultry has also been permitted to be irradiated with a minimum dose of 1.5 kGy and a maximum dose of 3.0 kGy (USDA, 1992). The seemingly low doses allowed for irradiation of pork and poultry in the U.S. stem from concerns of irradiation reducing spoilage micro-organisms while allowing germination of *Clostridium botulinum* spores and production of toxin. The same concerns of *C. botulinum* toxin production led the FDA and USDA to allow only aerobic packaging of irradiated poultry. Consideration of the safety for consumption of irradiated foods, the areas of radiological safety, toxicological safety, microbiological safety, and nutritional adequacy required testing by the FDA (Pauli and Tarantino, 1995).

In the U.S., irradiation is classified as a food additive and is thus regulated by the FDA. Also, labeling of irradiated pork and poultry requires the "radura" symbol as well as statements such as "treated with ionizing radiation" or "treated by irradiation" (Nielsen, 1987; Pauli and Tarantino, 1995). If irradiated ingredients are added to foods that have not been irradiated, no special labeling is required (Pauli and Tarantino, 1995). While there are numerous advantages to the process of food irradiation, little is known about this technology by consumers. Kampelmacher (1983) concluded the main reason for the lack of acceptance of the process by consumers and governments is probably the emotional resistance against nuclear energy.

# **Consumer Awareness and Acceptance of Irradiated Foods**

The availability of irradiated food products is very limited. Also, many consumers perceive irradiated products in a negative connotation based on their knowledge of nuclear weapons and nuclear energy. Nonetheless, recent events involving foodborne disease and microbial contamination of meat products has reemphasized the importance of irradiation technologies in reducing pathogenic and spoilage organisms to produce a wholesome food supply. Results of a consumer study by Resurreccion et al. (1995) indicated that the market potential for irradiated muscle foods would far exceed that of produce when based on consumer attitudes. A store owner in Pszczola's (1993) article pointed out a bad melon is easy to tell, whereas food pathogens are imposible to detect. Thus, four retail stores have been successfully selling irradiated chicken with a significantly reduced potential for salmonellosis and other foodborne illnesses (Pszczola, 1993).

Because of a limited supply of irradiated products and bad perceptions of the words "irradiated" and "radiated", the key to successful marketing of irradiated meats is probably consumer education. Numerous studies have demonstrated that acceptance of irradiated products increases when consumers are provided with information about the specific advantages of the irradiation process (Bruhn, 1995). Consequently, consumer awareness of irradiation processing had increased from 23% in 1984 to 60% in 1989

(Resurreccion et al., 1995) allowing consumers to become further educated about irradiation processing of foods.

Concerns about irradiated products have also been exploited by the media and activist groups. Typically, attacks of irradiation processing by the media and activist groups have lacked proper information and education, and are based on single agendas rather than the feelings of consumers as a whole. (Satin, 1993a). Lagunas-Solar (1995) pointed out concerns are being exploited by consumer activist groups by using the generalized misconceptions linking radiation to cancer and death. Also, the media's use of sensationalism rather than responsible journalism has lead to misconceptions about irradiation.

Unwarranted concerns of certain activists groups deal with the irradiation of spoiled food. These activist think spoiled food can be made to taste like a fresh wholesome product when it is irradiated. Satin (1993a) reported that you can not make spoiled food fresh by irradiating it. Nevertheless, many activist groups feel that good manufacturing practices may be disregarded if the product is to be irradiated. It is most likely that product to be irradiated will be of the highest quality since the irradiation process is a value added process. Also, contamination does not simply refer to high bacteria counts. Yogurt, certain cheeses and fermented sausages and other foods have high levels of bacteria. Contamination, thus refers to bacteria or foreign objects which exert some negative effect on the food and to those consuming it. Contaminated foods lose taste, texture, proper smell or good appearance, while they can also transmit disease. Thus, irradiation only prevents spoilage, it can not hide it.

In a consumer study Resurreccion et al. (1995) found that over 30% of consumers believe that irradiated foods are radioactive. But consumers over

the past ten years are less concerned about irradiation than they are about food additives, pesticide residues, animal drug residues, growth hormones, and microbial contamination (Bruhn, 1995; Resurreccion et al., 1995). While the number of incidences of microbial food contamination increase, consumers are more likely to accept food irradiation as long as they understand the chemical changes occurring in the irradiated products. Lagunas-Solar (1995) showed that public health and safety concerns center mostly on the chemical effects caused by the absorption of radiation energy, in particular, toxic radiolytic products, decreasing nutritional value, and modification of sensory properties. Lastly, the risk of workers becoming ill, environmental pollution and increasing food prices were of more concern to consumers than the food becoming radioactive (Resurreccion et al., 1995).

#### **Physical Effects of Irradiation on Fresh Meat**

Irradiation can have many effects on enzymes which have an active part in the proteolysis of meat. Enzymes within fresh meats can lead to the degradation of intermediate and thin filament of the myofibril which leads to increased tenderization (Huff-Lonergan et al., 1996). Enzymes may also lead to free amino acid build up, off-flavor, and off-odor development. Chiambalero et al. (1959) noted that total proteolytic activity appeared to be higher in pork than in beef. Drake et al. (1961) found that refrigerated temperatures minimized proteolytic activity of beef irradiated at 45 kGy.

Still, enzymes remaining within meat systems after irradiation have been noted for playing an important role in the development of irradiation odor of fresh meats. Lynch et al. (1991) reported that the irradiation odor of meats

might be composed of 2 elements, one being that of protein denaturation producing sulfurous compounds. It was also thought that enzymes remain active after irradiation which could lead to further proteolysis and a build up of free amino acids in the meat, leading to off odors. Drake et al. (1961) also discussed how during unrefrigerated storage of irradiation sterilized (45 kGy) raw ground beef the action of endocellular cathepsins caused off flavors.

Raw meat normally can not be stored for extended periods at refrigerated storage due to food spoilage microorganisms caused off-odors and off-flavors and texture degradation caused by the presence of proteolytic enzymes. While there is still some question to the effectiveness of irradiation in reducing active enzymes within meat, most researchers have found some interaction. Consequently, irradiation can reduce proteolytic enzymes within muscle foods, which may reduce the aging process and tenderization processes within meat. Lakritz and Maerker (1988) stated there is a negative relationship between increasing dose and enzymatic activity. Thus, the higher the dose, the greater the destruction of more proteolytic enzymes. The authors went on to indicate that between 1-10 kGy low level ionizing radiation can reduce the activity of some endogenous proteolytic enzymes in muscle. The decrease in activity is of course dose and enzyme activity dependent.

Because proteolytic enzymes remain active in meat and irradiation can reduce the active enzymes, numerous researchers have looked at the amount of proteolytic activity and tenderness of muscle foods to determine the effect of irradiation on enzymes. Drake et al. (1961) found proteolysis was evident in beef steaks receiving 5 kGy, while radiation induced proteolysis was not extensive. Twenty kGy had no effect on reducing proteases at -80°C for beef, while pork and chicken proteases were reduced by 13 percent (Elias, 1985). Lakritz and Maerker (1988) reported at 10 kGy the enzymatic activity of  $\beta$ glucuronidase was not affected, acid phosphatase activity was reduced by 8% and general proteolytic activity was reduced by 42%. In contrast, Chiambalero et al. (1959) stated 50 kGy had no significant effect on the proteolytic enzymes of beef and pork while Groninger et al. (1956) showed that sterilizing doses were not able to deactivate the succinoxidase system of beef, pork, and fish.

Procter et al. (1952) described how free radicals produced by irradiation of fresh meats can oxidize enzymes and flavor compounds within the meat. Consequently, irradiation can form radicals which damage enzymes as well as split the enzyme. Yang and Perng (1995) suggested that the permeability of the sarcoplasmic reticulum and nuclear membranes in shrimp muscles remained in tact after 5 kGy irradiation allowing functional release of calcium ions for 8 days at 4°C. The controls degraded and became more tender. Calcium ions play an important role in meat tenderization during postmortem aging, causing fragmentation of the myofibril by enzymes (Huff-Lonergan et al., 1996). It can be assumed that irradiation damaged enzymes which typically degrade the sarcoplasmic reticulum and other nuclear membranes are not as functional after irradiation.

Irradiation of fresh meats can cause changes within the structure of meat, reduce nutrients and have effects on water holding capacity and pH. Irradiation with a dose of 10 kGy only produces a 2.4°C increase in 1 kg of food with the heat capacity of water(Lagunas-Solar, 1995). This is about 3 percent of the energy required to boil one liter of water at 100°C. Thus, heating and conventional cooking result in substantially higher amounts and concentrations of free radicals than irradiation. This also accounts for limited

color and textural differences between irradiation and conventional thermal processing.

Satin (1993a) recorded some nutrient and vitamin loss by irradiation of fresh and processed meats. Elias (1985) compared the nutritional losses by irradiation of meats to commercial preservation techniques and found irradiation comparable or less. While certain vitamins are very stable to thermal and irradiation processing, others are more susceptible to damage. According to Groninger et al. (1956) and Lagunas-Solar (1995) riboflavin, pyridoxine, and niacin are relatively stable to irradiation in beef, pork and poultry while thiamine was very labile to irradiation and thermal processing.

Ionizing radiation has the ability to split off atoms from molecules or to split molecules into smaller molecules, thus creating free radicals. The same principal of this ability which forms radicals and disrupts the DNA of bacteria and other living organisms holds true for disrupting or denaturing protein substances. Irradiation can denature or break apart myofibril filaments as well as collagen, therefore making muscle foods slightly more tender (Taub et al., 1979). Groninger et al. (1956) reported small textural changes resulted in the sterilizing radiation of meats.

In a later study of the effects of irradiation on the structure of the myofibril, Lakritz et al.,(1987) found at 10 kGy minimal changes occured in the muscle structure of beef, but at levels above 30 kGy at 0 to 4°C major increases in myofibril fragmentation and decreases in tensile strength of raw and cooked muscles were noted. A decrease in myosin content was also found, while increasing dosages enhanced fragmentation of the myofibrils. The authors also mentioned that sarcomere length before and after irradiation remained

constant. Lescano et al. (1991) showed irradiated chicken breasts were more tender than controls.

Fresh meat, and beef in particular, is aged to increase tenderness. Previous tenderization techniques included "dry aging" beef by allowing carcasses or cuts to hang in coolers for numerous weeks. Thus, proteolytic and microbial proteolytic enzymes were active and the meat became more tender. Because of the vast quantity of cooler space required most processors have changed to a process of "wet aging" in which product is cut down to wholesale and/or retail cuts and packaged in vacuum bags. The product then becomes more tender as proteolytic enzymes become active without the presence of air. Wet aging also minimizes protein degradation by bacteria due to anaerobic conditions.

Drake et al. (1961) reported the typical "aged" meat flavor was not present in irradiated steaks possibly because of reduced bacterial proteolysis. Thus, dry aged beef has a very recognizable and distinct flavor. In a further study in this area, Lee et al. (1996) discovered aging 2 kGy irradiated prerigor beef at 30°C for 2 days in MAP resulted in similar Warner-Bratzler shear values as beef conventionally wet aged at 2°C for seven and fourteen days. The increased tenderness here is probably because of slight fracturing of the myofibril as well as the meat being prerigor. Also, the use of only 2 kGy probably had only limited effects on proteolytic enzymes. Nevertheless, it is very unlikely that prerigor meats will be irradiated on a commercial basis, other than for sausage manufacturing.

Fracturing of the myofibril and other structural changes within meat may affect the water holding capacity of meat. Lescano et al. (1991) stated the water holding capacity of chicken breast was reduced by 2.5 kGy, but higher

doses enhanced water holding capacity. Rhodes and Shepherd (1966) pointed out irradiation at 4 kGy of beef and lamb led to an increase in weep within packages. Nevertheless, Lakritz and Maerker (1988) noted the pH of meat was unaffected by irradiation. This is somewhat surprising since higher pH's within meat result in increased water holding capacities. Thus, the decreased water holding capacity of irradiated fresh meats might be caused by radiation denaturation of the myofibril. In contrast, Heath et al. (1990) indicated irradiation at 1, 2, and 3 kGy reduced cooking loss by 6.1, 3.6, and 3.7 percent respectively, in chicken breast tissues which had not been aged. This is most likely explained by to the product loosing weep due to decreased water holding capacity after irradiation, prior to weighing before cooking.

#### Irradiation Induced Chemical Changes on Fresh Meat

Ionizing radiation causes numerous chemical changes within fresh meats and other food products. Radiation may cause peroxidation of lipids, increase free fatty acids within foods, break peptide bonds, and split apart proteins, as well as create radiolytic compounds which become free radicals. Irradiation can cause chemical changes of meat such as deamination, decarboxylation, reduction of disulfide linkages, oxidation of disulfhydryl groups, amino acid side group decomposition, increase or decrease in peptide linkages, and change in valence state of metal ions (Taub et al., 1979).

Studies with meats have shown that the origin of radiolytically induced compounds can be attributed to precursors in the meat such as protein and fat (Merritt et al., 1985). When lipids or triglycerides are irradiated various hydrocarbons are produced from the fatty acids as well as carboxylic acid

radicals and  $CO_2$ . When proteins are irradiated, sulfur containing compounds, aromatic hydrocarbons and  $NH_3$  may be produced (Merritt et al., 1978a). Also, water may form ions and radicals such as  $H_3O$  and OH which induce further reactions. Oxygenated compounds such as alcohols and carbonyl compounds are only produced by irradiation from meats in small amounts. When triglycerides are irradiated, some of the major stable products formed are hydrocarbons from the loss of  $CO_2$  and  $CH_3COOH$  in various free radical reactions (Morehouse et al., 1993).

The most abundant radiolytic hydrocarbons are formed during various free radical reactions as a result of the loss of  $CO_2$ . Primary free radicals can undergo many reactions to form secondary radicals and other stable products (Morehouse and Ku, 1992). When lipids are treated with ionizing radiation a series of radiolytically generated hydrocarbons are formed from the decarboxylation (n-1) and deacetylation (n-2) of the fatty acids (Morehouse et al., 1993; Morehouse and Ku, 1992). Thus irradiation forms a series of saturated and unsaturated hydrocarbons from termination of alkyl radicals (from the parent fatty acid). Of the radiolytically generated hydrocarbons, the decarboxylation products predominate and constitute the major hydrocarbons formed from triglycerides (Morehouse and Ku, 1992).

In studying the effects of irradiation on shrimp fats Morehouse and Ku (1992) found shrimp fatty acids, which are highly unsaturated, form the hydrocarbons pentodecane, 8-pentodecene, heptodecane, 8-heptodecene, and 6,9-heptodecadiene. Thus, irradiation of unsaturated fatty acids can lead to many different hydrocarbons. Also, irradiation of unsaturated fats can lead to hydroperoxides which decompose into aldehydes, alcohols, ketones and other carbonyl compounds.

In studying the effects of irradiation on meat structures, researchers have reported a variety of chemical changing responses. Taub et al. (1979) reported radiolytic effects on connective tissue proteins, can lead to some degradation in peptide chains or in cross-linkages, thus increasing collagen solubility. Analysis of amino acids of beef irradiated at -30°C at a dose of 47 to 72 kGy showed no detectable difference compared with unirradiated controls. Batzer et al. (1959) discovered irradiation increased amounts of hydrogen sulfide, methyl mercaptan, acid-salt soluble carbonyl compound and pH. Coleby et al. (1961) noted irradiation at 25 kGy destroyed 42 and 43 percent of the glutathione in raw beef and pork respectively at 0°C. It was not until a temperature of -196°C was employed during irradiation was that over 90 percent of the glutathione remained. If irradiation destroys a portion of meat glutathione, this reduction could disrupt the natural biochemical antioxidation properties of meat, there by allowing increased peroxide formation. Also, Groninger et al. (1956) showed the porphyrin ring of the hematin compounds was stable to radiation at 0 to 9.3 kGy.

Temperature variations in respect to irradiation treatment of meat products is used in two different ways. First, meat products may be heated or cooked prior to irradiation. Thompson et al. (1961) found heating beef to 150°F prior to irradiation inhibited the release of amino acids from parent proteins. Cooking shrimp before or after irradiation neither increased or decreased the quantity of radiolytic hydrocarbons in irradiated as well as control shrimp (Morehouse and Ku, 1992). Thus, cooking cannot disguise the effects of irradiation. The other use of temperature in irradiation processing of meats is either freezing the product or using refrigerated temperatures during irradiation. Coleby et al. (1961) found that the actual temperature during irradiation influenced the degree of protection from chemical change. While only having a minimal effect, low temperature decreased the radiolytic yield during and after irradiation of shrimp (Morehouse and Ku, 1992).

Radiolytically induced hydrocarbon yields arising from fresh meats have been shown to be dependent upon the fatty acid composition of the meat (Merritt et al., 1985 and 1978a; Morehouse et al., 1993; and Morehouse and Ku, 1992). Also, researchers have shown that the amount of these radiolytically generated hydrocarbons increases with absorbed dose (Morehouse et al., 1993; and Morehouse and Ku, 1992). Therefore, as dose increases on product with the same fat content, so does the quantity of hydrocarbons.

Irradiation of meats has been known to create peroxides for years. Hydrogen is readily available in irradiated meat products because irradiation can cause further unsaturation of hydrocarbons as well as produce  $H_2$  from bond cleavages. Consequently, oxygen becomes a limiting factor in the irradiation formation of peroxides in fresh meats containing fat. Also, the amount of peroxides within fresh meats are typically used as indicators of the quantity of oxidation as well as in determining the rancidity of fresh meats. Lea et al. (1960) reported irradiation induced oxidation, as indicated by the peroxide values, which were greatest in the proximity to the surface. Irradiation in  $N_2$  MAP and then stored in air greatly reduced development of peroxides in fat when compared to aerobically packaged and irradiated products (Lea et al., 1960).

Therefore, aerobically packaged meat product when irradiated have increases in peroxide values as well as accelerated oxidations during storage (Groninger et al., 1956; Lea et al., 1960; and Lefebvre et al., 1994) Irradiation of meat products with the exclusion of  $O_2$  during irradiation either through vacuum packaging or  $N_2$  MAP inhibits peroxide formation (Groninger et al., 1956; Hansen et al., 1987; Rhodes and Shepherd, 1967; and Taub et al., 1979). Thus, most of the peroxides in vacuum packaged or MAP irradiated fresh meats should be formed prior to irradiation. This especially holds true when products are packaged and enough time is allowed for the enzymes of meat to use up any oxygen within the package. Also, Groninger et al. (1956) wrote that higher peroxide values of irradiated pork was undoubtedly due to greater unsaturation of the lipids.

Peroxide amounts formed or oxidation within lipids can be expressed using TBA or TBARS values. Ahn et al. (1993) showed how the TBARS values of cooked patties increased as the degree of oxidation of meat increased. Nonetheless, Heath et al. (1990) and Lambert et al. (1992a) reported no significant differences in TBA values of fresh poultry and pork due to irradiation. While Heath et al. (1990) used aerobic packaging it should be noted that Lambert et al. (1992a) used MAP only.

While irradiation typically causes the formation of hydrocarbons and radicals from triglycerides, free fatty acids may be formed. This may take place by proper splitting with ionizing radiation or through radical reactions. Thompson et al. (1961) indicated a dose of 1 and 5 kGy produced free fatty acids in beef. Nevertheless, Rhodes and Shepherd (1967) and Lefebvre et al. (1994) showed that free fatty acid values were not different due to irradiation.

### Irradiation Production of Radiolytic Volatiles in Meat

Radiolytic volatiles consist of compounds created by irradiation mostly in gas forms which dissipate when exposed to air. Consequently, volatiles

trapped within vacuum or modified atmosphere packaging either dissipate or react very fast when the packaging is opened and exposed to air. Merritt et al. (1978b) and Schreiber et al. (1993) have reported finding over 100 different volatiles in products of irradiated meat.

Therefore, there are numerous volatile compounds produced in a variety of amounts by irradiation. Merritt et al. (1975) reported the various trace volatile compounds produced by irradiation of several meats to consist of predominantly hydrocarbons, sulfur compounds, certain alcohol, and carbonyl compounds. Batzer and Doty (1955) found gases produced by a 14.9 kGy dose on beef to contain hydrogen sulfide, methyl mercaptan, and other sulfur containing compounds. Analysis of radiolytic volatiles of meat have also indicated levels of octane, 1-octene, hexanal, and nonane (Hansen et al., 1987). Acetaldehyde, acetone, methyl ethyl ketone, methanol, ethyl alcohol, methyl mercaptan, dimethyl sulfide, dimethyl disulfide, ethyl mercaptan, and isobutyl mercaptan have likewise been found in irradiated beef (Merritt et al., 1959).

The principle products of irradiated fresh meats are hydrocarbons such as alkanes, alkenes, alkynes, and alkadienes. Merritt et al. (1978b) noted 95 percent of the total volatile compounds in irradiated meats are constituted by alkanes and alkenes. This holds true for meat products which are not low in fat quantity. As fat quantity decreases in irradiated meats, so does the amount of hydrocarbons. Thus, the quantity and quality of hydrocarbons produced by irradiation vary with fat composition. For instance, Burks et al. (1959) indicated ammonia was 92 to 95 of the total volatile bases in 23 and 37 kGy irradiated beef, respectively. This result is most likely because the beef was very low in fat percentage (2 to 3%). Another class of compounds found in

abundance among irradiation of triglyceride products in various meats is propane dioldiester (Merritt et al., 1985).

Radiolytic volatiles of irradiated meat consist of many compounds which are continually changing. They consist of compounds such as ketones, aromatics, aldehydes, or sulfur compounds which produce various off odors. They may also react with other substances to form highly odorous compounds. Consequently, volatile bases produced by irradiation of meats are partial contributors to the odor of irradiated beef (Burks et al., 1959).

When meat is irradiated, hydrocarbons and oxygenated compounds are formed predominantly from lipids and the sulfur compounds are formed from protein (Hansen et al., 1987). Nevertheless, because oxygen is a rate limiting factor in the oxidation of fats and hydrocarbon products, oxygen content can vary the amount of volatile production. Hydrogen sulfide formation has also been found to be independent of the presence of oxygen (Batzer and Doty, 1955). Ionizing radiation results in the formation of highly reactive free radicals and hydrogen peroxide (Kilcast, 1990). Bond rupture in a triglyceride occurs preferentially at the bonds adjacent, or near to the ester linkages. If rupture occurs at the  $\alpha$ Carbon to the carboxyl group, the predominant compounds would be expected to be the alkanes and alkenes having one less carbon atom than the corresponding fatty acid (Merritt et al., 1975). The next most preferred cleavage is at the  $\beta$ carbon to the carboxyl which leads to alkanes and alkenes having two less carbon atoms than the corresponding fatty acid (Merritt et al., 1975).

In general radiolytic volatile compounds are found in beef, pork, mutton, lamb, veal, and poultry in about the same proportions, when irradiation occurs at the same temperature and dose (Merritt, 1972). Radiolytic volatiles are many of the same volatiles found in thermally processed meats. Using 50 kGy to sterilize beef, Wick et al. (1965) showed that non-irradiated samples had many of the same compounds as irradiated samples, just in smaller amounts. Merritt et al. (1959) described the same trend in irradiated and non-irradiated beef.

Irradiation induced volatiles in meats typically follow certain trends. As temperature of the product rises at the point of irradiation, radiolytic volatile yields increase (Merritt et al. 1978b and 1975). As the dose increases, so does radiolytic yield. The relationship of dose and volatile yield has been shown to be a linear function (Merritt et al., 1978b; and Morehouse et al., 1993). Also, some researchers have shown that volatiles may be reduced in quantity during subsequent storage after irradiation. According to Wick et al. (1965) the n-alkanals and methional are major volatiles component of freshly irradiated beef and minor components of six month stored and irradiated beef. Hansen et al. (1987) also reported the amount of total volatiles was greater in fresh irradiated samples than in samples stored for six months. Thus, it appears there may be some dissipation of many radiolytic volatiles of meats if they are packaged in containers or packaging which allows some gas exchange.

# **Identifying Irradiated Fresh Meat**

Treatment of food products with ionizing radiation reduces food pathogens and increases the shelflife of products. Therefore, irradiation is a value added process. There is a need to be able to identify irradiated food products to prevent the mislabeling of unirradiated products as irradiated foodstuffs. Also, there is a need to prevent irradiating products more than once to minimize toxicological hazards to the public. Various researchers have reported different means of identifying irradiated meats as well as indicating the dose applied.

Using gas chromatography for evaluation of irradiated chicken, pork, and beef, with hydrocarbons as markers Schreiber et al. (1993) was able to identify irradiated from non-irradiated samples correctly 98.3 % of the time, three to six months after irradiation. Using hydrocarbons as markers for irradiation with gas chromatography is only possible if the fatty acid composition of the irradiated product is known. Also, this technique was not able to be used for dose estimations. Morehouse and Ku in 1992 also noted that the absence of radiolytically generated hydrocarbons is a good indication that fresh shrimp have not been treated with ionizing radiation, whereas the presence of hydrocarbons is a good indication that shrimp have been irradiated.

Looking for other markers of irradiation with the use of gas chromatography, Furuta et al. (1992) found the level of carbon monoxide (CO) could be used as a probe in irradiated frozen meat and poultry. This technique is only affective in frozen meats since frozen products retain the CO gas for up to a year whereas the gas is released from refrigerated meats. This method is comparable to the ESR method with respect to sensitivity and the detectable period, but it has a distinct advantage of being useful for boneless products also.

Electro Spin Resonance (ESR) spectroscopy exhibits great promise for the identification of bone containing foods that have been treated with radiation. When bone is irradiated, a characteristic ESR signal develops and is easily monitored. The relative intensity of the ESR signal is dose dependent and displays a linear relationship to absorbed dose (Morehouse et al., 1993). Thus, ESR technique has been used to identify irradiated shell-fish and meats containing bone (Morehouse and Ku, 1992; and Stevenson and Gray, 1990).

There are factors such as the degree of ossification within bones and temperature which affect the ESR signal. Stevenson and Gray (1989) reported the ESR signal strength increased significantly as irradiation dose increased. Also, bones stored at 5°C showed a significantly greater reduction in free radical concentration than those stored at -20°C. Therefore the degree of calcification of the bones at different ages may influence the ESR signal strength because it is thought that the signal arises from structural defects in the crystal lattice of the hydroxyapatite of bone (Stevenson and Gray, 1989).

# Gamma versus Electron Radiation

Electron radiation or  $\beta$  radiation involves the application of accelerated electrons onto the face of a product. Electrons produced from a Van de Graaff generator slow down rapidly as they enter food products. The absorbed dose increases underneath the surface of the product, while the electrons moving further into the product move more slowly with less energy being absorbed (Olson, 1995). As electron radiation penetrates a food product two possibilities for energy disbursement exist. Elastic scattering occurs when electrons are deflected by the electrostatic field of an atomic nucleus (Woods and Pikaev, 1994). Elastic scattering involves scattering of the radiation without loss of energy. Secondly, when electrons come in contact with an electrostatic field and result in ionization there is an energy loss which results from an absorption of a dose. This ionization results in the formation of free radicals. Electrons of higher level energies can penetrate into products to a greater depth. Electron accelerators used in irradiation of foods have a maximum energy level of 10 million electron volts (MEV). Also, to be affective at least 5 MEV must be used to produce a dose to penetrate foods. At 10 MEV and irradiating both sides of a food product the greatest penetration a dose will have is 8.9 cm (3.5 inches) (Olson, 1995).

Gamma ( $\gamma$ ) radiation and X-rays consist of photons rather than electrons. Both gamma rays and X-rays while slightly different have lower energies in comparison to electrons, although they have a deeper penetrating ability. The absorbed dose from photons is highest at the surface of the product and diminishes exponentially as it penetrate through the product (Olson, 1995). Thus the absorbed dose of gamma and X rays are measured in a maximum-minimum ratio. To receive a better max/min ratio, products treated with X-rays and gamma rays are typically irradiated on both sides.

When photons come into contact with the product being irradiated various reactions may happen. Coherent scattering involves photons being scattered with little loss of energy. The photoelectric effect results from a photon ejecting a single electron from an atom of the stopping material. Where Compton scattering occurs, a photon interacts with an electron so that the electron is accelerated and the photon is deflected with reduced energy. Paired production involves the complete absorption of a photon in the vicinity of an atomic nucleus (Woods and Pikaev, 1994). The Photoelectric effect, Coherent scattering, Compton scattering and paired production all result in the formation of radiolytic free radicals.

Electron and photon radiation of foods result in two reactions, the formation of free radicals and solute molecules, or the formation of two free

radicals. The formation of two free radicals such as  $H \cdot + \cdot OH \rightarrow H_20$  is typical of high dose rate applications as with electron irradiation and at frozen temperatures (Diehl, 1982). The author also went on to write that second and tertiary radicals as with gamma radiation and X rays will react exclusively by a bimolecular termination reaction, therefore only a slight dose rate effect is discernible. Consequently, there should be little or no difference in radiolytic yields between electron and gamma irradiation. Studies on radiation sterilized meats using both electrons and photons confirmed there was no difference in radiolytic yields between  $\beta$  and  $\gamma$  radiation as well as X rays (Hannan and Shepherd, 1959; and Merritt et al. 1978a and 1978b).

#### Irradiation Effects on Meat Color

Irradiation of fresh meats typically causes a darkening of lean color. In 1959 Batzer et al. noted irradiated beef steaks were always darker than unirradiated controls. Groninger et al (1956) also reported with increasing radiation dosage the red color of beef was changed to a dull red and at 279 kGy to a tan color. When fresh meat products are vacuum packaged oxymyoglobin, which leads to the typical red color of beef, changes to deoxymyolobin which is a purplish red color. Hannan and Shepherd (1959) found with the absence of oxygen in irradiated chicken samples caused various shades of brown and green, presumably due to oxidative breakdown of the myoglobin.

When vacuum packaged beef is irradiated, its color changes to brown and represents a change in the trivalent iron of metmyoglobin and oxidation by a hydroxyl radical resulting in the loss of  $O_2$  (Thayer et al., 1993). Thus, the reduction of deoxymyoglobin to metmyoglobin during irradiation is caused by a small percentage of electrons reacting with the pigment. When exposed to air, a portion of metmyoglobin is gradually converted back to oxymyoglobin. The sensitivity of radiolytically reduced deoxymyoglobin to oxidation might be very dependent on the conformation of the denatured pigment (Taub et al., 1979). Ginger et al. (1955) also mentioned the reactions produced by ionizing radiation would favor the oxidation of free iron and of iron in cytochrome c. The authors went on to state that it is possible the reduction of metmyoglobin to oxymyoglobin was dependent on the existence of redox conditions. Thus the available information suggests that the heme as well as the protein moiety are adversely affected by irradiation (Clarke and Richards, 1971).

The main effect of irradiation of raw beef samples noted by Batzer et al. (1959) was the production of a red pigment, more stable to alteration or destruction either because of its own inherent stability or because of conditions in the irradiated sample. At 40 kGy and in some cases at 20 kGy, the authors reported a bright red pigment was formed, which was similar to oxymyoglobin and was stable at 35 and 60°F. Thus, irradiation has the ability to alter the structure of meat pigments as well as affect the state of the heme iron. The altering of myoglobin by irradiation has been reported to occur at doses greater than 3 kGy (Ginger et al., 1959; Batzer et al., 1959). Also, because beef is the most pigmented of red meats and poultry it is the most susceptible to the effects of irradiation on color changes.

Uncured cooked meats, exposed to a pasteurizing dose of irradiation in the absence of oxygen become pink or reddish. Upon exposure to oxygen the normal brown or gray color of metmyoglobin returns. This sequence of color changes is associated with the reducing action of free radicals leading to a reduced myoglobin derivative. This red myoglobin is red in color and is easily

oxidized to the usual brown color of cooked meats (Urbain, 1978; Thayer et al. 1993b; and Taub et al., 1979). Therefore, fresh vacuum packaged meats contain deoxymyoglobin which is oxidized by irradiation to metmyoglobin, and cooked vacuum packaged meats contain metmyoglobin which is reduced to a pigment similar to oxymyoglobin.

In studying the effects of irradiation on Hunter labscan values Lebepe et al. (1990) reported irradiation increased Hunter labscan 'a' values in vacuum packaged pork while Lambert et al. (1992a) found Hunter labscan L 'a' and 'b' values increased. In contrast, Luchsinger et al. (1995b) found Hunter labscan CIE L\*, a\*, and b\* values in raw ground beef were initially lowered by irradiation, but stabilized during storage. Oxygen within a modified atmosphere packaged product can affect Hunter L, 'a', and 'b' values of irradiated meat also. Lambert et al. (1992a) reported pork samples with 20 %  $O_2$  and irradiated at 1 kGy had higher L values, lower 'a' values, and higher 'b' values compared to controls. This indicated that samples packaged with  $O_2$ and irradiated resulted in more white, less red, and more blue pork. This may be attributed not only to the presence of oxygen which oxidizes myoglobin, but also to the enhanced oxidation of meat pigment when samples were irradiated in the presence of oxygen. Lambert et al. (1992a) also noted meat color of irradiated and non-irradiated pork loins was not affected in 100% N<sub>2</sub> MAP.

While beef may be the most sensitive meat to adverse irradiation induced color changes, Niemand et al. (1981) discovered when 2 kGy irradiated beef cuts were removed from vacuum packaging and allowed to develop a natural color in air, irradiated samples had significantly higher scores than controls based on a hedonic scale. Lefebvre et al. (1994) found the color of the raw irradiated (1, 2.5 and 5 kGy) samples packaged aerobically were considered more pleasant by panelists than that of the fresh references. Using a nine point hedonic scale Rhodes and Shepherd (1967) found irradiation (4.4 kGy) caused only slight adverse effects on the color of green back bacon. Consequently, irradiation has the ability to alter pigments which may or may not be desirable to consumers.

### **Irradiation Caused Off-Odors**

Irradiation of raw meat samples has been shown to produce products which are less pleasant or desirable to trained panels and consumer panels. Irradiation can have an adverse effect on the color of raw meat products; while also affecting the natural occurring odor of fresh meat. Consequently, not only does irradiation cause panelists to score color low, but the odor and aroma of the raw meats are scored lower. Groninger et al. (1956) and Lefebvre et al. (1994) reported that raw irradiated meat samples were consistently less acceptable or desirable by panelists than non-irradiated controls and references. Lescano et al. (1991) also found the irradiation odor of raw meat samples was unpleasant.

Many compounds have been shown to make up or cause the irradiation off-odor of meats. The make up of the irradiation off-odor is dependent upon the type of meat sample being irradiated, package type, headspace composition, and many other factors. For instance, irradiation of a high fat meat in the presence of  $O_2$  would produce numerous hydrocarbons. In 1959 Burks et al. noted it seemed evident that many different compounds are responsible for the odor of irradiated beef. Some compounds such as amines and ammonia may have definite effects on the over all odor when they are in combination with similar compounds, although each may be present in a concentration that would be undetectable if the compound were alone. Some of the compounds responsible for irradiation off-odors of meats include hydrogen sulfide, methyl mercapatan and carbonyl compounds (Dempster, 1985), volatile amines and ammonia (Elias, 1985), and other compounds with active hydrogens, probably sulfur containing compounds (Hedin et al., 1959). Exclusion of  $O_2$  during irradiation should decrease the irradiation off-odor of irradiated meat caused by irradiation decomposition of fatty acid hydroperoxides(Hansen et al., 1987; Huber et al., 1953; Lambert et al., 1992a).

Numerous researchers have worked on pin pointing the exact dose at which an irradiation off-odor exists and other undesirable organoleptic changes take place. Grant and Patterson (1991a) reported a threshold dose of 1.75 kGy for pork while Sudarmadji and Urbain (1972) found the threshold dose for poultry and beef to be at 2.5 kGy. The irradiation off-odor of chicken has been detected at 1 kGy, 2.5 kGy, and 5.0 kGy by Heath et al. (1989), Lescano et al. (1991), and Mercuri et al. (1966), respectively. Lynch et al. (1991) also reported an irradiation off-odor of turkey at 2.5 kGy. The irradiation off-odor of poultry has been characterized as sour, rancid, mature, metallic, sulfur, burnt feathers, and as bad meat. Niemand et al. (1981) found the irradiation off-odor in 2 kGy treated beef while Lea et al. (1960) found the off-odor in .93 and 1.86 kGy irradiated beef.

Lambert et al. (1992a) reported no difference was detectable by the sensory panel between the non-irradiated treatments and the  $N_2$  packaged pork samples irradiated at 1 kGy. Luchsinger et al. (1995b) also found no off odors in either 2.0 or 3.5 kGy irradiated raw ground beef samples. This finding is most likely due to a long period of time transpiring between

removing samples from vacuum packaging and oxygen permeable bags and the sensory panel scoring the samples. Thus, if samples had been scored immediately after removal from packaging there would probably have been a difference than allowing samples to air out.

Many researchers have noticed that after exposing vacuum packaged low dose irradiated fresh samples to air for several minutes that the off-odor diminishes and sometimes disappears (Dempster, 1985; Luchsinger et al., 1995a; Niemand et al., 1981). The same holds true for products stored in high oxygen transmission or permeable packaging, or products stored without packaging (Rhodes and Shepherd, 1967). Also, package types containing a great amount of branched polymers such as polyethylene have lead to taint transfer resulting in off-odors and off-flavors (Trip, 1959).

Many factors have an effect on increasing or decreasing the off-odor of irradiated fresh meats. Researchers have shown that irradiation off-odors increase with the dose applied (Hansen et al., 1987; and Merritt et al., 1975). Also, as the irradiation temperature and storage temperature rises the irradiation off-odors of meat increases (Hanis et al., 1989; and Merritt et al. 1975). The odor intensity of fresh meat has also been shown to increase with irradiation (Lescano et al., 1991) as well as with higher doses and irradiation temperatures (Kosaric et al, 1973a and 1973b).

Irradiation off-odors also have been reported to decrease with storage time (Drake et al., 1961; Mercuri et al., 1966; and Wick et al., 1965). Part of the reduction of off-odors during storage is due to the volatiles escaping through high oxygen permeable packaging. Another part of the reduction of off-odors is the fact that volatiles can form more stable products over time. While Grant and Patterson (1991) reported the irradiation off-odor of MAP pork chops did not change over the storage period, a few researchers have found that off-odors increase in intensity over time. Coleby et al. (1961a) and Lambert et al. (1992a) reported that the irradiation off-odor of treated samples progressively became stronger and less pleasant for panelist during storage, most likely due to the growth of spoilage micro-organisms.

The last factor which affects the irradiation off-odor is cooking. Drake et al. (1961) and Lefebvre et al. (1994) found that the irradiation induced off-odor is significantly reduced by cooking the meat products. Other researchers have found that cooking not only reduces the off-odor of irradiation but it can also eliminate the off-odor of fresh meats (Lescano et al. 1991; Luchsinger et al., 1995b; and Rhodes and Shepherd, 1967).

#### **Irradiation Off-Flavors**

The volatiles which are formed from irradiation of fresh meat products and taint transfer from the irradiation of plastic packaged meat result in offodors which can also lead to off-flavors. Cooking has generally been noted for improving the acceptability of irradiated meats when compared to raw counterparts. Cooking of poultry meat irradiated with 0.5. 1.9, 5.0, and 10.0 kGy was noted by Hanis et al. (1989) to diminish and eliminate the negative sensory effects of irradiation. Nevertheless, researchers such as Coleby et al. (1961) and Tarkowski et al. (1984b) have reported that unirradiated control samples were clearly preferred over irradiated samples of beef and pork based on flavor. It should be noted that many researchers such as Coleby and his associates have reported a "wet-dog" or "metallic" off-flavor when meats are sterilized by using extremely high doses. While low dose irradiation may produce off-flavors, they are not as extreme and intense as those produced at sterilizing doses.

The type of cooking and cooking temperature used in preparing irradiated products may also vary irradiation off-flavors. Hannan and Shepherd (1959) and Hanis et al. (1989) found that steaming irradiated samples led to greater off-odors and off-flavors than stewing or frying samples. It should also be noted that frying and stewing in these cases were done at higher temperatures which can lead to the development of more cooked flavors.

Irradiation off-flavors of meats have been listed as being rancid, metallic, sweet, warm, stale, flat, old, acidic, and wet dog (Risvik, 1986). Typically, at low doses the irradiation off-flavor of meat is less harsh and less noticeable than it is at sterilizing doses. Numerous researchers have investigated the threshold dose at which an irradiation off-flavor appears in individual meat samples. Huber et al. (1953) and Coleby et al. (1961a and 1961b) were some of the first to reveal beef is most sensitive to the development of irradiation off-flavors followed by lamb, veal, chicken, and pork. Rhodes and Shepherd (1966) found the maximum dose which could be applied to fresh beef and lamb in anaerobic packaging without producing off-flavors is 4 kGy; while Sudarmadji and Urbain (1972) found a threshold dose was 2.5 kGy for beef and 6.2 kGy for lamb. Lefebvre et al. (1994) found ground beef developed off-flavors at 1 kGy also, and Luchsinger et al. (1995b) noted 2.0 and 3.5 kGy increased bloody, fat-like, animal hair, and metallic flavors of ground beef.

Sudarmadji and Urbain (1972) also reported a threshold dose of 1.5 kGy for turkey, 1.75 kGy for pork, and 2.5 kGy for chicken. Rhodes and Shepherd (1967) reported 4.4 kGy produced no irradiation off-flavors in bacon, while Mattison et al. (1986) found 1 kGy produced off-flavors in pork loins. Hannan and Shepherd (1959) found 2.3 kGy produced off-flavors in chicken. Lastly, using triangle tests, panelists were not able to distinguish between 2 and 5 kGy irradiated ground turkey and ground beef samples and their counterpart controls (Murano et al., 1995).

Off-flavors are caused by irradiation formed from free radicals oxidizing flavor compounds and meat (Proctor et al., 1952). While most irradiation offflavors originate from substances formed from the meat being irradiated, packaging can create radicals which taint the meat and lead to off-flavors. Tripp (1959) reported that volatiles produced from polyethylene packaging during irradiation of packaged food products lead to taint transfer and offflavors. Meanwhile, Keay (1968) noted taint from polyethylene and polypropylene packaging disappeared after cooking. Nonetheless, researchers such as Proctor et al. (1955) have developed additives like sodium ascorbate which when added to fresh meats destined for irradiation, reduce or eliminated irradiation off-flavors.

Various factors such as dose, temperature, and storage can affect the quantity and quality of irradiation off-flavors of meats. As irradiation dose increases the amount and intensity of off-flavors and off-tastes increases (Cain et al., 1956; Lefebvre et al., 1994; Merritt et al., 1975; and Risvik, 1986). Typically, as temperature of the meat during irradiation increases again, so does the amount and intensity of irradiation off-flavors (Merritt et al., 1975 and Niemand et al., 1981). In contrast, Cain et al. (1956) reported irradiation offflavor was independent of temperature using a dose of 4.65 to 18.60 kGy.

The factor of storage temperature affecting the amount of irradiation offflavors is very much associated with dose. At sterilizing doses a high storage temperature may be used. While using low dose irradiation treatments, fresh meats must still be refrigerated to maintain shelf life. Nevertheless, Coleby et al. (1961) has reported storage of irradiation sterilized meats produced a stronger, more nauseating bitter flavor at higher temperatures (37°C), possibly due to increased proteolysis of the raw products rather than at refrigerated temperatures. Merritt et al. (1975) showed irradiation flavors decreased during storage while Hannan and Shepherd (1959) found storage at 0°C and below had little affect on the off-flavors of irradiation sterilized meats.

Another factor affecting off-flavors of irradiated fresh meats is microbial count and spoilage. Once a fresh meat is spoiled, a typical spoiled and rancid off-flavor develops. If spoiled meat is irradiated, an off-flavor still persists, in which the off-flavor of irradiation is combined with the spoiled off-flavor. This combination of irradiation spoiled meat still leads to a bad taste for panelists (Lefebvre et al., 1994). Irradiation of meat which appeared spoiled and had a  $10^6$  to  $10^7$  g<sup>-1</sup> of spoilage and or pathogenic bacteria produced samples which were not preferred or acceptable by panelists (Grant and Patterson, 1991a). Consequently, the low dose irradiation of spoiled meat will lower microbial counts to acceptable levels, while still leaving the sensory factors of the meat at unacceptable levels.

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# EFFECTS OF LOW DOSE IRRADIATION AND STORAGE TIME ON AROMA AND LEAN COLOR OF RAW BEEF PATTIES IN ANAEROBIC AND AEROBIC PACKAGING

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#### Abstract

The effects of electron beam irradiation, aerobic and anaerobic packaging, and storage times on the lean color and aroma of raw ground beef patties were investigated. Lean trim was coarse ground at 3 days postmortem, then fine ground, pattied and packaged at 3, 6, and 9 days postmortem. Patties were irradiated immediately after packaging or 3 days after packaging at 2 kGy, then stored in a display case between 1 °C and 4 °C for 4 days. Nonirradiated controls were held under similar conditions. After 4 days of storage for each postmortem time, Hunter color and sensory evaluations were performed on all samples. Irradiated beef patties were found to be a darker red color (P < 0.05) than controls by the sensory panel. Hunter "a" value for irradiated patties were lower (P < 0.05) than non-irradiated controls. Irradiated and non-irradiated patties with the shortest postmortem storage times had the most desirable aroma scores (P < 0.05). Anaerobic packaged controls had more desirable aroma scores (P < 0.05) than irradiated patties in anaerobic packaging.

Key Words: Beef patties, irradiation, sensory attributes, color, odor.

## Introduction

Recent events involving the meat industry and food-borne infections have increased industry, governmental and consumer awareness to possible contaminants and pathogens such as Escherichia coli 0157:H7, Salmonella spp, and Staphylococcus aureus. Concerns with the safety of fresh meats have reemphasized the importance of the implementation of technologies useful in prevention or reduction of pathogenic bacteria (Bruhn, 1995). While not a new technology, irradiation has proven to be effective in reducing pathogenic bacteria and gram-negative micro-organisms while extending shelf life (Ehioba et al., 1988; Monk et al., 1995; Radomyski et al., 1994; Thayer and Boyd, 1993).

Several researchers have shown that D-values (the required dose to kill 90% of the micro-organisms present in the product) of 1 kGy and less eliminated pathogenic bacteria and gram negative spoilage microorganisms (Clavero et al., 1994; Lefebvre, et al., 1992; Mattison et al., 1986; and Tarkowski et al., 1984). By using conventional plate counts, Thayer et al. (1993) reported no detectable surviving microflora in any samples of lean ground pork from 2 day postmortem loins that received a 1.91 kGy dose or higher, even after refrigerated storage for up to 5 weeks. Thayer and Boyd (1993) concluded that a dose of 1.5 kGy eliminated *E. coli* 0157:H7 in meat challenged with  $10^{4.8}$  CFU/g. at 0 °C following 20 hours of temperature abuse at 35 °C. Thus, by using low dose irradiation a substantial protection against *E. coli* 0157:H7 and other pathogens can be offered to the consumer.

Various packaging films have been shown to suppress spoilage microorganisms and extend shelf life and prevent recontamination of fresh meats (Lee et al., 1995; Farber, 1991; and Radomyski et al., 1994). Consequently, the combination of irradiation and barrier packaging films could be an effective and valuable technology in providing safer, and more wholesome and palatable meat, while augmenting consumer confidence.

Radiolytic compounds are produced from free radicals that are formed when meat products are irradiated. In 1981, the World Health Organization's (WHO) Expert Committee on the Wholesomeness of Irradiated Food found there was no toxicological hazards from foods irradiated up to a dose of 10 kGy (WHO, 1981). Radiolytic compounds, however, are known to cause off odors and discoloration of fresh meat (Lambert et al., 1992; Lee et al., 1995; and Lefebvre et al., 1994). Irradiation caused radiolytic compounds are of importance because consumers perceive fresh meat quality to be a desirable combination of appearance, color, and aroma when the package is opened (Lambert et al., 1992). The higher the dose the more radiolytic compounds are formed resulting in stronger off-odors and discoloration (Mattison et al., 1986; and Murano et al., 1995). Thus, the use of low dose irradiation has proven to limit off- odors and the discoloration of fresh meats.

Factors such as dose, temperature, anaerobic or aerobic packaging, and the existing microflora content have been shown to affect the quality of meat products (Lee et al., 1995; Monk et al., 1995; and Radomyski et al., 1994). Other factors requiring research before commercial application of irradiation involve the effects of postmortem age of fresh meats prior to irradiation, and the storage time prior to irradiation on the quality characteristics of fresh meats, especially beef patties. While Lakritz and Maerker (1988) reported 1 to 10 kGy was beneficial to reducing proteolysis caused by endogenous enzymes in 24 hour postmortem beef and Lee et al. (1996) found 2 kGy was effective in accelerated postmortem aging of prerigor beef in 4 and 2 days in comparison to conventional wet aging, neither of these research groups were looking at the direct effects of postmortem age and storage time on the aroma and color of fresh beef.

The objective of this study was to determine the effects of postmortem storage time, the time interval between packaging and irradiation, aerobic and anaerobic packaging, and electron beam irradiation on the color and aroma of fresh beef patties.

## **Materials and Methods**

## Sample Preparation and Storage

Raw beef shank meat from a commercial packing plant was obtained at 3 days postmortem for each of 3 replications, coarse ground through a .95 cm plate, and mixed at the Iowa State Meat Lab. For each of the replications the batch of mixed coarse ground beef was split into 3 equal amounts and placed in plastic lugs. Two lugs were then placed in the cooler and maintained at 0  $^{\circ}$ C until postmortem day 6 and 9, respectively. The 3 day postmortem coarse ground beef was fine ground through a .32 cm plate, and pattied (114 g on

average) using a Hollymatic patty machine (model number 54). Half of the 3 day postmortem patties were next packaged anaerobically in Cryovac B620 barrier bags. The other half of the 3 day postmortem patties were packaged aerobically using a Poly(vinyl Chloride) overwrap film. Half of the total 3 day postmortem patties, consisting of half each of the anaerobic and aerobic packaged patties, were placed back into plastic lugs and stored at 0 °C for three more days. The other half of the 3 day postmortem patties were further split in half consisting of 25% control aerobic patties, 25% control anaerobic patties, 25% treated aerobic patties, and 25% treated anaerobic patties. The control patties were placed in a self service display cooler and maintained between 1 °C and 4 °C, under fluorescent light. The treated patties were irradiated (2 kGy) at the Iowa State University Linear Accelerator Facility, and then stored with the controls in the display cooler between 1 °C and 4 °C. The other half of the 3 day postmortem patties were treated in the same manner being split in half 3 days after packaging. The 6 and 9 day postmortem coarse ground meat were treated in the same manner as the 3 day postmortem ground beef on day 6 and 9 postmortem, respectively.

#### Sensory Evaluation

Sensory evaluations of patties were made 4 days after being irradiated and placed in the display cooler. Preliminary studies had indicated there was no difference in the sensory qualities of irradiated ground beef when performed one or four days after irradiation. Consequently, sensory evaluations were performed 4 days after irradiation to represent the time commercially irradiated patties would be in transport to grocers and consumers. Sensory evaluations for non-irradiated control patties were done at the same time as their counterpart treated patties. Patties were evaluated by a trained panel for aroma and color (Cross et al., 1978). Each panelist received one patty from each of the treatment and control groups. Initial aroma by the panel was conducted immediately after removal of the patties from the packages. Subsequently patties were evaluated 30 minutes later for aroma and color. The aroma scores were based on an 8 point scale, 1 being extremely undesirable, and 8 being extremely desirable. The color scale used to evaluate lean color can be seen in Table 2.

#### **Physical and Chemical Analysis**

Two patties per treatment and control group were evaluated for L, a, and b values (where L = lightness, a = redness, b = yellowness) by a Hunterlab Labscan instrument( model LS 5100). Illuminat A/10 was used with a 4.4 cm diameter aperture. Patties were removed from the package, allowed to bloom for 15 minutes, and then three measurements were made on both patties within each group, and then Hunter L, a, and b scores were averaged. Lipid oxidation of two raw ground beef patties per treatment was determined using the 2-thiobarbituric acid (TBARS) method of Tarladigis et al. (1960).

#### **Statistical Analysis**

A split-plot design was used to analyze the data. The data set was arranged into two sets based on package type for the analysis of variance. SAS~GLM was used in determining means, standard errors of the means, and the analysis of variance. Least significant differences (LSD) were calculated to separate means. An alpha level of P < 0.05 was used to determine significance. The experiment was replicated three times.

#### **Results and Discussion**

Ground beef patties packaged aerobically and anaerobically at a postmortem storage time of 3 days were found to have significantly (P < 0.05) more desirable initial and 30 minute aromas than those stored with 6 and 9 day postmortem storage times (Table 1). Batzer et al. (1959) reported sensory qualities never increased, only deteriorated as postmortem age increased prior to irradiation. Aerobic packaged (PVC) patties irradiated on the day of packaging had more desirable aromas (both initial and 30 minute) (P < 0.05) than patties irradiated 3 days after packaging (Table 1). Anaerobic packaged (VAC) non-irradiated controls were found to have more desirable aromas (P < 0.05) than irradiated patties (Table 1). Lambert et al. (1992) reported similar results in which irradiated (0.5 and 1 kGy) fresh pork had lower or less desirable sensory odor scores than controls.

Irradiated patties had less desirable aroma scores than controls and the trend remained consistent over postmortem storage times regardless of PVC and VAC packaging (Figures 1 and 2). Lefebvre et al. (1994), also found that lean ground beef packaged in polyethylene bags had less pleasurable odors when irradiated with 1, 2.5, and 5 kGy than non-irradiated controls. Irradiated patties in PVC and VAC produced moderately undesirable aroma scores over postmortem storage times (Figures 1 and 2). Aroma scores for VAC non-irradiated control patties decreased from moderately desirable on postmortem storage day 3 to slightly desirable on postmortem storage days 6 and 9 (Figure 1). Aroma scores for PVC non-irradiated control patties decreased from postmortem storage days 3 to 9 (Figure 2). This is most likely due to off-odors from microbial

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caused degradation of the meat (Dempster et al., 1985; and Radomyski et al., 1993).

Aroma scores for VAC control patties were higher than irradiated patties that were irradiated day 0 and 3 after packaging. Mattison et al. (1986) noted that panelists could detect irradiation off-odors in vacuum packaged pork loins irradiated with 1 kGy at storage day 7, but not after storage day 14. The control and irradiated aroma scores for ground beef patties remained consistent on day 0 and 3 of irradiation regardless of packaging (Figure 3). Aromas of irradiated patties in PVC also remained consistently "very undesirable," (a score of 2), on both day 0 and 3 of irradiation after packaging. Aromas of control patties in PVC decreased from day 0 of irradiation after packaging in comparison to day 3, from a score of "slightly desirable" to "moderately undesirable" (Figure 4). Lee et al. (1995) observed irradiation may result in more off-odors when fresh beef is packaged with oxygen such as PVC, rather than VAC packaged patties.

Panelist found non-irradiated controls in either PVC or VAC to be lighter (P < 0.05) and to have higher Hunter "a" values (P < 0.05) than irradiated patties (Table 2 and 3). Control patties in VAC also had higher L, "a", and b values (P < 0.05) than irradiated patties. Dempster et al. (1985), also found higher "a" values of beef burgers treated with 1.5 kGy on day 0 in comparison to control samples. Controls had higher and increasing "a" values over postmortem storage time in PVC, where "a" values decreased over postmortem storage times for irradiated patties (Figure 5). In contrast, Lebepe et al. (1990) detected that irradiation significantly increased Hunter "a" values over the non-irradiated samples in vacuum packaged pork loins. Hunter "a" values of PVC patties irradiated the same day as packaging decreased over the

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postmortem storage time, although "a" values of patties irradiated 3 days after packaging increased over postmortem storage times (see Figure 6).

Vacuum packaged non-irradiated controls and irradiated patties had higher initial aroma scores than 30 minute aroma scores over all postmortem storage times and the days of irradiation after packaging (Table 1). Dempster et al (1985) also reported off-odors improved in irradiated (1.03 to 1.54 kGy) vacuum packaged samples when opened and exposed to the air. Conversely, PVC non-irradiated controls and irradiated patties had lower initial aroma scores than 30 minute aroma scores over all postmortem storage times and the days of irradiation after packaging (Table 1).

The largest difference between anaerobic and aerobic packaged patties (both irradiated and controls) was VAC patties consistently had higher color scores as observed by the panel than their counterpart PVC patties over postmortem storage times and the day of irradiation after packaging. Oxygen in the package when irradiation occurs adversely affects sensory quality by increasing discoloration, as opposed to irradiating meat packaged in vacuo (Lambert et al., 1992; and Lee et al., 1995). Also, on the 0 and the 3 day of irradiation after packaging as well as postmortem storage day 3, 6, and 9 VAC patties consistently had higher Hunter "a" values than PVC patties (Table 2 and 3). Luchsinger et al. also found VAC beef patties with a 0, 2, and 3.5 kGy had higher "a" (redness) values than aerobically packaged counterparts. Irradiated PVC patties had higher Hunter L values than irradiated VAC patties over postmortem storage times and the irradiation day after packaging. Although, Lefebvre et al. (1994) accounted that color preference by his panel was for irradiated rather than control samples, numerous researches have found otherwise. Dempster et al. 1985 found beef burgers when exposed to 1.03

and 1.54 kGy had higher surface color scores by the panel than non-irradiated samples at day 0, but scores decreased over time. The principal effect on meat color by irradiation has been reported to be the destruction of the heme pigment (Batzer et al., 1959).

In aerobic packaging, the presence of  $O_2$  initially promotes a bright red color of beef because of the oxygenation of myoglobin to oxymyoglobin. When irradiation is applied to the meat, oxidation of oxymyoglobin to brown metmyoglobin is enhanced (Lambert et al. 1992). A trained panel found no changes in the sensory attributes of 30-36 hour postmortem beef top round treated with 2 kGy in contrast to controls (Rodriguez et al., 1993). Their color and odor attributes were not measured on raw product, but on cooked beef. Still, Tarkowski et al. (1984) reported a taste panel found 38% of beef filets treated with 1 kGy were not acceptable from a sensory standpoint.

Aroma scores (both initial and 30 minute) for non-irradiated VAC controls were higher than non-irradiated PVC controls over postmortem storage times and irradiation days 0 and 3 after packaging. The 30-minute aroma scores for irradiated VAC patties were higher than 30 minute aroma scores for irradiated PVC patties, but initial aroma scores for irradiated PVC and VAC patties were not different. This indicates when packages are first opened, panelist find equal disagreeable odors for all patties irradiated, but in 30 minutes VAC packaged patties become more desirable while PVC patties do not become more desirable in aroma. A possible reason for this difference might be attributed to the structural and physical differences between the two package types. A poly(vinyl chloride) overwrap film would run the risk of forming chlorine containing radicals and other polymer radicals when exposed to radiation. These radicals could then migrate into the foodstuffs and then react with the meat causing undesirable aromas. Buchalla et al. (1993) reported that poly (vinyl chloride) carried a risk of tainting food products when irradiated with low dose levels of radiation, especially in the presence of oxygen.

There were no differences in thiobarbituric acid (TBARS) values (P > 0.05) between irradiated and non-irradiated patties over postmortem storage and different packaging days. The only significant differences were due to replication. This may be attributed to a wider range of storage temperatures in the display cooler in the second replication versus the first and third replications. Aerobically packaged patties TBARS were 4.90 (0 kGy) and 6.39 (2 kGy) respectively. On the other hand TBARS for anaerobically packaged patties were 1.29 (0 kGy) and 1.32 (2 kGy) for controls and irradiated patties respectively. As would be expected the TBARS results for aerobically packaged patties were higher than anaerobically packaged patties. Lebepe et al. (1990) and Mattison et al. (1986) also found no significant differences in TBAR values between irradiated and non-irradiated samples.

## Conclusions

Coarse ground beef at 3, 6, and 9 day postmortem and then fine ground and pattied at each of these postmortem times, irradiated with 2 kGy had slight discoloration and off-odors. The irradiated and control beef patties with the shortest postmortem storage time (day 3) had significantly (P < 0.05) more desirable aroma scores (versus 6 and 9 day). Beef patties irradiated with a 2 kGy dose were darker than controls (P < 0.05). The irradiated and control beef patties in aerobic packaging with the shortest interval between packaging and irradiation, 0 versus 3 days, had more desirable aroma scores (P < 0.05). Control beef patties anaerobically packaged were found to have more desirable initial and 30-minute aroma scores (P < 0.05) than irradiated beef patties. Aroma scores for aerobic packaged patties did not increase 30 minutes after the package was opened whereas aroma scores for anaerobic packaged beef patties increased 30 minutes after the package was opened.

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difuci objedily ili	01,01401	TREATMENTS				
		Anaerobic	Anaerobic	Aerobic	Aerobic	
		Initial	30 Minute	Initial	30 Minute	
<b>D</b>		<u>Aroma</u>	Aroma	<u>Aroma</u>	Aroma	
Postmortem	3	<b>4.78</b> <sup>a</sup>	<b>5.35</b> <sup>a</sup>	<b>3.95</b> <sup>a</sup>	3.75 <sup>a</sup>	
Storage	6	<b>3.94</b> <sup>b</sup>	<b>4.76</b> <sup>b</sup>	<b>3.21</b> <sup>b</sup>	<b>3.20</b> <sup>b</sup>	
Time (days) <sup>g</sup>	9	<b>3.91</b> <sup>b</sup>	<b>4.54</b> <sup>b</sup>	$2.79^{\mathrm{b}}$	<b>2.61</b> <sup>c</sup>	
	SEM	0.23	0.19	0.15	0.18	
Irradiation	0	4.34	4.86	3.65°	<b>3.48</b> <sup>d</sup>	
Day After	3	4.08	4.91	3.03 <sup>d</sup>	2.95°	
Packaging <sup>g</sup>	SEM	0.19	0.16	0.13	0.14	
Dose (kGy)	0	<b>5.28</b> <sup>c</sup>	5.65°	3.71	3.55	
	2	3.14 <sup>d</sup>	<b>4.11<sup>d</sup></b>	3.05	2.94	
	SEM	0.19	0.16	0.13	0.14	

Table 1. Means<sup>f</sup> showing the effects of postmortem storage times, irradiation day after packaging, and irradiation on the aroma of raw beef patties packaged <u>anaerobically in Cryovac B620 bags or aerobically in Poly(vinyl Chloride).</u>

a-e Superscripts indicate significant differences within columns (P<.05).

- f Mean scores were based on an eight point scale, 1 being extremely undesirable and 8 being extremely desirable.
- g Means for postmortem storage time and the irradiation day after packaging are combinations of control and irradiated patties.

	COLOR <sup>c</sup>	HUNTER	HUNTER	HUNTER
	(BLOOM)	L VALUE	<u>a VALUE</u>	<u>bVALUE</u>
3	5.66	39.0	9.9	7.10
6	5.53	39.7	9.7	7.03
9	5.48	39.2	10.1	7.52
SEM	0.10	0.83	0.42	0.18
0	5.53	39.0	9.8	7.30
3	5.58	39.6	10.0	7.15
SEM	0.08	0.68	0.34	0.15
0	<b>6.00</b> <sup>a</sup>	<b>40.3</b> <sup>a</sup>	10.8ª	<b>7.65</b> ª
2	$5.12^{b}$	38.2 <sup>b</sup>	<b>9.0</b> <sup>b</sup>	<b>6.79</b> <sup>b</sup>
SEM	0.08	0.68	0.34	0.15
	6 9 SEM 0 3 SEM 0 2	BLOOM)         3       5.66         6       5.53         9       5.48         SEM       0.10         0       5.53         3       5.58         SEM       0.08         0       6.00 <sup>a</sup> 2       5.12 <sup>b</sup>	BLOOM       L VALUE         3       5.66       39.0         6       5.53       39.7         9       5.48       39.2         SEM       0.10       0.83         0       5.53       39.0         3       5.53       39.0         0       5.53       39.0         3       5.53       39.0         3       5.58       39.0         3       5.58       39.6         SEM       0.08       0.68         SEM       0.08       0.68         2       5.12 <sup>b</sup> 38.2 <sup>b</sup>	(BLOOM)L VALUEa VALUE35.6639.09.965.5339.79.795.4839.210.1SEM0.100.830.4205.5339.09.835.5839.610.0SEM0.080.680.3406.00 <sup>a</sup> 40.3 <sup>a</sup> 10.8 <sup>a</sup> 25.12 <sup>b</sup> 38.2 <sup>b</sup> 9.0 <sup>b</sup>

Table 2. Means showing the effects of postmortem storage times, irradiation day after packaging, and irradiation on the color of raw beef patties packaged anaerobically in Cryovac B620 bags.

a-b Superscripts indicate significant differences within columns (P<.05)

- c The color scale was an eight point scale, 1 for dark brownish-greenish gray, 2 for light brownish-greenish gray, 3 for light gray, 4 for moderately dark red, 5 for slightly dark red, 6 for cherry red, 7 for moderately light cherry red, and 8 for very light cherry red.
- d Postmortem storage time and the irradiation day after packaging are combinations of control and irradiated patties.

			-		
		COLOR	HUNTER	HUNTER	HUNTER
De entre existence	3	<u>(BLOOM)</u>	L VALUE	<u>a VALUE</u>	b VALUE
Postmortem	3	2.50	43.29	6.93	7.86
Storage	6	2.50	41.53	6.93	7.57
Time (days) <sup>d</sup>	9	2.73	40.96	7.22	7.62
	SEM	2.57	0.87	0.36	0.20
Irradiation	0	2.57	42.02	7.22	7.51
Day After	3	2.57	41.84	6.82	7.86
Packaging <sup>d</sup>	SEM	0.12	0.71	0.29	0.16
Dose (kGy)	0	<b>2.94</b> <sup>a</sup>	41.25	<b>7.85</b> ª	7.64
	2	2.26 <sup>b</sup>	42.61	6.20 <sup>b</sup>	7.73
<u> </u>	SEM	0.12	0.71	0.29	0.16

Table 3. Means showing the effects of postmortem storage times, irradiation day after packaging, and irradiation on the color of raw beef patties packaged aerobically in Poly(vinyl Chloride).

a-b Superscripts indicate significant differences within columns (P<.05)

- c The color scale was an eight point scale, 1 for dark brownish-greenish gray, 2 for light brownish-greenish gray, 3 for light gray, 4 for moderately dark red, 5 for slightly dark red, 6 for cherry red, 7 for moderately light cherry red, and 8 for very light cherry red.
- d Postmortem storage time and the irradiation day after packaging are combinations of control and irradiated patties.

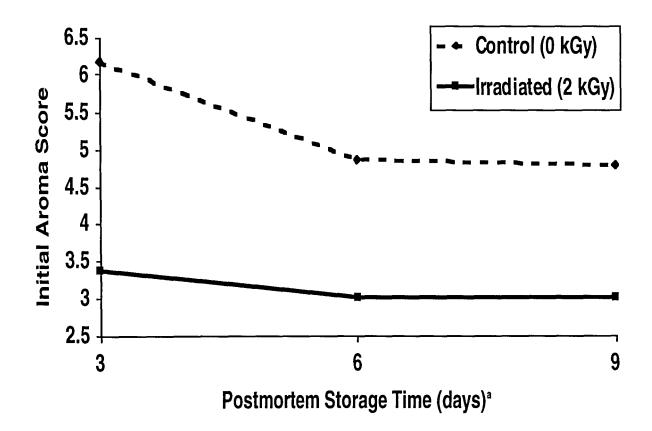
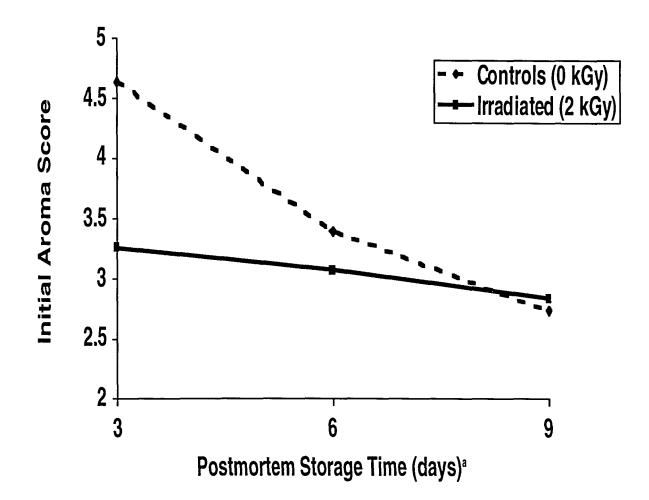


Figure. 1. Initial aroma scores for anaerobic packaged patties at different postmortem storage times and doses.

- a. 30 minute aroma scores followed similar patterns as initial aroma scores.
- b. Postmortem storage time reflects the age of the beef when first processed into patties.



- Figure 2. Initial aroma scores for aerobic packaged patties at different postmortem storage times and doses.
  - a. 30 minute aroma scores followed similar patterns as initial aroma scores.
  - b. Postmortem storage time reflects the age of the beef when first processed into patties.

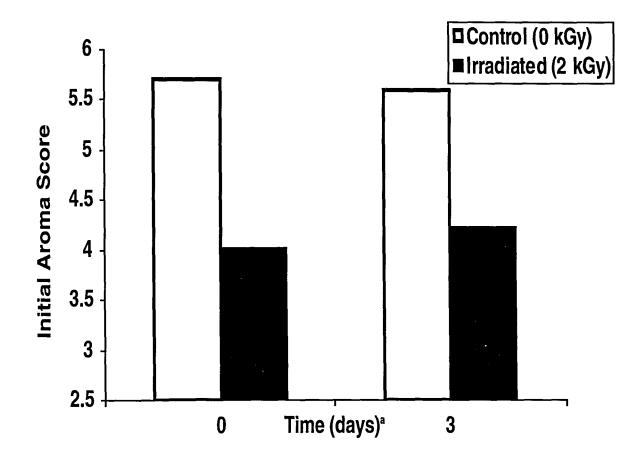
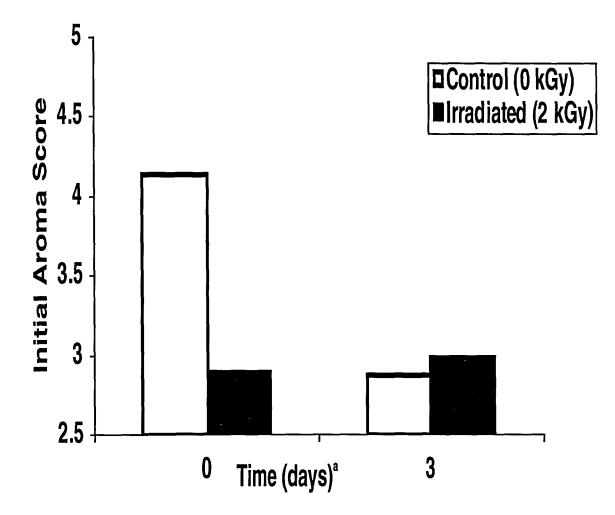


Figure 3. Initial aroma scores for anaerobic packaged patties irradiated 0 or 3 days after packaging.

- a. 30 minute aroma scores followed similar patterns as initial aroma scores.
- b. Patties were processed into patties and packaged on the same day.



- Figure 4. Initial aroma scores for aerobic packaged patties irradiated 0 or 3 days after packaging.
  - a. 30 minute aroma scores followed similar patterns as initial aroma scores.
  - b. Patties were processed into patties and packaged on the same day.

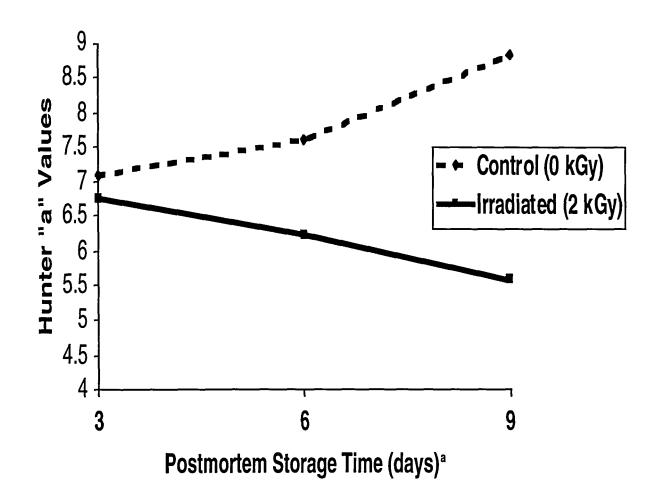


Figure 5. Hunter "a" values for aerobic packaged patties at different postmortem storage times and doses.

a. Postmortem storage time reflects the age of the beef when first processed into patties.

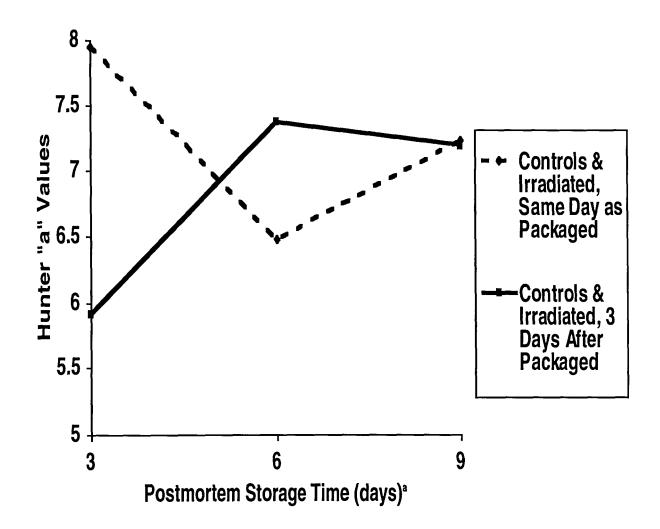


Figure 6. Hunter "a" values for aerobic packaged patties at different postmortem storage times.

- a. Postmortem storage time reflects the age of the beef when first processed into patties.
- b. Lines are a combination of both irradiated and control patties within either a 0 or 3 day irradiation time once packaged.

# THE EFFECTS OF IRRADIATION, STORAGE TIME, AND HIGH AND LOW OXYGEN TRANSMISSION ANAEROBIC PACKAGING ON RAW AND COOKED SENSORY ATTRIBUTES AND COLOR OF GROUND BEEF PATTIES

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# Abstract

The effects of electron beam irradiation, high and low oxygen transmission anaerobic packaging, and storage time on the raw lean color, raw odor, and cooked sensory attributes of ground beef patties were investigated. Beef trim was coarse ground and split into two groups on day one. Group one was fine ground, pattied and packaged immediately; group two was fine ground and packaged six days latter. Patties were held either as controls or irradiated with an average dose of 2 kGy one day following packaging and stored at 0 °C. Sensory evaluations of controls and treated patties were conducted four days after irradiation. Irradiated beef patties had greater (P < 0.05) raw aroma intensities, raw off-odors, and off-flavors, lower (P < 0.05) Hunter CIE L\*, a\* and b\* values, and were darker red (P < 0.05). Seven-day raw beef patties had greater aroma intensities (P < 0.05), higher b\* values and were less juicy (P < 0.05) than raw day one beef patties. Irradiated patties had greater (P < 0.05) off-odors than controls for both day one and day seven beef patties. Hunter b\* values were also lower (P < 0.05) for irradiated patties than controls for both one day and seven day beef patties.

Key Words: Beef patties, irradiation, vacuum packaging, color, sensory attributes.

# Introduction

Fresh meat, ground beef especially, is highly perishable and its shelf-life is limited by the growth of aerobic and psychrotrophic strains of bacteria under refrigerated aerobic storage. While the growth of pathogenic micro-organisms is limited by normal refrigerated storage conditions, they pose potential public health threats if fresh meat is temperature abused. Gram positive lactobacillus which are present in ground beef may also lead to spoilage. The combination of vacuum or modified atmosphere packaging and irradiation has been shown to reduce or eliminate pathogenic and spoilage organisms (Lee et al., 1995; Monk et al., 1995).

Ionizing radiation of meats forms highly reactive and unstable ions or free radicals which react to form stable compounds called radiolytic compounds. While the irradiation of foodstuffs with a maximum dose of 10 kGy has been reported by the World Health Organization (WHO, 1981) to pose no toxicological hazard, several researchers have reported irradiation causes discoloration, off-odors, and off-flavors of fresh meats (Fu et al., 1995a; Lefebvre et al., 1994; and Sudarmadji and Urbain, 1972). At a threshold dose of up to 2.5 kGy off-flavors start to develop in beef (Sudarmadji and Urbain, 1972). Clarke and Richards (1971) also noted beef myoglobin was oxidized by irradiation and that heme was structurally changed by irradiation.

The interaction of ionizing radiation with flexible packaging materials forms radiolytic compounds as a result of chain scission within the carbon chains of the polymers involved. At low doses Rojas De Gante and Pascat (1990) found irradiation of flexible food packaging formed radiolytic organic compounds, typically ketones, aldehydes, and carboxylic acids. These radiolytic compounds could conceivably contribute to negative sensory factors of the food within the package. The combination of vacuum packaging and irradiation can reduce the microbial load of fresh meats and extend the shelflife of raw meats. Prior to commercial application of low dose irradiation treatment of ground beef patties, a proper vacuum package which releases radiolytic compounds while maintaining a long shelf-life of the beef is needed.

Heat stabilizers, antioxidants, lubricants, and plasticizers are used for the processing and the stability of food packaging materials. When flexible packaging is irradiated these additives may degrade and migrate into the packaged food leading to discolorations, off-odors, and off-flavors (Bourgés et al., 1993). Lox et al. (1995) demonstrated that  $\gamma$  irradiation increases the migrational behavior of products within the films at low doses (0 - 10 kGy) and in the case of  $\beta$  irradiation there was a nearly continuous rise of migration as a function of the dose. Also,  $\beta$ -ray irradiation had a less destructive effect on the compounds composing the plastic material which resulted in lower migration values into foodstuffs Because of the potential negative quality and sensory factors which radiolytic compounds may cause there is a need for a vacuum plastic package which will release radiolytic compounds into the air while minimizing global migration into the packaged meat. Furthermore, consumer studies indicate a growing support for irradiated foods and public willingness to buy irradiated products increase when the public is properly educated (Bruhn, 1995; and Resurreccion et al., 1995).

The objective of this study was to determine the effects of storage time of raw beef, high and low oxygen transmission vacuum plastic packaging, and low dose electron beam irradiation on color, and on raw and cooked sensory attributes of ground beef patties.

# **Materials and Methods**

### **Preparation of samples**

Two piece boneless chucks were purchased from a commercial packing plant for each of the 3 replications, and fabricated into 85% beef trim at the Iowa State Meat Laboratory. For each of the replications the batch of beef trim was coarse ground through a 1.27 cm plate, mixed for three to five minutes and placed into plastic lugs. Half of each replication of coarse ground beef was fine ground through a .32 cm plate, and formed into patties (114 g on average) by using a Hollymatic patty machine (model type 54). The ground beef patties were further split into two packaging groups consisting of either a high oxygen permeability beef vacuum bag (Cryovac 37 cc/m<sup>2</sup>/24 hr.) or a low oxygen permeability vacuum bag (Cryovac 10 cc/m<sup>2</sup>/24 hr.). Day-one storage patties consisted of patties from this group. Packages were vacuum sealed by a Multivac Ag 800 vacuum packaging machine. The other half of each replication was stored in covered plastic lugs at 0 °C±1 °C for 6 days. After 6 days of storage the second half of each replication coarse ground beef was fine ground and packaged in the same manner as the first half of each replication. Consequently, this group represented a storage time of day-seven patties.

# Irradiation and storage

The control and treated patties were placed in cardboard boxes and maintained at 0 °C±1 °C. The treated patties were irradiated the day after patties were formed, and packaged at the Iowa State University Linear Accelerator Facility in a single layer one cm thick with an average dose of 2.14  $\pm$  .16 kGy. Samples were irradiated by 10 MeV electron beam at an average dose rate of 32.6 kGy/m/min. The average dose represents an average of the top and bottom surface doses of the samples.

Absorbed doses were determined using alanine pellets as the dosimeter. After irradiation the treated patties were once again stored with the controls at 0 °C $\pm$ 1 °C for another four days. The second half of the 3 replications were divided into control, and treated groups and handled in the same manner as the first half of the replication after the end of the 6 day storage period of the coarse ground beef.

## Sensory evaluation

Sensory evaluations of the treated and control patties of both package types were conducted 4 days after the treated patties were irradiated. The sensory panel was composed of 14 experienced and trained panelists from the faculty, staff, and students in Meat Science at Iowa State University. Panelist had been previously trained (Cross et al., 1978) in experiments in which they were required to detect irradiation off-odors and off-flavors as well as to determine color differences of raw meat and other cooked meat attributes. Raw beef patties consisting of both control and treated patties were evaluated by the trained panel for beef patty aroma intensity, irradiation off-odor, and color in morning sessions. Different patties from the same treatment groups were cooked and evaluated for cooked aroma intensity, irradiation off-odors, juiciness, tenderness, flavor intensity, and irradiation off-flavors by the panel in an afternoon session on the same day.

In the morning raw patty session panelists removed the patties, one at a time from the vacuum package by cutting open the package with a knife and placing the patty on a white paper plate. The patty was then evaluated for beef aroma intensity and irradiation off-odors. After allowing the color to develop for about fifteen minutes the panelists evaluated the patties for lean color. Panelists had been shown two scored fresh reference samples for aroma intensity, color, and off-odors, prior to making their evaluations in each morning session. Samples were identified with a three digit code number.

The beef patty aroma intensity scale was an 8 point scale where 8 was extremely strong and 1 was extremely weak. The off-odor (irradiation) scale was a 5 point scale where 5 was extremely off-odor, and 1 was no off-odor. The color scale was also an 8 point scale consisting of 1 for dark brownish/greenish gray, 2 light brownish/greenish gray, 3 light gray, 4 moderately dark red, 5 slightly dark red, 6 cherry red, 7 moderately light cherry red, and 8 very light cherry red.

For the afternoon cooked session all patties were cooked on a Wolf gas grill from a thawed state (0 °C) for 5 to 7 minutes to 71°C (AMSA, 1995). For each of the four groups 4 patties were cooked. Each patty was cut into eighths, mixed within the group, and two pieces were selected at random for each panelist(AMSA, 1978; and AMSA, 1995). Panelists were served the 2 samples per group on a white plate with a three digit code under red fluorescent lighting. Panelists made evaluations for cooked aroma intensity, irradiation off-odors, juiciness, tenderness, flavor intensity, and irradiation off-flavors. The scales for off-odors and off-flavors (irradiation) were 5 point scales, 5 being extremely off-odor or extremely off-flavor and 1 being no off-odor or no offflavor. Cooked aroma intensity, juiciness, tenderness, and flavor intensity were all 8 point scales with 8 being extremely strong, juicy, tender, and intense, respectively; and 1 was extremely weak, dry, tough, and bland, respectively. Samples were distributed in 4 minute intervals to allow the panel to evaluated each sample thoroughly. Each sample was served hot, shortly after cooking. Panelists cleansed their palate with unsalted crackers and distilled water at room temperature between samples.

#### **Color analysis**

Two patties from each of the four groups from all of the raw sensory analysis were analyzed for CIE L\*, a\*, and b\* values (Illuminat A/10°) by a Hunter Labscan Spectrocolorimiter (4.4 cm diameter aperture, Hunter Associates Laboratory, Inc., model LS 5100). Each patty was removed from the vacuum bag and allowed to bloom over a 15 minute period, and then individually tightly wrapped in a color neutral film. Three measurements were made on both patties within each of the 4 groups and then L\*, a\*, and b\* values were averaged.

# Microbiological analysis

Samples for each half of all the replications were taken prior to fine grinding for aerobic plate counts. Aerobic plate counts were determined using procedures defined by the U. S. FDA (1995).

### Statistical analysis

All experiments were replicated 3 times. Statistical Analysis System~GLM (SAS Institute, Inc., 1994) was used in determining means, standard errors of the means, Fisher least significant differences (LSD) for separation of Least Square Means at P<0.05, and the analysis of variance. Data were analyzed as a 3 by 2 completely randomized block design.

# **Results and Discussion**

Raw aroma intensity was found to be significantly (P < 0.05) higher in irradiated samples than non-irradiated control beef patties (Table 1). The raw aroma intensity was also significantly (P < 0.05) higher for the seven-day beef samples than the one-day samples (Table 1). The increased aroma intensity of the 7-day samples is most likely explained by the higher levels of lactic acid producing micro-organisms being present in those samples (Table 4). Microbial degradation of meat has been reported by Zhao et al. (1996) and Fu et al. (1995b) to lead to increased off-odors of irradiated samples. Consequently, microbial off-odors of meat may have lead to the increased aroma intensity of the raw beef patties in the older samples.

Raw off-odors of the irradiated samples were found to be significantly (P < 0.05) higher than non-irradiated control beef patties (Table 1). Lefebvre et al. (1994) also reported odors of lean ground beef to be judged less pleasant in irradiated than non-irradiated samples. Luchsinger et al. (1995a) also noted that in packaging off-odors were greater in irradiated beef steaks than in controls.

Because panelists found the irradiated beef patties to have higher offodor than controls, the increased aroma intensity of the irradiated samples

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may most likely be due to irradiation off-odors. It has also been shown that the irradiation off-odor is composed of many different compounds (Burks et al., 1959). Irradiation of meat forms many radiolytic products typically hydrocarbons, which may further react and form compounds such as aldehydes, alcohols, ketones, and other carbonyl compounds. Some compounds such as amines and ammonia may have definite effects on the overall odor when they are in combination with similar compounds, although each may be present in a concentration that would be undetectable if the compound were alone.

The irradiation off-odor has been described by Heath et al. (1990) as "burned oil" and "burned feathers" in poultry, as "sour," "rancid," "mature," "bad meat," and "putrid" by Lynch et al. (1991), and even as "wet dog" by Hedin et al. (1960). Lynch et al. (1991) also noted that irradiation off-odor was unlike the sulfurous notes previously associated with protein degradation. Fatty acid composition, dose, and oxygen ( $O_2$ ) within the package during irradiation may contribute to the irradiation off-odor.

The interaction of dose and storage time of the coarse ground beef was also found to be significant (P < 0.05) for irradiation off-odors of raw beef patties. Grant and Patterson, (1991) and Lambert et al. (1992) recorded that offodors of irradiated raw meat samples was a combination of both microbiological spoilage odors and irradiation off-odors. Figure 1 illustrates that one-day and seven-day irradiated samples had greater off-odors than nonirradiated controls. While the irradiation of spoiled meat will lower microbial counts to acceptable levels, the odor of meat may still be unacceptable to consumers. Thus, the irradiation of spoiled ground beef patties produced an odor which consisted of the combination of both spoilage off-odors and irradiation off-odors.

The panel found the color of the raw irradiated beef patties to be significantly (P < 0.05) darker red than the control patties, which were more of a cherry red color (see Table 1). Control patties were also found to have higher (P < 0.05) Hunter CIE L\*, a\*, and b\* values (see Table 2). Fu et al. (1995b) and Luchsinger et al. (1995b) reported raw L\*, a\*, and b\* values of ground beef patties were initially lowered by irradiation. In contrast, Lambert et al. (1992) reported irradiation increased the Hunter L, a, and b values of raw, vacuum packaged pork. Table 2 shows that irradiation causes raw ground beef

When fresh meats are vacuum packaged meat myoglobin changes to deoxymyoglobin which is purplish red and is converted to oxymyoglobin in the presence of oxygen, which is the typical meat color described as "cherry red". When vacuum packaged beef is irradiated, its color changes to brown, representing a change to the trivalent iron of metmyoglobin and oxidation by a hydroxyl radical resulting in the loss of  $O_2$  (Thayer et al., 1993). Thus, the oxidation of deoxymyoglobin to metmyoglobin during irradiation is caused by a small percentage of electrons reacting with the pigment. When re-exposed to air, a portion of the irradiation metmyoglobin is gradually reduced to oxymyoglobin. Consequently, irradiation has the ability to alter the structure of meat pigments, the protein moiety, the state of the heme iron, and heme (Clarke and Richards, 1971).

Day-one beef patties had lower (P < 0.05) b\* values than aged, seven-dayold beef patties (see Table 2). The increased blueness of the older patties may be attributed to slight oxidation of the myoglobin during storage. Also, slight structural changes of the myoglobin or proteins during storage may have raised b\* values.

The interaction of storage time and irradiation for b\* values was also significant (P < 0.05). Control patties consistently had higher b\* values than the irradiated patties over the storage period (Figure 2). This interaction is most likely significant because irradiation increases b\* values while storage time increases the b\* values of the control samples only slightly. Moreover, the interaction of storage time and package type for b\* values was significant (P < 0.05). While the high  $O_2$  transmission packages had b\* values increasing over the storage period, the low  $O_2$  transmission packaging had even higher b\* values (Figure 3). The interaction is most likely more of a storage time effect, than a packaging effect due to the lack of packaging effects found throughout the experiment.

Doses were not significantly different for cooked aroma intensity, cooked off-odors, overall juiciness, overall tenderness, or cooked beef flavor intensity. The refrigerated storage time of the meat also did not significantly affect cooked aroma intensity, cooked off-odors, overall tenderness, cooked beef flavor intensity, or cooked beef off-flavors. Day one ground beef patties, however, were significantly (P < 0.05) juicier, or less dry than the seven-day beef patties (Table 3). As coarse ground beef is stored in open lugs, it looses moisture as purge develops within the package. This loss of fluid from the meat could have attributed to the increased dryness of the seven-day-old patties when compared to the fresh, day one samples.

Irradiated cooked beef patties had greater (P < 0.05) off-flavors than the non-irradiated controls. The volatiles and radiolytic compounds which are formed from irradiation of raw meat and taint transfer products from

packaging materials (Tripp, 1959; and Keay, 1968) result in off-odors as well as lead to off-flavors. Irradiation off-flavors of meats have been listed as being "rancid," "metallic," "sweet," "warm," "stale," and "acidic," (Risvik, 1986). Huber et al. (1953) and Coleby et al. (1961) were some of the first to reveal beef is the most sensitive meat to the development of irradiation off-flavors. Nevertheless, Sudarmadji and Urbain (1972) reported turkey and pork flavors were more sensitive to irradiation than beef. Sudarmadji and Urbain (1972) also reported beef had a threshold dose of 2.5 kGy before irradiation off-flavors developed.

Cooking has generally been noted for eliminating the problems of irradiated raw meats. Hanis et al. (1989) reported cooking diminished and even eliminated the negative sensory effects of irradiation. Untrained panelists were also unable to distinguish between 2 and 5 kGy irradiated ground beef samples and controls for Murano et al. (1995). Nonetheless, Tarkowski et al. (1984) and Lefebvre et al. (1994) found panelists were able to easily and significantly distinguish between irradiated and non-irradiated controls. Consequently, while the trained panel was able to consistently and significantly identify irradiation off-flavors, there were only slight off-flavors present in the irradiated beef patties. It should be also noted that panelists were not asked if the irradiation off-flavor present was desirable or undesirable.

# Conclusions

An objective of our study was to determine if there was a significant difference between the high and low oxygen transmission vacuum packaging in reducing irradiation off-odors and off-flavors. Package type was not a

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significant influence on any of the main factors measured. Thus, a larger difference in oxygen transmission may be necessary in vacuum package material to release radiolytic gases which cause the irradiation odor and flavor of irradiated meats. Dose was not a significant influence for cooked aroma intensity, cooked off-odors, juiciness, tenderness, or flavor intensity. Storage time did not have a significant affect on raw off-odors, panel color, CIE L\* and a\* values, cooked aroma intensity, cooked off-odors, tenderness, flavor intensity, and off-flavors.

Irradiation significantly (P < 0.05) increased raw aroma intensity, raw off-odors, off-flavors, produced a darker red color as determined by panelists, and lowered Hunter CIE L\* a\* and b\* values. Older beef (day-seven patties) had a greater (P < 0.05) raw aroma intensity, lower b\* values, and had dryer cooked patties. Both seven-day and one-day irradiated patties had greater (P < 0.05) off-odor scores and lower b\* values than controls. High oxygen transmission packaging had higher b\* values increase more than low oxygen transmission vacuum packaging for both storage times (P < 0.05).

Accordingly, irradiation played it's greatest role on raw factors of the beef patties. Once cooked there was a small increase in off-flavors found by the trained panel. Thus finding a vacuum package which would allow radiolytic gases to escape out of a high permeable vacuum bag may reduce some of the negative quality factors of irradiation. Also, irradiated patties having greater off-odors for both one-day and seven-day samples than controls partly showed that irradiation off-odors can be compounded with spoilage off-odors. While irradiation will lower microbial counts, once meat has high microbial counts the meat will continue to have negative quality factors after irradiation. Thus, the irradiation of high microbial count meat will produce off-odors as well as other negative sensory qualities, which are combinations of irradiation and microbial off-odors.

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ground beef on aroma inten	<u>sity, off-odors, and</u>	<u>d color of raw beef</u>	patties.
		<b>EVALUATIONS</b>	5
	RAW BEEF	RAW BEEF	RAW BEEF
	AROMA	OFF-ODORS	$\mathrm{COLOR}^{\mathrm{g}}$
	<u>INTENSITY</u> <sup>e</sup>	_(IRRADIATION	J) <sup>f</sup>
DOSE			
Control	<b>4.3</b> <sup>a</sup>	1.5 °	<b>5.5</b> <sup>a</sup>
Irradiated (2 kGy)	<b>4.9</b> <sup>b</sup>	<b>2.5</b> <sup>b</sup>	<b>4.0</b> <sup>b</sup>
PACKAGE TYPE			
High $O_2$ Transmission	4.8	2.1	4.6
$Low O_2$ Transmission	4.5	1.9	4.8
STORAGE TIME			
Day 1	<b>4.4</b> <sup>c</sup>	2.0	4.6
Day 7	<b>4.8</b> <sup>d</sup>	2.0	4.8
SEM's	0.14	0.09	0.14

Table 1. Means of the effects of dose, package type, and storage time of the	,
ground beef on aroma intensity, off-odors, and color of raw beef patties.	

a-d Superscripts indicate significant differences within columns (P < 0.05).

e Scores were based on an eight point scale, 8 being extremely strong and 1 extremely weak.

f Scores were based on a five point scale, 5 being extremely off-odor and 1 no off-odor.

g Scores were based on an eight point scale, 1 for dark brownish-greenish gray, 2 for light brownish-greenish gray, 3 for light gray, 4 for moderately dark red, 5 for slightly dark red, 6 for cherry red, 7 for moderately light cherry red, and 8 for very light cherry red.

		HUNTER LABSCAN CIE SCORES					
	CIE	CIE	CIE				
	<u>L* VALUE</u>	<u>a* VALUE</u>	<u> </u>				
DOSE							
Control	<b>45.3</b> ª	<b>30.8</b> ª	<b>25.6</b> <sup>a</sup>				
Irradiated (2 kGy)	<b>43.4</b> <sup>b</sup>	27.0 <sup>b</sup>	22.4 <sup>b</sup>				
PACKAGE TYPE							
High $O_2$ Transmission	44.3	28.8	24.1				
Low $O_2$ Transmission	44.4 28.9		24.0				
STORAGE TIME							
Day 1	44.1	29.0	23.7°				
Day 7	44.5	28.7	<b>24.4</b> <sup>d</sup>				
SEM's	0.19	0.18	0.11				

Table 2. Means of the effects of dose, package type, and storage time of the
ground beef, on the Hunter Labscan CIE values of raw beef patties.

a-d Superscripts indicate significant differences within columns (P < 0.05).

overa	<u>all-juiciness of cooked bee</u>	f patties.		
			EVALUATIONS	
		AROMA	OFF-ODORS	OVERALL
		<u>INTENSITY</u>	(IRRADIATION)	JUICINESS
DOSI	E			
	Control	5.3	1.4	4.9
	Irradiated (2 kGy)	5.3	1.5	5.0
PAC	KAGE TYPE			
	High O <sub>2</sub> Transmission	5.3	1.4	5.0
	Low O <sub>2</sub> Transmission	5.3	1.4	4.9
STO	RAGE TIME			
	Day 1	5.3	1.4	<b>5.</b> 4ª
	Day 7	5.2	1.4	$4.5^{\mathrm{b}}$
	SEM's	0.12	0.06	0.21
a-b	Superscripts indicate sig	gnificant differe	nces within column	s ( $P < 0.05$ ).
С	Scores were based on an	eight point scal	e, 8 being extremel	y strong and 1
	extremely weak.			
-	a 1 1 (			<u>∧ 1 1 - 1</u>

Table 3. Means of the effects of dose, package type, and storage time of the ground beef, on the cooked beef aroma intensity, cooked beef off-odors, and overall-inciness of cooked beef patties.

d Scores were based on a five point scale, 5 being extremely off-odor and 1 no off-odor.

e Scores were based on an eight point scale, 8 being extremely juicy and 1 extremely dry.

<u>flavors of cooked patties.</u>				
		EVALUATION	S	
	OVERALL	COOKED BEEF		
	<b>TENDERNESS<sup>c</sup></b>	FLAVOR	<b>OFF-FLAVORS</b>	
		INTENSITY	<b>IRRADIATION</b> <sup>e</sup>	
DOSE				
Control	6.0	5.2	<b>1.8</b> <sup>a</sup>	
Irradiated (2 kGy)	5.9	5.4	<b>2.2</b> <sup>b</sup>	
PACKAGE TYPE				
High O <sub>2</sub> Transmission	6.0	5.3	1.9	
$Low O_2$ Transmission	5.9	5.3	2.1	
STORAGE TIME				
Day 1	6.0	5.4	2.0	
Day 7	5.8	5.2	2.0	
SEM's	0.15	0.11	0.08	

Table 4. Means of the effects of dose, package type, and storage time of the ground beef, on the overall-tenderness, cooked flavor intensity, and cooked off-flavors of cooked patties.

a-b Superscripts indicate significant differences within columns (P < 0.05).

c Scores were based on an eight point scale, 8 being extremely tender and 1 extremely tough.

d Scores were based on an eight point scale, 8 being extremely intense and
1 being extremely bland.

e Scores were based on a five point scale, 5 being extremely off-flavor and 1 no off-flavor.

	Storage Time of the Coarse Ground Be		
	Day 1	Day 7	
<b>REPLICATION 1</b>	4.52	5.2	
<b>REPLICATION 2</b>	6.80	>9.00 <sup>b</sup>	
REPLICATION 3	7.76	>9.00 <sup>b</sup>	

Table 5. Colony forming units<sup>a</sup> per replication over the storage times of the <u>coarse ground beef for the meat samples.</u>

a Numbers are Log 10 CFU per gram.

b Numbers are estimations due to high microbial loads.

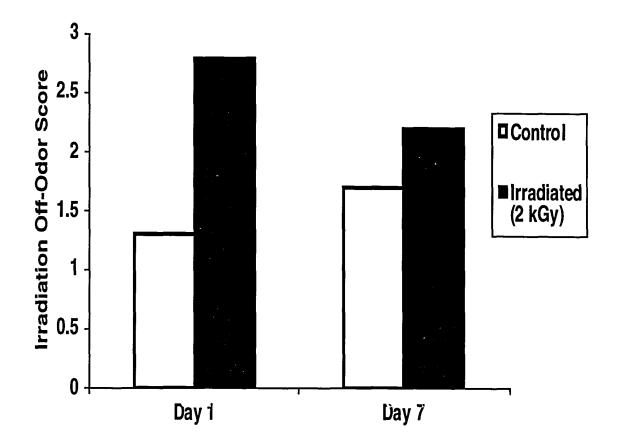


Figure 1. Effects of the storage time of the coarse ground beef and irradiation on the raw off-odor (irradiation) of raw ground beef patties.

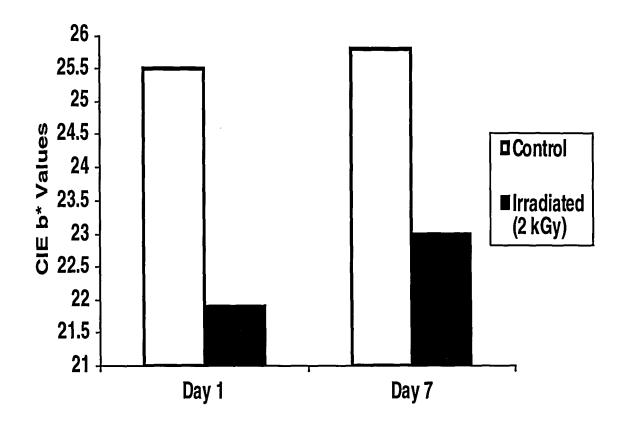


Figure 2. Effects of storage time of the coarse ground beef and irradiation on Hunter Labscan CIE b\* values of raw ground beef patties.

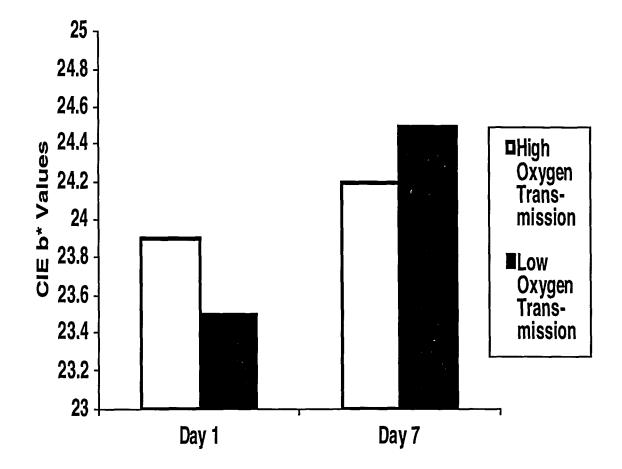


Figure 3. Effects of storage time of the coarse ground beef and package type on Hunter Labscan CIE b\* values of raw ground beef patties.

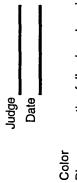


6. Moderately Strong	5. Slightly Strong	
	6. Moderately Strong	6. Moderately Strong 5. Slightly Strong

Please use the following descriptions. Off-Odors(Irradiation) 5. Extremely Off-Odor

- Very Off-Odor
   Moderately Off-Odor
   Slightly Off-Odor
   Slightly Off-Odor
   No Off-Odor

A. Slightly Weak
 Moderately Weak
 Very Weak
 Extremely Weak



Please use the following descriptions. 8. Very light cherry red

- 7. Moderately light cherry red

- 6. Cherry red
  5. Slightly dark red
  4. Moderately dark red
  3. Light gray
  2. Light brownish/greenish gray
  1. Dark brownish/greenish gray

Color Score			
Off-Odor Score			
Aroma Score			
Sample Number			

Comments:

Figure 4. Sensory panel evaluation sheet for raw ground beef patties.

<b>Beef Pattles</b>	
of Ground	Patties
Evaluation	Cooked
Sensory	

Judge Date

Please use the following descriptions for each category.

Off-Flavor(Irradiation)	8. Extremely intense 5. Extremely Off-Flavor	4. Very Off-Flavor		2. Slightly Off-Flavor	1. No Off-Flavor				
Flavor Intensity		7. Very intense		5. Slightly intense	4. Slightly bland	3. Moderately bland	2. Very bland	1. Extremely bland	
<b>Overall Tenderness</b>	8. Extremely tender	7. Very tender	6. Moderately tender	5. Slightly tender	4. Slightly tough	3. Moderately tough	2. Very tough	1. Extremely tough	
<b>Overall Juiciness</b>	8. Extremely juicy	7. Very juicy	6. Moderately juicy	5. Slightly juicy	4. Slightly dry	3. Moderately dry	2. Very dry	1. Extremely dry	
Off-Odors(Irradiation) Overall Juiciness Overall Tenderness	5. Extremely Off-Odor	4. Very Off-Odor 7. Very juicy 7. Very tender	3. Moderately Off-Odor	2. Slightly Off-Odor	1. No Off-Odor				
<b>Cooked Aroma Intensity</b>	8. Extremely Strong	7. Very Strong	6. Moderately Strong	5. Slightly Strong	4. Slightly Weak	<ol><li>Moderately Weak</li></ol>	2. Very Weak	1. Extremely Weak	

Off-Flavor				
Flavor Intensity				
Overall Tenderness Flavor Intensity				
Overall Juiciness				
Off-Odor				
Aroma				
Sample #				

Comments:

Figure 5. Sensory panel evaluation sheet for cooked ground beef patties.

## **GENERAL SUMMARY**

From the two studies contained in this work several conclusions can be made. First, low dose irradiated raw beef patties have greater aroma intensities, off-odors, and have less desirable aromas. Increased aroma intensities are most likely due to irradiation off-odors. Also, irradiation produces a slight off-flavor. The off-flavors present are most likely at the threshold dose, and their effect on overall desirability due to flavor and odor is not fully understood.

Secondly, aging or storing meat prior to irradiation lowers sensory attributes. Raw off-odors increase for irradiated patties over longer storage periods. Thus, lower postmortem ages of meat increase sensory evaluations. Meat should be less than six days postmortem to irradiate, meat of three days postmortem and less is the most desired for irradiation. Also, ground beef needs to be irradiated as soon as possible after packaging. This is especially true if the ground beef is packaged aerobically.

Third, irradiation caused ground beef patties to be scored darker red by trained sensory panels. Irradiation also lowers Hunter a\* values. Irradiation also lowered L\* and b\* values of ground beef patties which were packaged in a anaerobic vacuum packages. Nevertheless, a\* values or the redness of ground beef is the most affected in comparison to L\* and b\* values.

Lastly, anaerobic vacuum packaging improves sensory qualities of irradiated, refrigerated ground beef samples when compared to aerobic packaging. Vacuum packaging especially prolongs the sensory shelflife of raw, refrigerated irradiated ground beef patties. Irradiated patties packaged aerobically with Poly(vinyl Chloride) were not significantly different from non-

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irradiated controls due to microbial spoilage and oxidation to the controls. There was also not a significant effect between the high (37 cc) and low (10 cc) oxygen transmission anaerobic vacuum packaging on the sensory attributes of either the controls or irradiated ground beef patties. Thus, a larger oxygen transmission than 37 cc may be great enough to allow radiolytic compounds causing off-odors and off-flavors to escape from within the anaerobic package while maintaining a vacuum.

Several factors such as dose, temperature, microbial count, postmortem age and package type affect the sensory qualities and color of irradiated ground beef patties. The storage period of beef prior to irradiation was a major focus of the two studies. It was found if ground beef samples had a high microbial count and negative sensory qualities prior to irradiation, the negative sensory qualities continued after irradiation. Thus, only the freshest beef samples should be used in producing irradiated ground beef products. Further research needs to be conducted on the sensory qualities of irradiated ground beef in anaerobic packaging over longer storage periods. Lastly, consumer acceptability of irradiated and noon-irradiated ground beef within anaerobic vacuum plastic packaging needs to be determined.

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