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PRESERVATION OF MEATS BY IONIZING RADIATION - AN UPDATE

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INTRODUCTION:

Preservation of foods, including meats, by ionizing radiation, is an important peaceful application of atomic energy and has an international scope. At present, more than 50 countries have some form of food irradiation research and application (1,2). Most of them are members of the United Nations' International Atomic Energy Agency (IAEA) and the Food and Agriculture Organization (FAO). Except for activities conducted in the United States, most of the food irradiation and related research and development is on low-dose irradiation of agricultural commodities, such as potatoes and onions for sprout inhibition; insect disinfestation of grain, flour, and cereal products; extension of the shelf-life of fruits and vegetables; delaying of ripening of certain fruits, like mangoes and bananas; and extension of shelf life by reducing bacteriological contamination of spices and certain meat, poultry, and seafood products.

Although the Health Authorities in sixteen countries have approved at least one of seventeen irradiated foods, the major effort worldwide is now directed toward obtaining scientific evidence to show that foods irradiated under conditions envisaged for commercial application are safe to eat, i.e., are wholesome and nutritionally adequate. Several excellent reviews and books are available for detailed information on the subject (1, 3, 4, 5, 6, 7, 8, 9). In the United States, the preservation of food by ionizing radiation gained great impetus when President Dwight D. Eisenhower, in December 1953, proposed the ATOMS FOR PEACE program to the United Nations.

The United States' main effort today is in the field of high-dose radiation sterilization (radappertization) of meats, poultry, and selected seafood items as conducted primarily under the U. S. Army's Food Irradiation Program. Since 1971, most of the activities have been concerned with the wholesomeness of radappertized enzyme inactivated beef in preparation for petitioning the U. S. Food and Drug Administration (FDA) and U. S. Department of Agriculture (USDA) for a regulation permitting the unlimited human consumption of irradiated beef. The status of the petitioning and the wholesomeness testing of radappertized beef and other meats will be presented in this paper. The application of ionizing radiation for processing of raw, fresh meats and poultry will also be discussed.

IRRADIATION OF MEAT AND POULTRY

1. Irradiation Sources

The three basic types of ionizing radiation used for processing of foods are gamma rays from cobalt-60 and cesium-137, electrons having a maximum energy of 10 million electron volts (Mev) (5 Mev in the United Kingdom) and X-rays

(5 Mev maximum energy) produced by electrons in an X-ray target. The gamma rays electrons and X-rays cause temporary ionizations and excitations of the molecules in the food. The ionized and excited molecules, together with unstable secondary products, inactivate the microorganisms. The number of the food irradiation pilot plants the world over stood at 27 in 1972 and is continuing to increase as the activities on food irradiation increase (2).

At the U. S. Army Natick Development Center (NDC) there are two pilot scale irradiation sources: (a) 3,000,000 curies cobalt-60 gamma source and (b) 10 MeV versatile electron accelerator (LINAC) (10).

2. Possible Applications of Irradiation in Meat Industry

Table 1 lists the possible applications of ionizing radiations in the meat industry.

a. Terminology.

The terminology used in the food irradiation field was established and approved by an IAEA committee of specialists on terminology of radiation processing (11,12).

(1) Radurization - the term is derived from the words "radiare", to radiate, and "durare", to prolong. The process reduces the numbers of spoilage microorganisms and results in an increase in refrigerated storage time. This term is to replace such terms as "radiation pasteurization" and "irradiation by non-sterilizing doses".

(2) Radicidation - the term is derived from the words "radiare" and "-cida, caedere", to kill, and is used for the radiation process that reduces or eliminates specific organisms of public health significance.

(3) Radappertization - "Radappertization" or "radiation appertization" is the radiation process that commercially sterilizes (destroys microorganisms in the food). The process corresponds closely to commercial heat sterilization of foods. The term is derived from the name of the French confectioner, Appert, who suggested the method for thermal preservation of canned foods.

b. Radurization

An increase of even a few days in the shelf-life of refrigerated fresh meat has great economic value. This is especially important when meat carcasses and products must be transported for some distance, e.g., by sea. Russian investigators reported that irradiation with 0.5 Mrad doses increased the storage time of beef at 3°C up to 6 months, sides of lamb irradiated with 0.4 Mrad up to 8 weeks when stored at 1°C, and raw, vacuum packed (in Saran) pork up to 4 months when irradiated at 0.9 Mrad and stored at 2° to 4.5°C (5).

Various meat dishes made from the red meats and poultry, vacuum packed in flexible plastic films and irradiated with 0.8 Mrad, had good taste properties as verified by professional taste panels and by consumers (5.6). With

Table 1: Possible applications of ionizing radiation in meat industry

Application	Doso Range (Mrad)	Irradiation Temp. (°C)
Radurization for extension of refrigerated storage (0°C to 5°C), e.g., meat, poultry, and fish.	0.05 to 0.5	5° ± 5°
Radicidation - destruction of specific pathogens and parasites, e.g., salmonellae from meat, poultry, and animal feeds; trichinae, tapeworms, and liver flukes in meats.	0.1 to 1.0	5° ± 5°
Sterilization of food ingredients, e.g., spices.	1 to 2	Ambient
Radappertization (sterilization) to allow long-term unrefrigerated storage, e.g., for meats, meat products, poultry, and fish.	2 to 6	-30° ± 10°C
Reduction of nitrite in cured radurized and radappertized meats.	0.5 to 4.0	5° to -40°C

the approval in 1967 by the USSR Public Health Ministry, the vacuum-packed, raw and precooked meat and poultry products irradiated to 0.6 to 0.8 Mrad received a high acceptance in a large-scale consumer test conducted in train dining cars (5).

Three problems may occur during extended storage life of radurized raw meats: discoloration, fat oxidation, and exudation of meat juices. Experiments conducted in the United States on retail units of beef (13) showed that these problems can be controlled by: (a) treatment of the retail meat cuts with 10% solution of sodium tripolyphosphate (TPP) containing 0.25% ascorbic acid to a number of the wrapped retail cuts to form a bulk shipping or wholesale package; (c) irradiation in the chilled state; and (d) transportation of the bulk package under refrigeration to the retail outlet, where it is opened prior to placing the retail cuts in the refrigerated display case to restore the red color (oxymyoglobin) on the surface of the meat cuts. Retail cuts of beef so processed, irradiated with 0.1 to 0.25 Mrad, have been shown to have an increased saleable life at 4.4°C from an average of 4 days to at least 15 days (13,14). The process has definite promise for the meat industry in connection with centralized packing of the retail cuts of beef, pork, and lamb, but additional work is needed to complete the development of commercially suitable processes.

The use of radurizing doses of ionizing radiation to control microbiological spoilage and to increase the saleable shelf-life of fresh, eviscerated chicken has been investigated widely in the United States and other countries. Normal maximum shelf-life of fresh poultry depends mainly on the

Table 2: Radiation resistance (D_{10} in Krad) of some meat spoilage microorganisms irradiated at 5°C to 25°C

Microorganism	Medium	(D_{10}), Krad
C. botulinum type A	Food product	400
C. botulinum type B	Buffered solution	330
Micrococcus radiodurans	Beef	250
C. welchii	Meat	240
C. sporogenes	Buffered solution	210
C. botulinum type E	Bouillon	200
B. stearothermophilus	Buffered solution	100
S. typhimurium	Egg mixture	70
S. typhimurium	Buffered solution	20
Streptococcus faecalis	Bouillon	50
E. coli	Bouillon	20
Pseudomonas species	Buffered solution	4

Source: Ref. 5, Table 19.

storage temperature and is about 6 days at 4°C, 8 days at +1°C, and 10 days at -1°C (15). On the other hand, eviscerated chicken irradiated with 0.4 to 0.6 Mrad doses may be stored for 34 days at +1°C as found in the USSR (5). The doses of 0.1 to 0.3 Mrad are considered necessary for the destruction of the spoilage microorganisms of refrigerated poultry such as Pseudomonas and Achromobacter (5,16,17), while doses of 0.5 to 1.0 Mrad are needed for the destruction of Salmonellae (18,19,20) to improve the hygienic quality (radicidation).

Research conducted at the NDC (18) showed that radurization of fresh eviscerated chicken with a dose of 0.13 to 0.28 Mrad gave carcasses that were free from microbiological spoilage and were of excellent quality for 15 to 21 days at 1.6°C. The only commercially available process which approximates this saleable shelf-life is the low-temperature (-1.0 to -2.0°C) storage and distribution of fresh chicken. Irradiation with 0.5 Mrad gave a product of excellent quality after roasting or broiling even after storage of 35 days at 1.6°C, or 21 days at 4.4°C. The irradiation of the Kosher-processed fresh eviscerated chicken resulted in an additional extension of the shelf-life by 2 to 3 days in comparison with the non-Kosher process (18).

Radurization doses of ionizing radiation of 0.1 to 0.5 Mrad are also very effective in reducing the bacterial population, with the resulting shelf-life extension, in ground raw meats and vacuum-packed luncheon meats and frankfurters stored under normal refrigeration temperatures of 2 to 4.4°C (5,6).

In a study on ground beef, it was shown that the beef obtained from a centrally operated plant contained 2.3 x 10^5 microorganisms per gram, while in the retail store, the bacterial count of non-irradiated ground beef had an average of 5.5 x 10^7 per gram. With 0.204 Mrad irradiation, a 3 log-cycle reduction in the total microflora and a shelf-life extension to approximately 2 weeks at 2°C were attained (21). Much of the residual flora in ground beef was attributed to Morexella-Acinetobacter, which was a common contaminant of all sources of red meats. Psychrotrophic bacteria have been shown to be extremely sensitive to radiation, e.g., Pseudomonas fluorescens gave a D_{10} value of 12 Krad in low-fat ground beef; D_{10} values for other bacteria were Escherichia coli 43, Salmonella typhimurium 64, and Staphylococcus aureus 58 Krad. The bacteria were more sensitive to radiation in a high-fat meat than in a low-fat meat (21).

Table 2 presents the radiation resistance of some of the meat spoilage microorgnisms (5). Radurization is effective in eliminating or greatly reducing the microorganisms listed in the lower half of the table, while higher doses (radappertization) are required for the elimination of C. botulinum species and Micrococcus radiodurans.

c. Radicidation

The low doses of ionizing radiation are very effective also in controlling the growth and reproduction of such parasites as Trichinae (Trichinella spiralis) and tapeworms (Cysticercus bovis and Echinococcus granulosus) with a dose of 15 to 30 Krad (22,23); a dose of 0.4 to 0.5 Mrad (24) may completely eliminate or kill these parasites. In spite of the thorough meat inspection and meat handling in the United States and in other countries, parasites of this kind are still a problem in meats (23,24,25).

The most important application in the use of ionizing radiation is in the use of higher radiciding doses (0.5 to 1.0 Mrad) for the irradiation of fresh poultry, red meats, and animal feeds to eliminate salmonellae. The resistance values (D_{10}) of salmonellae at 4°C vary from 51 to 80 Krad, depending on the species (5,6,20,21). The 0.5 Mrad dose, recommended for the elimination of Salmonellae from fresh poultry (19), would also reduce the numbers of Staphylococcus, Shigella and spoilage organisms by a factor of at least 10^7, and of Clostridium spores by a factor of 10 to 100.

At present limited clearances for low-dose irradiation of fresh, eviscerated chicken for shelf-life extension and/or salmonellae control have been issued in several countries: experimental batches in the Netherlands (300 Krad) and Soviet Union (600 Krad); and fresh and frozen eviscerated poultry, irradiated with a maximum dose of 0.75 Mrad has been approved by the Canadian Government for test marketing.

The dose of 1.0 to 2.0 Mrad can be successfully used to eliminate salmonellae from animal dry feeds and from fish meal (23,26). Israel Health Authorities have approved radicidation of poultry feed using 1.5 Mrad as maximum dose (27). Experiments conducted in England have shown that there is no adverse effect on the nutritive value of animal feeds irradiated at 0.1 to 0.5 Mrad and that the irradiation at 2.0 Mrad was superior to heat treatment with respect to retention of protein quality (26).

d. Irradiation of spices.

Spices generally have high bacterial counts and, since other foods are seasoned with spices, the spices serve as the foci for rapid bacterial growth. Although ethylene oxide has been used as a bactericidal agent, it may leave an undesirable residue (29). Heat is unsatisfactory because it drives off, or reduces the desirable volatiles. Work by Polish (30) and Hungarian (31) investigators has shown that irradiation is highly effective and can be substituted for ethylene oxide. An investigation on feeding rats with a diet containing various levels of spice mixtures irradiated up to 1.5 Mrad is being conducted in Hungary under contract to the International Project in the Field of Food Irradiation with the objective of obtaining the approval by the Health Authorities of irradiated spices.

e. Radappertization.

The scientific and technological feasibility of using ionizing radiations to preserve highly perishable animal protein foods, such as meats, poultry, and some sea foods for long periods of time under non-refrigerated conditions has been proven under the U. S. Army Radiation Preservation of Food Program, presently conducted at the NDC.

Technology is well advanced for radappertized ham, bacon, pork sausage, beef, corned beef, and codfish cakes and, except for the determination of the specific irradiation dose requirements, for shrimp, lamb, turkey and ground beef, pork and chicken with the additives, 0.75% NaCl and 0.3% sodium tripolyphosphate (TPP) (4,23,32,33,34,35,36).

The radappertization process basically involves a pre-irradiation treatment with heat to an internal temperature of 65° to 75°C to inactivate autolytic enzymes; packaging over a partial vacuum in a sealed container impermeable to moisture, air, light, and microorganisms; bring the food package to the temperature at which it will be irradiated; and then exposing the food package to ionizing radiation until the required absorbed dose is obtained.

Table 3 shows the minimum required doses for radappertized foods that have been successfully produced in the laboratory. Bacon irradiated at temperatures at or below 25°C is of excellent quality, but other products develop off-flavors when irradiated at temperatures above freezing. Low temperature (below -5°C) irradiation is used for producing acceptable beef (33,34,35), and other foods can be improved by irradiating them in the frozen state (-30° $\pm$ 10°C). However, as temperature is lowered below 0°C, increasingly higher irradiation doses are required to achieve the same degree of biocidal effect. Also the cost of freezing increases as temperature is lowered below the limit of mechanical refrigeration, i.e., about

Table 3: Minimum required doses (MRD) for radappertization[a]

Food	Irradiation temperature (°C)	MRD (Mrad)
Bacon	5 to 25	2.5
Beef[b]	-30 ± 10	3.7
Ham[c]	5 to 25	3.1
Ham[d]	-30 ± 10	3.5
Pork	5 to 25	4.3
Codfish Cakes	-30 ± 10	3.2
Corned Beef	-30 ± 10	2.4
Pork Sausage	-30 ± 10	2.7

[a]Based on 10^{12} reduction in numbers of spores of C. botulinum (12-D) as determined by the Spearman-Karber method (28);
[b]With the additives: 0.75% NaCl and 0.375% sodium tripolyphosphate;
[c]Regular (high) $NaNO_2/NaNO_3$ (156/700 mg/kg);
[d]Reduced $NaNO_2/NaNO_3$ (25/100 mg/kg);
Source: Mr. Abe Anellis, US Army Natick Development Center

-30°C. Therefore, the most favorable balance of quality, cost, and required irradiation dose appears to be at about -30° ± 10°C.

The minimum radiation doses (MRD) given in Table 3 were obtained in accordance with the 12D concept of microbiological safety. The MRD data indicate the radiation dose in megarads needed to reduce the numbers of viable spores by a factor of 1×10^{12}, based upon the recovery data of the most radiation resistant strains of Clostridium botulinum used in inoculated pack studies with the individual foods in sealed cans as the substrate (28,37). The MRD values depend on the food as well as its temperature during irradiation. The foods containing curing agents (ham, corned beef, bacon) generally have lower MRD's than similar foods without these ingredients.

Mixtures of about 0.75 percent sodium chloride and 0.25 to 0.5 percent food grade phosphates, such as TPP are excellent binding agents both for radappertized hamburger (ground beef), and for formed rolls of beef, chicken, pork, and lamb. Weight loss during enzyme inactivation was reduced from the normal 30-35 percent loss with no additives to 10-15 percent with these additives, thus improving the juiciness of the products. The amount of added TPP of 0.3% is sufficient for the intended purpose (38). All products retained their shape through extended room temperature storage and during kitchen preparation, and meat rolls were readily sliced after reheating.

To protect the radappertized foods from bacterial recontamination after irradiation and during long-term non-refrigerated storage, durable packaging of the food prior to irradiation is required. Two program goals have guided progress in the field: (a) determining reliability of commercially available metal containers for low temperature radappertization of commercially available metal containers for low temperature radappertization of pre-packaged foods, and (b) developing flexible, light-weight containers capable of withstanding rough handling and storage, retaining the protective qualities during storage without any adverse effects on the food contained therein. There is no problem in the irradiation of tinplate containers at doses up to 7.5 megarads at temperatures as low as -90°C, provided the can enamels used are of the epoxyphenolic or phenolic types and the end-sealing compounds are a blend of cured and uncured butyl elastomers, a blend of polychloroprene and butadiene-styrene elastomers, or a blend of polychloroprene and uncured butyl elastomers (39).

The FDA has approved four plastic films as food contactants for foods radappertized by exposure to the gamma rays from Co-60 or Cs-137 to a maximum of 6.0 Mrad: polyethylene, polyethylene terephthalate (Mylar), vinyl chloride-vinyl acetate copolymer and polyiminocaproyl (Nylon 6) (23,40). Other films currently being investigated include the ethylene-butene copolymer, vinylidine chloride-vinyl chloride copolymer, polystyrene, plasticized polyvinyl chloride, polyiminoundecyl (Nylon 11) and a blend of ethylene-butene copolymer and polyisobutylene. These films are used as the food contactant layer in a laminated structure with aluminum foil (middle layer) as a moisture and oxygen barrier, and either Mylar or Nylon 6 as the outside layer to give strength to the laminate in the form of pouches. The laminated flexible package consisting of chemically bonded Mylar and medium density polyethylene as the food contactant layer, aluminum foil (middle layer) and Nylon 6 (outside layer) was found to be very reliable for packaging radappertized foods (40). Over 400,000 such flexible packages were used during 1972 - 1974 for vacuum packaging of more than 40,000 kg. of beef with less than 0.01% failures after vacuum packaging and electron irradiation at -40 to -5°C between 4.7 and 7.1 Mrad. Both metal containers and flexible packages have to be sealed under vacuum to prevent rancidity of the lipids in the foods packaged for radappertization.

Table 4 shows examples for the quality of radappertized meats using the 9-point hedonic scale for preference (41). In case of meat and poultry products, the rating of 5 ("neither like nor dislike") is considered to be threshold of acceptability. A rating of 7 or above indicates a highly acceptable product.

It is of more than passing interest that irradiated ham (with 150/600 mg/kg additions of $NaNO_2/NaNO_3$) was eaten by the astronauts of the Apollo 17 flight to the moon in December 1972. The ham slices, 12 mm in thickness and weighing approximately 105 ± 5 g, were eaten at three meals (including one meal on the moon itself) in sandwiches made with radurized bread (50,000 rads) using radiation insect-disinfested rye flour (50,000 rads). They reported "The juicy, chewy (irradiated) ham and cheese on (irradiated) rye was one of the space culinary delights enjoyed by the Apollo 17 astronauts) (42). Radappertized ham slices were also orbited in Skylab III as an emergency back-up food, along with canned bread made from irradiated (50 Krad) wheat flour. In response to a National Aeronautics and Space Administration request for foods for the Apollo-Soyuz Test Program (ASTP), the NDC provided radappertized

Table 4: Acceptance of radappertized meats

Product	Mrad at -30°C	Recipe	No. Raters	No. Tests	Ave. Rating[a]
Beef	4.7 to 5.3	Onion Gravy	33	2	6.41
"	"	Roastau jus	89	4	6.21
"	"	Brown Gravy	85	4	6.49
Ham[b]	3.7 to 4.4	Grilled	32	2	8.10
"	"	Baked	201	8	7.44
Pork Sausage	2.7 to 3.3	Fried	91	4	7.38
Chicken	4.5 to 5.4	Breaded-Fried	79	2	7.00
Cooked Salami	2.5 to 2.9	Cold	64	2	6.40
		Apollo - Soyuz Test Meats:			
Ham[c]	3.7 to 4.3	Cold	64	2	7.65
Beef Steaks	3.7 to 4.3	Fried	64	2	6.95
Corned Beef	2.5 to 2.9	Cold	64	2	6.95
Turkey Slices	3.7 to 4.3	Cold	64	2	6.35

[a] 9-point-hedonic scale; [b] Regular (high) $NaNO_2/NaNO_3$ (150/600 mg/kg);
[c] Reduced $NaNO_2/NaNO_3$ (25/100 mg/kg)

Table 5: Acceptance of high and low nitrite ham (Consumer Panel: n = 32)

mg/kg added		Irradiation Source	Acceptance Ratings[a]	
$NaNO_2$	$NaNO_3$		Irradiated[b]	Non-Irradiated
150	600	Cobalt-60	6.8 ± 1.5	7.3 ± 1.5
		Electrons	6.5 ± 2.0	
25	100	Cobalt-60	6.4 ± 1.5	7.2 ± 1.4
		Electrons	7.0 ± 1.2	

[a] Paired sets of samples, Cobalt-60 vs Electrons, for low and high nitrite-nitrate hams; [b] 3.7 to 4.4 Mrad at -30°C ± 10°C.

ham (with 25/100 mg/kg additions of $NaNO_2/NaNO_3$), corned beef, turkey slices, and beef steaks for evaluation by prospective pilots, both Russian and American, of the ASTP flight. These products were selected by the astronauts and cosmonauts and were eaten during flight between July 15-24, 1975.

In the United Kingdom, considerable experience has been obtained with animal feeds radappertized at 2.5 Mrad for pathogen-free feeds and at 5.0 Mrad for germ-free feeds (43). Irradiated feeds have been shown to be more nutritious than thermally sterilized feeds. Thermal sterilization (autoclaving) lowers considerably the concentration of individual amino acids, particularly the essential amino acids, lysine, methionine and tryptophan. This was shown by animal feeding studies with rats and mice using thermally sterilized versus irradiation sterilized animal feeding mixtures (44).

In Italy, the irradiation of foot and mouth disease virus with doses of 3 and 4 Mrad was conducted (45). Irradiation in the dry state with 4 Mrad reduced the number of the virus particles by 10^7; the same degree of reduction in the liquid state was achieved with 3 Mrad. Radappertization at cryogenic temperatures in sealed containers in the absence of oxygen offers a means of eliminating this virus in many infected animal products.

The health authorities of the United Kingdom and the Netherlands in 1969 and in Germany in 1972, approved radappertized foods for hospital patients which have either received organ transplants or are being treated for leukemia and have their immune responses suppressed to minimize rejection. Because the suppression of the immune response makes the patients hypersusceptible to bacterial infections, they are kept in a sterile environment and are fed sterile diets. Although heat can be used to sterilize diets, it is not a suitable method for all foods and limits the variety of foods the patients can eat. Radappertization permits a much wider selection of foods and helps stimulate the patient's appetite with improvement in morale and nutritional condition (23,43).

The effect of ionizing radiation on the nutritional value of meat proteins, even when processed with radappertized doses, is less damaging than that of heat (35,44); the effect on vitamins is not markedly different in degree from that of other methods of preservation (23,46). Protection of nutrients is improved by holding the food at low temperature during irradiation and reducing or excluding free oxygen from the foods by vacuum packaging (4, 23, 32, 35, 46).

f. Reduction of nitrite in cured meats.

Recently the researchers of the NDC investigated the possibility of reduction in the additions of nitrite and nitrate in radappertized cured meats such as ham and bacon. Nitrite and nitrate benefit organoleptic qualities such as characteristic flavor and pink color of cured meats. Nitrite, in combination with other curing agents, also inhibits toxin production by *C. botulinum* in thermally processed meats. The use of these curing agents, however, has been under reappraisal by the meat industry and health regulatory agencies because under certain conditions nitrite may react with free amines in food to form nitrosamines, which are carcinogenic (47, 48). In addition, the residual nitrite left in cured meats after processing may react in the gastrointestinal tract with free amines, forming carcinogenic N-nitroso compounds. Model experiments with laboratory animals have shown that high concentrations of nitrite and certain amino compounds induced tumors characteristic of the corresponding N-nitroso compounds (47, 49). Because of the formation of nitroso compounds from nitrite and amines in the stomach, it appears prudent to reduce the intake of nitrite as much as possible and in particular to reduce the amount added to our foods. This was strongly recommended by the toxicology experts of the International Symposium of Nitrite in Meat Products that took place in Zeist, The Netherlands, September 11 to 14, 1973.

The experiments on radappertization conducted at the NDC show that the additions of nitrite to cured, smoked ham and bacon can be reduced from 156 mg/kg the amount commonly used by the meat industry, to 25 mg/kg, without affecting the characteristic color, odor, flavor, and overall acceptance of the product and with the guarantee that no C. botulinum toxin will be formed (50, 51). Table 5 shows the preference data for the low and high nitrite radappertized ham. The data indicate the high quality of products containing only 25 mg/kg sodium nitrite and 100 mg/kg sodium nitrate added to the products during curi instead of the commonly used 150-156 mg/kg nitrite and 500-600 mg/kg nitrate. The important factor in achieving this notable 83% reduction is the fact that radappertization destroys C. botulinum, thus eliminating the need for the larger amount of nitrite required for controlling C. botulinum in nonirradiat cured meats. No nitrosamines (dimethylnitrosamine, methylethylnitrosamine, diethylnitrosamine, nitrosomorpholine, nitrosopyrrolidine, or nitrosopiperidine) were detected in any of the radappertized ham samples, shortly after pr cessing and after 14 months non-refrigerated storage (50, 51). Determination of the nitrosamines in low nitrite-nitrate bacon (raw and pre-fried), with emphasis on nitrosopyrrolidine, continues. The study is being extended to other radappertized cured meats.

WHOLESOMENESS OF IRRADIATED FOODS.

Wholesomeness, in general implies: (a) nutritional adequacy, (b) microbiological safety, (c) zero induced radioactivity, (d) acceptable organolepti and esthetic characteristics, and (e) absence of toxic, carcinogenic, mutagenic, and teratogenic effects. Despite the continuing controversy revolving around whether food irradiation should be regarded as a "food additive" or a "food processing technique" (1, 3, 23) the existing statutes in the United States, i.e., the 1958 Food Additive Amendment to the Federal Food, Drug and Cosmetic Act, have legally defined the intentional exposure of food to ionizi radiation as adulteration of that food by a food additive. Consequently, if irradiated foods are to be permitted for unrestricted human consumption withi the United States, the wholesomeness of irradiated foods must be established according to these statutes. The general principles adopted for testing intetional food additives, therefore, are generally applied to the testing of irradiated food but with certain distinctions (12, 52, 53). Absolute proof o safety of any food additive is not possible in the strictest sense, because t do so would require long-term feeding studies with human test subjects. Consequently, animal models are used as test systems to demonstrate possible kno harmful effects. Data from such experiments are then interpreted from the viewpoint of extrapolating the results to man.

Within the United States, the Department of Army and the Atomic Energy Commission have conducted numerous long-term animal feeding experiments utilizing irradiated foods. Reviews of the accomplishments of these two agencies through 1966 are available (54,55). It is concluded in these reviews that no evidence of untoward biological effects on animals or impairment of nutritional quality have been found and that foods irradiated with gamma rays or 10 MeV electrons up to an absorbed dose of 5.6 Mrads are as wholesome as non-irradiated foods.

Table 6: Diet groups of dogs

Group Designation	Diet[a]
Group I - - - - - - - - - -	100% commercial dog ration
Group II - - - - - - - - - -	35% frozen, enzyme-inactivated beef[b].
Group III - - - - - - - - - -	35% thermally sterilized, enzyme-inactivated beef[b].
Group IV - - - - - - - - - -	35% gamma ray (^{60}cobalt) radappertized, enzyme-inactivated beef[b].
Group V - - - - - - - - - -	35% electron (LINAC) radappertized, enzyme-inactivated beef[b].

a - Dry weight basis; b - Plus 65% commercial dog ration

Table 7: Diet groups of rats and mice

Group Designation	Diet[a]
Group I - - - - - - - - -	100% modified semipurified diet.
Group II - - - - - - - - -	35% frozen, enzyme-inactivated beef[b].
Group III - - - - - - - - -	35% thermally sterilized, enzyme-inactivated beef[b].
Group IV - - - - - - - - -	35% gamma ray (^{60}cobalt) radappertized, enzyme-inactivated beef[b].
Group V - - - - - - - - -	35% electron (LINAC) radappertized, enzyme-inactivated beef[b].
Group VI - - - - - - - - -	100% commercial rodent ration.
Group VII - - - - - - - - -	35% frozen, enzyme-inactivated beef[c].
Group VIII - - - - - - - - -	35% thermally sterilized, enzyme-inactivated beef[c].
Group IX - - - - - - - - -	35% gamma ray (^{60}cobalt) radappertized, enzyme-inactivated beef[c].
Group X - - - - - - - - -	35% electron (LINAC) radappertized, enzyme-inactivated beef[c].

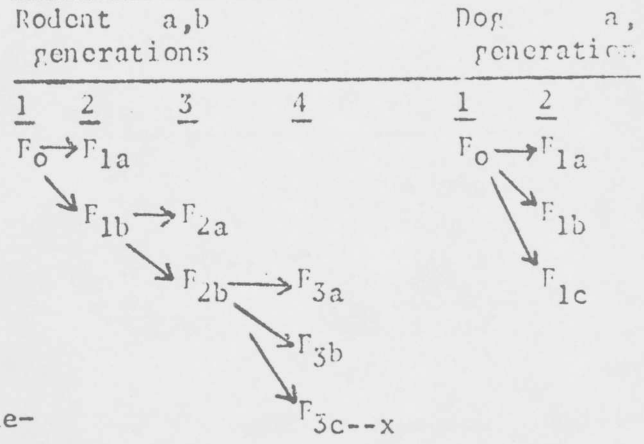
a - Dry weight basis; b - Plus 65% modified basal diet; c - Plus 65% commercial rodent ration.

Despite the enormous amount of work previously accomplished, the general lack of approvals of irradiated foods by regulating agencies indicates that earlier protocols for wholesomeness testing proved inadequate when viewed against more contemporary yardsticks brought about by ever increasing knowledge. As experience was gained with succeeding animal feeding experiments, new parameters for study were then added. As an example, irradiated bacon was approved by the FDA in 1963. This approval was rescinded in 1968 upon re-examination of the same experimental data which was resubmitted in support of a petition for irradiated ham. The reasons for this rescission were based on the fact that the work conducted in the 1950's as reevaluated in light of the "state-of-the art" in the 1960's was found to be insufficient to prove wholesomeness (4,56).

Table 8: Animals per diet group

Species	Males	Females
Rats (Sprague-Dawley)	70	70
Mice (Swiss Albino)	75	75
Dogs (Beagle)	10	20

Table 9: Breeding program and generations



a - F_0 Generation is derived from stock animals fed the respective diets prior to conception and through weaning of the F_0 Generation. b - F_0 Generation maintained on study for 2 years. c - F_0 Generation maintained on study for 3 years.

In spite of the FDA 1968 decision, we believe that irradiated foods are wholesome and have continued to demonstrate this wholesomeness through additional animal feeding studies. Many countries outside the United States have reported a number of short-term and long-term animal feeding studies that have been completed since 1969 (1,3,5,6,43,53,57). None of the investigations indicated any incidence of chronic toxicity or carcinogenicity with the irradiated food. Some of these investigations also included mutagenicity and teratogenicity tests, all with negative results.

In 1971, the United States Department of the Army initiated a broad based study to establish the wholesomeness of radappertized, enzyme-inactivated beef (56,58). The various areas of this study included microbiology, induced radioactivity, radiation chemistry and food technology aspects as well as the multigeneration animal feeding studies which are outlined here.

1. Diet groups and test diets

Table 6 lists the five diet groups in the dog feeding portion of the studies. Because the nutrition of the dog has not been as well defined as for the rodents, and partly because of economic factors, a commercial dog ration was selected as the negative control diet and served as the basal ingredient (65% by dry weight) of the other four diets.

The original intent of the study was to have a similar five-group design for rats and mice in which a semipurified diet would be used in lieu of a commercial ration as the negative control diet and as the basal ingredients of the other four diets. However, apparent nutritional inadequacies of the semipurified diet initially used in these studies became evident during the course of the study and the current protocol defines two different negative control diets for both rats and mice. This change has resulted in ten diet groups for each rodent species (Table 7).

The primary purpose of the negative control groups in these studies is to serve as an indicator of comparability of the husbandry and management practices of the individual replicates. The meaningful comparison in these

Table 10: General Parameters

Growth and Body Weight
Food Consumption
Reproduction Performance
Longevity (Mortality)
Pathology:
Gross and Microscopic
Ophthalmoscopic (Rats and Dogs)
Urinalysis (Dogs)
Semen Examination (Dogs)

studies is intended to be between the frozen, enzyme-inactivated beef and thermally sterilized, enzyme-inactivated beef groups on the one hand, and the two radappertized (^{60}Cobalt and Electron Linear Accelerator), enzyme-inactivated beef groups on the other.

Table 11: Routine Analyses

Hematology and Chemistry	
Total and Differential Cell Counts	
Hemoglobin and Hematocrit	
Serum Protein, Albumin and Globulin	
Prothrombin Time, SGOT, SGPT	
Serum Alkaline Phosphatase (Dogs)	
Serum Creatinine (Dogs)	
BSP Liver Function (Dogs)	
Diets	
Thiamine	Riboflavin
Pyridoxine	Niacin
Ascorbic Acid	Vitamin A
Vitamin E	
Fatty Acids	Amino Acids
Calcium	Phosphorus
Proximate (Protein, Fat, Ash, Moisture)	
Peroxide Number, TBA, pH	

Table 8 shows the number of animals per diet group. The large number of rodents (140 rats, 150 mice) per diet group is required to permit a suffici number of rodents to be available after two years for adequate statistics on longevity and to allow for the periodic sacrifice of animals for histopatholog cal examinations at three-month intervals. The large number of dogs per diet group (thirty) is necessary for adequate reproduction data.

The breeding program is outlined in Table 9, and follows, in general, the recommended procedure for multigeneration studies (12). Because reproduct performance was questioned in previous studies, and in order to study this mor intensely than the usual breeding recommendations, the F_{2b} (third generation) rats will be bred continuously throughout their life span to determine whether under this type of challenge some defect will be exhibited either in shortened reproductive life or through some lesion.

2. Parameters of interest

Data are collected for the general parameters listed in Table 10, whic are, in general, the generally accepted parameters for such studies (12). Any gross pathology is determined on every animal at autopsy, and if lesions are observed, microscopic examinations are conducted. However, tissues are routin examined microscopically from rats that are routinely sacrificed (four of each sex/diet group), from all animals that are moribund or die during the course o the study, and from all F_o generation animals. The routine analyses conducted during the study are shown in Table 11, and supporting studies are shown in Table 12.

3. Current evaluation

The animal feeding studies will be completed in the summer of 1975 for the dogs and in the summer of 1976 for the rodents. Any preliminary evaluatio

Table 12: Supporting Studies

I. Animal Feeding Studies
A. Antimetabolites to:
1. Thiamine
2. Pyridoxine
B. Teratogenicity
C. Mutagenicity
II. Irradiation Effects on:
A. Fats and Lipids
B. Protein and Amino Acids
III. Microbiology
A. "12-D" Determinations
B. Indigenous Microflora

of the present results must take into account that the animal feeding experime are based on biological entities and whc composite status undergoes minor variati from day to day. Until all studies are completed and the data analyzed, no atte can be made to draw definite conclusions regarding the wholesomeness of irradiate beef. This position is commonly taken i all investigations based upon such biolo cal indicators, and is not unique to the studies. However, to date, there have b no indications from these studies that , reflect adversely on the wholesomeness o radappertized beef. Other studies, foll ing similar procedures as used for the animal feeding with beef, will be initi- ated with radappertized pork, chicken, and low nitrite-nitrate ham in 1976 to assure the wholesomeness of these additional three radappertized meats.

CONCLUDING REMARKS

Irradiation of meats, poultry, and other high protein foods has a great po- tential for economic and social benefits for the developed and developing coun- tries of the world. Radurization and radicidation could reduce significantly the incidence of food-borne and animal feed-borne salmonellae, trichinosis fro pork, and beef tapeworms which are a problem in many countries where food is eaten raw or undercooked. It could also eliminate liver fluke and fish infec- tions which are common in Japan and some Scandinavian countries where raw fish is eaten. Radurization of subprimal cuts of meats, packed in suitable flexible films, will be a great aid to the industrial implementation of centralized pac ing and distribution of the meats.

Radappertization of enzyme-inactivated, precooked meats, poultry, and seaf products, hermetically packed in different size metal cans or flexible pouches could increase the line of the shelf-stable, grocery-type items in the superma kets by increasing the variety, as well as the quality of the presently existi thermally processed items. Radappertization, as a "cold" process, would elimi ate the adverse quality changes, such as destruction of texture, "canned meats" off-flavor, loss of natural juices and destruction of nutrients, such as vitami and essential amino acids, which take place during thermal sterilization of her metically sealed foods.

Another advantage of the radappertization process is its flexibility: it c preserve a variety of meats in the range of sizes and shapes ranging from 2 to kg Pullman or round metal cans for institutional use to ½kg can or 100 grams flexible pouch for individual use. Such meats are compatible with the trend fo greater convenience, simplicity in preparation and reduction of labor in the ki chen.

Radappertized meats, poultry, and seafood, packed in large Pullman or round cans can be used for stockpiling for future emergencies or for the utilization of surplus meats from more productive years for consumption in the following years when the meat production is low; thus, market fluctuations in meat distribution could be stabilized.

There is an industrial interest within the U.S. for stable, high quality meat, poultry, and seafood products, as shown by a survey conducted in March 1972 of ten packers, three seafood processing companies, two food retailers ar two trade organizations (35). However, until the wholesomeness problem is resolved, private industry will not invest the funds for food irradiation. Th extensive wholesomeness studies now conducted by the U.S. Army on radappertize beef, and similar studies to be conducted on radappertized pork, chicken and ham in 1976-1979, should provide the basis for commercialization of shelf-stat radappertized meats and poultry. After successful completion of the wholesome ness studies and following clearances from the FDA and USDA, engineering tests and evaluations by the military and marketing tests for the civilians are anti cipated for beef in 1979 and for pork, ham, and chicken in 1981 and 1982.

The wholesomeness studies conducted abroad (the Netherlands, Germany, Canada, USSR, Japan, India, Hungary) and by the International Project, coordin ated by the IAEA, could allow commercialization of selected low-dose irradiate items (potatoes, onions, spices, mushrooms, shrimp, fish, pork sausage, grain and flour, and poultry) prior to the anticipated marketing testing of radappertized meats in the United States.

Other main obstacles toward commercialization are: (a) the legal definiti of food irradiation as a food additive rather than a food processing method; (b) the stigma attached to the word "irradiation," and (c) lack of factual economical data upon which to base the planning of the industrial operations.

In 1972, a panel of experts met in Bombay, India, and recommended that in giving clearances, regulatory agencies consider food irradiation as a food process and not as a food additive, and that FAO and the IAEA convene a panel, with participation of WHO and representatives of the health authorities of individual countries, such as the FDA in the United States, to determine what must be done to implement the recommendation (8).

Extensive studies on the effect of irradiation on the chemical changes of food constituents (fat, proteins, carbohydrates, vitamins) conducted at the NL and other countries should allow interpolations of the wholesomeness data obtained on specific individual foods to other foods of similar basic composition of particular interest for individual food processors.

In regard to the fear of the "irradiation", education of the consumers wil be necessary by government agencies, such as the USDA and Department of Commer with participation of the FDA in case of the United States. Such educational campaigns, with participation of marketing experts, is anticipated in the Unit States after completion of the wholesomeness studies on beef, pork, chicken, a ham. In this respect a very successful consumer education and four marketing tests were conducted in Israel for irradiated potatoes and onions, and in Thailand for irradiated onions.

The economic feasibility of food irradiation was investigated by several investigators, based mostly on research data and theoretical considerations (1, 3, 7, 8, 9, 34, 59). For radappertization of meats, the estimated cost f(the processing is from 3¢ to 20¢ per kg, depending on factors such as the dos(of irradiation, temperature of irradiation, irradiation source, throughput pe: hour, and processing hours per year (34, 59). In the final analysis, the mea‹ industry will have to make a technology assessment of meat irradiation, takin; into consideration not only the cost of the irradiation and the quality of th: food preserved but also other tangible factors, such as savings in refrigerat: energy during storage and distribution, reduced needs for refrigerated space : grocery stores, introduction of new or improved items to the market, problem (salmonellae or botulinum, problems with meat additives now being used and consumers likes and dislikes.

Meat preservation by irradiation on a widespread commercial basis is stil: perhaps a decade in the future, although some specialized applications will come into use sooner. However, with tens of millions of the world's people still suffering from hunger and malnutrition, the use of irradiation to exten(storage time and to preserve foods, including meats, will contribute to makin; available more food which is disease free and better nutritionally.

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