# SENSORY DETERMINATION OF THE AMOUNT OF FLAVOR CHANGE CAUSED BY GAMMA IRRADIATION IN SELECTED ANIMAL PROTEIN FOODS

Thesis for the Degree of M. S.

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By

Slamet Sudarmadji

AN ABSTRACT OF A THESIS

Submitted to
Michigan State University
in partial fulfillment of the requirements
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ABSTRACT

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Irradiation of foods can cause a characteristic flavor change. In animal protein foods such as meat, there has been reported a species variation in this flavor development. The objective of this research was to measure the response of animal protein foods derived from twenty species of animals of different biological classifications.

A qualified expert panel of judges scored these foods on a flavor intensity scale of five. The foods were irradiated with a number of doses of gamma radiation over a range of 0 to 5 Mrad. Statistical analyses of the dose-flavor relationship were made and conclusions drawn as to flavor threshold dose and species variation in sensitivity.

Foods studied were: beef, lamb, pork, chicken, turkey, venison, bear, whale, sea turtle, hippopotamus, elephant, horse, rabbit, opossum, beaver, shrimp, lobster, trout, halibut and frog.

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# INTRODUCTION

The application of ionizing radiation technology to food preservation has been the subject of intensive scientific research. This research appears promising and feasible for treating many foods.

The values of radiation preservation of meat and poultry products are mainly the extension of product life at refrigeration temperatures with the pasteurizing doses, and indefinite preservation without refrigeration at a sterilizing dose. The irradiation process accomplishes these effects mainly by destruction of spoilage microorganisms.

Beef, for example, irradiated to 100 Krad has its storage life increased two to five times (Hannan and Thornley, 1957). For fishery products, 150-450 Krad is the optimum radiation dose range and the refrigerated life of these foods can be expected to be doubled or tripled. The radiation preservation process of fish and shellfish has been reported to be feasible (Holston, et al., 1967).

Regardless of achievements in the technology of food irradiation or of demonstration of the wholesomeness and of government clearances of the products or of

considerations of economic feasibility, the commercialization of the irradiated products will be largely determined by consumer reaction and acceptance of such products.

Consumer acceptance mainly is determined by factors such as significant physical or chemical changes in the products caused by irradiation process. Changes in flavor, odor and color can greatly influence the acceptance.

Undesirable flavor and odor can be developed by the irradiation process in protein foods. Although the flavor and odor are similar in character among the several kinds of meats, intensity of flavor and odor has been reported to be different. The intensity is reported to be greater in beef, for example, compared with chicken and pork (Hannan, et al., 1957).

Continued research on the development of the technology of food irradiation both in the United States and elsewhere may be expected to lead to a number of applications. Of special interest is in the use of low temperatures during irradiation to reduce off-flavor development.

While a great amount of work has been reported on the chemical nature of the irradiated flavor and odor, there are only scattered reports on the flavor response of different animal protein foods to irradiation.

This present work was undertaken to measure by taste panels the flavor changes obtained by irradiating protein foods from twenty species of food animals.

Five trained expert panel-members evaluated the irradiated flavor of protein food samples with a number of doses of gamma radiation over a range of zero to five Mrad.

Twenty selected food animals covering a wide distribution over the animal kingdom were used in this experiment. They were: beef, swine, lamb, chicken, turkey, deer, bear, whale, sea-turtle, hippopotamus, elephant, horse, rabbit, opossum, beaver, shrimp, lobster, trout, halibut and frog.

# LITERATURE REVIEW

For describing the degree of flavor and odor changes produced by the ionizing radiation process in the animal protein foods, no exact methods of chemical or physical measurement have been worked out to date. Despite the inaccuracy of the subjective evaluation method, it has been used widely. Because of the low concentrations of odor and flavor constituents, variabilities of the samples and differences of human responses, it is not unusual for disagreement to occur among the results from various laboratories in odor and flavor evaluation of irradiated foods.

It is not surprising also that there is some disagreement on the description of irradiated odor and flavor. Various terms or combination of those terms have been used to describe the human response to the irradiated odor and flavor, such as burnt, metallic, bitter, cured meat, reminiscent of cress, cheesy, goaty, wet dog, wet grain, acrylic, or unappetizing (Huber, et al., 1953; Mehrlich, 1966; Batzer, et al., 1959).

Regardless of the different terms used to describe the irradiation odor and flavor, Merritt, et al. (1967)

concluded that the irradiation odor in raw meat is a characteristic property, is the same for beef, pork, lamb and the other meats, and does not vary in type but only in intensity.

In trying to clarify the mechanism and the background of irradiated flavor and odor development, some research workers have evaluated the intensity of irradiated flavor and odor and ranked the meats according to their sensitivity to radiation. Hannan and Thornley (1957) reported their finding that beef is more sensitive than chicken, but pork is less sensitive. Another report ranks meats in order of decreasing sensitivity as follows: beef, lamb, veal and pork (Huber, et al., 1953). A statement by Coleby (1959) agrees with Huber's that beef and lamb are more sensitive than pork, chicken, turkey and bacon.

No ranking of irradiated odor and flavor sensitivity of other meats is available.

## The Critical Dose

No off-odor could be detected in beef up to 93 Krad but at 470 Krad or higher it could be detected readily (Anon., 1962). Hannan and Thornley (1957) suggested the critical dose for beef is about 100 Krad, below which dose no distinctive flavor change from the unirradiated control could be detected. They reported the figure for lean pork is 250 Krad. And only a slight detectable flavor change developed in commercial pork sausage, as high as the dose

of two Mrad. For the British bacon the critical dose is similar to that of the lean pork.

Huber, et al. (1953) believed that irradiated beef at 100 Krep has a better flavor than the unirradiated control.

The critical dose appears to be 250 Krad for irradiated chicken, stored anaerobically. The irradiated flavor at that dose is just detectable if the chicken is cooked by steaming (Coleby, 1959 and Thornley, 1957).

Holston, et al. (1967) reported the optimum dose range for irradiation of fish and shellfish is 150-450 Krad. No irradiated flavor could be detected in trout (Salvelinus namaycush) irradiated up to 0.5 Mrad, cooked by baking without seasoning at 350° F for ten minutes (Graikoski, et al., 1967a).

Groninger and coworkers (1956) examined tuna irradiated at two Mrep. This dose produced a desirable pink color but an undesirable burned odor. Graikoski, et al. (1967b) reported that whitefish was acceptable if irradiated up to 0.3 Mrad but not at 0.4 Mrad.

No perceptible odor change in cured ham was reported up to two Mrep. Bacon and corned beef were free from detectable irradiated flavor at 1.5 Mrep after cooking (Groninger, et al., 1956).

The Chemistry of Irradiated Flavor and Odor

Batzer, et al. (1959) described the odor produced in beef by irradiation up to two Mrad as "sulfide-mercaptan"-like odor, up to four Mrad as "wet grain" odor and up to eight Mrad as "slight burnt and wet grain" odor type.

These sensory descriptions give us a vague picture of the chemical changes quantitatively as well as qualitatively in beef with increasing irradiation dose.

Batzer and coworkers (1959) further found out that hydrogen sulfide, methyl mercaptan, acid-soluble carbonyl compounds and pH were increased in irradiated beef; on the other hand glutathione and glycogen disappeared.

The possible precursors of the undesirable odor components formed during gamma irradiation of beef are water soluble and probably contain nitrogen and/or sulfur (Huber, et al., 1953; Drake, et al., 1957).

Huber, et al. listed the possible source of the irradiated odor as:

1. Formation of hydrogen sulfide and other sulfur compounds.
2. Formation of isovaleraldehyde from free leucine.
3. Various soluble protein and other amino acids.
4. Cabbage odor from irradiated methionine solution.
5. Indole from tryphthophane.
6. Geranium odor from phenylalanine solution.

7. Reaction involving lipid and lipid soluble compounds.

The Role of Lipids

It was reported by Huber, et al. (1953) that in beef with higher fat content less destruction of glutathione and less hydrogen sulfide occurred, but there was no significant difference of irradiated flavor between lean and fat beef.

Groninger, et al. (1956) observed the increasing peroxide concentration in beef and pork during irradiation in the presence of oxygen. Rancidity was believed to have a linear correlation with peroxide content and in this way peroxides contribute to the overall irradiation odor and flavor. They made an observation on the correlation between peroxide value and unsaturated fatty acid content. Due to the higher unsaturated fatty acid content of pork than beef, the peroxide value in pork is two and a half times that of beef at 30 Mrep. Mitchell (1957) supported Groninger's statement by his finding that the source of irradiated flavor and odor appears to be the lipid components.

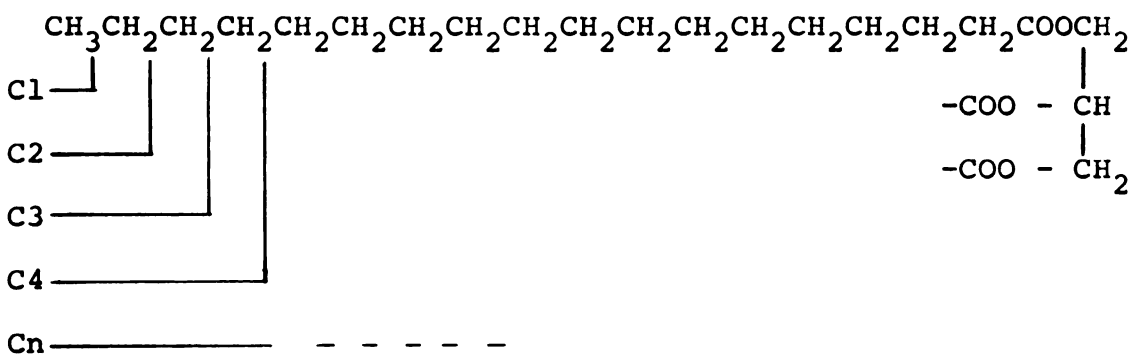
Although peroxides seem to be involved in the formation of irradiated flavor and odor, peroxides themselves are odorless. They play a role in oxidation process by producing secondary products such as aldehydes and ketones.

Through the work by Champagne and Nawar (1967), hydrocarbon series of n-alkanes, 1-alkenes, internally unsaturated alkenes and alkadienes were identified in irradiated beef and pork fat. They determined that a typical off-odor was detectable in beef and pork fat irradiated at two Mrad and was more intense at higher doses. It has been observed that the unsaturated hydrocarbons are much more odorous than the saturated ones. Champagne and Nawar also determined that the quantities of 1-heptene, and 1-octene in beef and pork fat irradiated at six Mrad exceeded their odor concentration threshold in mineral oil. At two Mrad beef and pork fat exhibit some off odor. At this dose, however, the concentrations of all hydrocarbons were found to be below the threshold levels for a detectable odor. These workers suggested the possibility, therefore, of additive or synergistic effects.

Merritt, et al. (1966) believed that the hydrocarbons found in irradiated meats can come only from the lipid. They also noted that the irradiated butter fat had a characteristic fat irradiation odor, unlike the typical rancid odor produced in an oxidized sample. Likewise they also observed that a large amount of hydrocarbons but only a small quantity of carbonyl compounds were produced in irradiated butter fat, whereas a large amount of carbonyl compounds and small amount of hydrocarbons were produced in oxidized butter fat. Their conclusion was that the

mechanism for irradiation production of volatiles is different from the mechanism for oxidation.

Another work by Merritt, et al. (1966) using gas chromatography and mass spectrometry determined the volatile components of irradiated meats. It was concluded that the radiation changes in lipids appear to be the result of radiation-induced direct bond cleavage of the lipid molecules. The scission of glycerol stearate at all points of the chain, will produce alkyl free radicals, and with recombination or hydrogen termination, all the n-alkanes from methane to heptadecane could be formed.




(From Merritt, et al.: Irradiation Damage in Lipids, 1966.)

Figure 1. The scission of glycerol stearate.

Their hypothesis was supported by their finding that the alkanes from methane to pentadecane occur in irradiated meat.

The homologous series of alkenes, which were also detected in moderate quantities, they theorized resulted from secondary collisions, in which a second electron was

extracted from the radicals. Alcohol production was explained as a reaction of alkyl free radicals with water molecules. They concluded that their data support the hypothesis that radiation products are primarily the result of direct bond cleavage, while the carbonyl compounds are produced by an indirect route through the absorption of oxygen by an alkyl free radical followed by decomposition.

The Role of Protein

Many researchers believed that water soluble proteins are a major source of the irradiated flavor and odor (Drake, et al., 1957; Huber, et al., 1953; Batzer, et al., 1955; and Harlan, et al., 1967).

Merritt, et al. (1966) stated that amino acids were responsible for sulfur and aromatic compounds production in irradiated meats. Drake, et al. (1957a) made an extensive study of irradiation of protein in dry and wet state. They found that the irradiated odor was more pronounced from a wet sample, whereas almost odorless products were derived from the dry protein sample. It was also noted that odor increased when water was added to protein which had been irradiated in the dry state.

That individual amino acids react differently to irradiation was proved by Drake, et al. (1957b) by their measurement of amino acid survival in 1 per cent insulin solution irradiated up to 40 Mrep. Cystine appeared to be

the most sensitive to irradiation followed by tyrosine, phenylalanine, proline, histidine and glycine.

Besides cystine, tyrosine, phenylalanine, proline and histidine, Drake, et al. (1957a) believed that methionine, cysteine and tryphtophane are likely the sources of irradiated odor and flavor in protein. Schweigert (1959) also reported an undesirable odor was produced from irradiated leucine.

Drake, et al. (1957a) reported on the specific odor and flavor produced by irradiation of amino acids. Methionine for example, if electron irradiated will produce a strong odor described as similar to cooked garlic and cabbage. A rose-like flowery odor was reported by them from irradiated phenylalanine, polyphenylalanine and n-acetylphenylalanine solutions.

The role of protein in irradiated odor and flavor development is unclear. Again Drake, et al. (1957a) reported the levels of free amino acid, for example, in highly acceptable protein foods such as pork or chicken are not dissimilar from those in a less acceptable food such as beef. They showed that the addition of certain amino acids to the sample prior to irradiation does not significantly lower its acceptability. These workers pointed out also that the odors from irradiated protein are not directly related to amino acid composition, but more closely to the availability of functional groups.

Using gas chromatography and mass spectrometry techniques, Merritt, et al. (1966) found the presence of components from irradiated meats which they concluded arose from the direct bond cleavage of amino acids. They believed that sulfides, disulfides and mercaptans are derived directly from cystine or methionine. Aromatic compounds such as benzene and toluene can be produced from phenylalanine and phenol and p-cresol from tyrosine. Some compounds such as hexyl mercaptan or ethyl-buthyl-disulfide, they explained as probably originating from free radicals derived from lipid and protein portions of meat.

Very similar to the finding by Merritt, Ronsivalli, et al. (1967) using Time of Flight Mass Spectrometer (TOFMS) detected at least 32 compounds in the volatile fraction of clam meat (Mya arenaria). They found that dimethyl sulfide was the dominant component and the source of the typical clam odor. They identified components which are present in irradiated meat such as sulfides, esters, alcohols, amines, carbonyls, carboxylic acids and hydrocarbons, can also be postulated as arising from the radiation effects on the protein and lipid portions of sea foods.

In work somewhat contrary to the postulated important role of fat and protein in producing irradiated odor and flavor, Volkova, et al. (1955) reported that lipid-protein complexes decrease some of the effect of irradiation.

## The Carbohydrates

Very little information is available on the possible role, if any, of carbohydrates in the formation of irradiated flavor and odor. Long, et al (1957) reported the depolymerization of carbohydrates by radiation. Formaldehyde was produced by gamma-irradiation of carbohydrates. They made an observation on irradiation of a d-glucose solution resulted in decomposition products such as acid and a reducing sugar different from d-glucose.

Because of the relatively small amount of carbohydrates present in animal protein foods, they cannot be considered to be a major source of irradiation flavor and odor.

## Synthetic Odor Approach

The role of lipids and protein in the contribution of irradiated flavor and odor has been previously reviewed. Many researchers believed that the irradiation flavor and odor in animal protein foods are caused by the volatile chemical compounds produced by radiation impact on the protein and lipids molecules (Huber, et al., 1953; Drake, et al., 1957a; Wick, et al., 1967; Merritt, et al., 1967).

Using gas chromatography techniques it is possible to trace very small concentration of volatile components in animal protein foods. With this method many odor components can be determined to a degree approaching the human sensitivity.

By using the results of chromatography and by trial and error methods, Wick, et al. (1967) succeeded in preparing a mixture of substances which had an odor resembling the irradiated odor of beef. This odor was produced by mixing just three components (which are also present in non-irradiated and irradiated beef), in the right concentration and proportion, i.e., methional, 1-nonanal, and phenylacetaldehyde at the concentration of 5.0 ppm, 0.5 ppm and 0.25 ppm respectively or proportion of 20:2:1. They believed that the three substances are not completely responsible for irradiated odor and flavor, however, they are the most important contributors.

## Elimination and Prevention of Irradiated Odor and Flavor

There are two kinds of molecular damage caused by irradiation. The first is a direct effect of radiation, in which a molecule is struct directly by an incoming particle or photon and split into fragments. The direct effect is very important in dry food materials or in very concentrated solutions (Frigerio, 1967; Drake, et al., 1957). The second is the indirect effect in which a molecule, such as water, is split into reactive fragments or radicals which then react with other molecules. With indirect action the diffusion of fragments or radicals may be sufficiently slow, that chemical protective agents may

be placed in their path, and sacrificed to protect more critical molecules (Frigerio, 1967).

If the irradiation is done at very low temperature, active fragments and radicals will not be able to move freely from their point of origin and their indirect action thereby reduced.

Harlan, et al. (1967) showed that beef steak irradiated with six Mrad at -196° C (-320° F) had a flavor comparable with the non-irradiated control. This finding apparently supports the belief that the irradiation flavor is caused largely by indirect effect of radiation.

As noted previously, besides the irradiation at low temperature, some chemicals have a preventive effect on the development of the irradiated odor and flavor.

Some vitamins, such as a mixture of alpha tocopherol, ascorbic acid and vitamin A can cause less irradiation flavor in beef (Anonymous, 1962).

The effect of formation of lipid-protein complexes in prevention of irradiation effect in meat has been mentioned earlier.

Mitchell (1957) further suggested that the elimination of oxidative changes related to the radiation flavor, can be achieved by removal of oxygen, exclusion of light, use of low temperature or a combination of those.

Cooking methods may help in reducing the irradiation flavor. Thornley (1957) reported that chicken

irradiated at 250 Krad cooked by steaming could be distinguished from control, but by roasting, chicken irradiated at 375 Krad could not be distinguished from the control. Waters, et al. (1969) conducted taste tests, and concluded that irradiated salmon steaks with doses up to 4.5 Mrad at -30° C (-22° F) were more acceptable when heated in oil rather than in an open air oven. Furthermore they found that tuna and salmon were highly acceptable when breaded and cooked, or when using hickory smoke flavor for masking the irradiated flavor.

Finally, Urbain (1965, 1971) suggested the following possible ways to reduce the irradiated flavor and odor by:

1. Exclusion of oxygen during and after irradiation.
2. Use substances as free radical acceptors.
3. Use low temperatures during irradiation.
4. Absorbents and flavoring for masking (spices, tomato).
5. Selection of foods insensitive to irradiation.
6. Using a small dose.

# MATERIALS AND METHODS

Because the main tool employed in this investigation of irradiated flavor is a subjective method using human sensory perception of flavor, broad representation of samples is needed. Only expert trained tasters were employed.

## Materials

The animal protein foods were selected to cover animal species as varied as possible, but still keeping in mind the possibility of the taster's acceptance as foods.

The animals used as sources of the foods can be placed into the phylla of Chordata (subphyllum Vertebrata) and Arthropoda (class Crustacea).

The systematic classification of the samples is as follows (Gray, 1965; Thomson, 1964; Walker, 1968):

Phyllum: Arthropoda

Class: Crustacea

--Shrimp
--Lobster

Phyllum: Chordata, subphyllum Vertebrata

Class: Mammalia

Order: Carnivora
--Bear

Order: Proboscidea
--Elephant

Class: Mammalia (cont.)

Order: Lagomorpha
--Rabbit

Order: Cetacea
--Whale

Order: Rodentia
--Beaver

Order: Perissodactyla
--Horse

Order: Artiodactyla
--Beef
--Deer
--Lamb
--Swine
--Hippopotamus

Order: Marsupialia
--Opossum

Class: Aves
--Chicken
--Turkey

Class: Reptilia
--Turtle

Class: Amphibia
--Frog

Class: Pisces
--Halibut
--Trout

The samples to be tested were supplied by Michigan State University Food Store in East Lansing or Czimer Foods, Inc. in Chicago. All the samples were kept frozen until shortly before use. The histories of the foods prior to receipt were unknown.

Preparation of Sample

The samples were cut into steaks (except shrimp, lobster, and frog leg) in frozen state, about one inch thick and about 0.5-0.75 lb for each cut. Altogether

twelve sample steaks were needed for each food, ten for the ten dose levels of irradiation plus two non-irradiated controls.

Each cut or reasonable division of shrimp, whole lobster or frog legs was packed and vacuum sealed in International Kenfield's I.K.D. Super Vacuum packaging pouch (All-Vak ≠ 13 F.B.R.). This gas-impermeable pouch consists of Mylar polyester base with a thin coat of polyvinylidine chloride (Saran) applied to the outer surface of a heavier coat of polyethylene as a sealant. The samples then sealed with the Kenfield Vacuum Sealer (Model C14AN). The samples were thawed prior to irradiation.

Irradiation Process

The source of radiation was the $^{60}Co$ irradiator in Food Science Building at Michigan State University, East Lansing. The main source consists of 24 BNL MK-1 radioactive Co strips, doubly encapsulated in stainless steel sheaths, arranged to form a cylinder or a well. The dose rate in the center well was about two Mrad/hr at the height of 15.2 centimeters. The temperature in the irradiation chamber was kept at 40-50° F using the refrigeration facilities. The sample temperature prior to irradiation was about 40-50° F and after removal from the source was about 60° F.

Samples were irradiated in I.K.D. plastic bags. One sample of each food was irradiated at one of the

following doses: 0, 10, 50, 100, 500, 1000, 2000, 3000, 4000, and 5000 Krad. Two non-irradiated samples were used as control. All the samples were irradiated in the center well of the source.

Cooking Method

It is known that the cooking method has an effect on taster's ability to detect the irradiation flavor (Hannan, et al., 1959). Chicken samples irradiated at two Mrad, and cooked by pressure cooking method and by steaming, had an irradiated flavor easily detected by panel. If the samples were grilled lightly, or stewed in water, or deep fat fried, the ability of panel to detect the irradiated flavor was decreased.

In this present study, the samples in I.K.D. plastic bags which were evacuated and sealed were cooked in boiling water for 30 minutes. This method assured a well-done degree of cook, no burning and no leaching by the cook water. The samples were cut into pieces suitable for the panel (about ten grams), and kept warm over warm water during the time of sample presentation to the panel (as long as 30 minutes).

Taste Testing Method

Taste testing methods and procedures were discussed in some publications (Anonymous, 1963; Anonymous, 1964; Kramer, 1966; Hirsh, 1970).

In this present work, the series of twelve samples for each food were divided into two groups, one for each of two panel sessions. The first session employed samples with doses of 0, 50, 500, 2000, and 4000 Krad with one non-irradiated control. The samples for the second session had doses of 10, 100, 1000, 3000, and 5000 Krad, with one non-irradiated control.

The type of test used was the rank order test (Committee on Sensory Evaluation of the Institute of Food Technologists, Anonymous, 1964) or ranking test (Anonymous, 1963). To harmonize the result of the flavor evaluation in each session the taste testers discussed and agreed to the value of the irradiated flavor at a particular dose. For this purpose, the first session determined the value of irradiated flavor of the 500 Krad sample; in the second session the 3000 Krad sample was used to serve this purpose. To help the tasters, one identified non-irradiated control was also served. After the tasters agreed upon the value of the score of irradiation flavor at that particular dose, the other samples were presented in individual booths, disguised under coded numbers, and concealed from color differences as much as possible by using blue fluorescent light. The irradiated flavor intensity was recorded by number scores on score sheets; the score range is one to five, representing:

1 = no irradiation flavor detected

2 = slight irradiation flavor

3 = moderate irradiation flavor

4 = strong irradiation flavor

5 = very strong irradiation flavor

The score sheets employed appear on the next two pages. Score sheet A was used in determining the value of the score of the irradiated flavor of the 500 and 3000 Krad samples. Score sheet B was intended for scoring of other samples.

The Taster and Panel Room

According to the Committee on Sensory Evaluation of the Institute of Food Technologists (Anonymous, 1964) for the rank order type test, three to ten trained panelists are needed, and two to seven samples per test can be served.

In this present work, the panelists were obtained from the staff of the Department of Food Science of Michigan State University in East Lansing. Prior to the actual taste testing, several training sessions to familiarize the panel with the irradiated flavor of beef, pork and chicken were conducted. In addition, triangle tests were given in order to select panel members who had good sensitivity to the irradiated flavor of animal protein foods. Five permanent members were selected, with five other members as alternates.

Score Sheet: A

CALIBRATING PROCEDURE FOR RANKING
IRRADIATION FLAVOR:

Name: _____ Material: _____

This is a ranking test with two samples. One untreated sample, Coded R, is your reference. Please taste the samples, discuss with other panel members, and decide in agreement one of the following categories:

| Irradiation flavor: | Sample number: |
|---|---|
| 1. None | _____ |
| 2. Slight | _____ |
| 3. Moderate | _____ |
| 4. Strong | _____ |
| 5. Very strong | _____ |

Score Sheet: B

FLAVOR INTENSITY SCORE

Judge: _____ Date: _____

Material: _____

---

Indicate the intensity of irradiated flavor, if any, by checking the appropriate box opposite the term which describes the degree of irradiated flavor.

Use as much time as you need.

Please use as reference:

| Sample No.: | Score: | Term: |
|---|---|---|
| _____ | _____ | _____ |

| Score/Term | SAMPLE NUMBER | | | | |
|---|---|---|---|---|---|
| | | | | | |
| 1. None | ____ | ____ | ____ | ____ | ____ |
| 2. Slight | ____ | ____ | ____ | ____ | ____ |
| 3. Moderate | ____ | ____ | ____ | ____ | ____ |
| 4. Strong | ____ | ____ | ____ | ____ | ____ |
| 5. Very strong | ____ | ____ | ____ | ____ | ____ |

Comments:

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Thank you.

The panel room used for testing was equipped with individual boots each with blue fluorescent light. The room was pressurized to avoid contamination with odors from the kitchen and was fully air conditioned.

The sessions were conducted in either morning or afternoon. No more than one session was held on a given day.

## The Statistical Methods

The statistical methods used in this analysis are described in "Principles and Procedures of Statistics" by Steel and Torrie (1960).

The procedure for the Analysis of Variance of Split-plot designs is summarized as follows:

Step one: Find the Correction Term and Total Sum of Squares.

$$\text{Correction Term} = \frac{X_{...}^{2}}{r.a.b} = C$$

X = grand total of observation
r = the number of blocks or judges
a = the number of animal foods or whole units per judge
b = the number of doses or subunits per whole unit

Total Sum of Squares (Total SS) of Subunits =

$$\sum_{ijk} X_{ijk}^{2} - C \quad *)$$

---

*) $X_{ijk}$ denote the score value in the ith block (or judge) from the subunit for the jth level of factor A (animal food samples) and the kth level of factor B (dose of irradiation).

Step two: Complete the Whole-unit Analysis.

$$\text{Whole Unit SS} = \frac{\sum_{ij} X^2_{ij.}}{b} - C$$

$$\text{Judge SS} = \frac{\sum_{i} X^2_{i..}}{a.b} - C$$

$$\text{SS (A = Animal Food Samples)} = \frac{\sum_{j} X^2_{.j.}}{r.b} - C$$

Error (a) SS = Whole Unit SS - Judge SS - SS (A)

Step three: Complete the Subunit Analysis.

$$\text{SS (B = dose)} = \frac{\sum_{k} X^2_{..k}}{r.a} - C$$

$$\text{SS (AB)} = \frac{\sum_{jk} X^2_{.jk}}{r} - C - SS(A) - SS(B)$$

Error (b) = Total SS (Subunit) - Whole Unit SS - SS(B) - SS(AB)

The F-test Calculation

$$\text{Mean Square} = \frac{SS}{\text{degree of freedom}}$$

$$\text{F animal food samples} = \frac{\text{Mean Square of Animal Foods}}{\text{Mean Square of Error (a)}}$$

Compare F calculated with F value from Table S (Rohlf and Sokal, 1969).

If F calculated > F table, there are significant differences among the animal protein food samples.

This F test method can be extended to test the difference among the treatments (radiation dose).

Duncan's New Multiple-range Test

If there are significant differences among the animal protein food samples or among the treatments, we may proceed to the Duncan's multiple range test to see its individual difference.

Procedure:

1. Determine $S_{\bar{x}} = \sqrt{\frac{\text{error mean square}}{r}}$

See Table A.7 (Steel and Torrie, 1960) for Significant Studentized Ranges (SSR).

Find the Least Significant Ranges (LSR) by multiplication of SSR by $S_{\bar{x}}$.

2. Rank the means. Arrange the means of the animal protein food samples in order from the smallest to the highest value.

3. Test the differences: largest minus smallest, largest minus second smallest, . . . , largest minus second largest, then second largest minus smallest, second largest minus second smallest and so on. Each difference is declared significant if it exceeds the corresponding LSR.

## RESULTS AND DISCUSSION

Statistical Analysis.

Analysis of Variance of Split-plot design (Steel and Torrie, 1960) was applied in treating the data.

The data secured with the taste panels are given in Table 1.

Calculation:

$$\text{Correction term} = C = \frac{X...^2}{r.a.b} = \frac{2404^2}{5 \times 20 \times 10} = 5779.2$$

$$\text{Total SS} = \sum_{ijk} X^2_{ijk} - C = 1^2 + 1^2 + \ldots + 5^2 - C$$

$$= 7178 - 5779.2 = 1398.8$$

$$\text{Whole Unit SS} = \frac{\sum_{ij} X^2_{ij.}}{b} - C = \frac{29^2 + 28^2 + \ldots + 23^2}{10}$$

$$- 5779.2 = 5881.2 - 5779.2 = 102$$

$$\text{Judges SS} = \frac{\sum_{i} X^2_{..}}{a.b} - C = \frac{468^2 + 481^2 + \ldots + 469^2}{20 \times 10}$$

$$- 2779.2 = 4.7$$

$$\text{SS A (Animal Food Samples)} = \frac{\sum_{j} X^2_{.j.}}{r.b} - C$$

$$= \frac{141^2 + 119^2 + \ldots + 122^2}{5 \times 10}$$

$$- 5779.2 = 60.7$$

Error (a) SS = Whole Unit SS - Judge SS - Animal

Food Sample SS (A) = 102 - 4.7 - 60.7 = 36.6

$$\text{SS B (Dose)} = \frac{\sum_k X^2_{..k}}{r.a} - C = \frac{130^2 + 122^2 + \ldots + 403^2}{5 \times 20}$$

$$- 5779.2 = 1058$$

$$\text{SS A-B (Animal Food Sample - Dose)} = \frac{\sum_{jk} X^2_{.jk}}{r} - C$$

$$- SS(A) - SS(B) = \frac{5^2 + 5^2 + \ldots + 23^2}{5}$$

$$- 5779.2 - 60.7 - 1058 = 141.9$$

Error (b) SS = Total SS - Whole Unit SS - SS (B)

- SS(AB) = 96.9

F Test Calculation

F calculated for animal food samples

$$= \frac{\text{Mean Square of food samples}}{\text{Mean Square of Error (a)}} = \frac{3.195}{0.482} = 6.63$$

Interpolated value of F (Table S:Rohlf and Sokal, 1969):

$$F\ (0.05, \nu 1, \nu 2) = F\ (0.05, 19, 76) = 1.73$$

F calculated > F table; there is a highly significant difference of irradiated flavor among the animal protein food samples or there is a significant difference in flavor sensitivity of animal protein food samples to gamma irradiation.

$$\text{F calculated for dose} = \frac{\text{Mean Square of Dose (B)}}{\text{Mean Square of Error (b)}}$$

$$= \frac{117.555}{0.134} = 877.27$$

Table S:

$$F(0.05, \nu 1, \nu 2) = F(0.05, 9, 720) = F(0.05, 9, \sim)$$

$$= 1.88$$

There is a very significant effect of irradiation dose on the irradiated flavor of animal protein food samples.

F calculated for interaction of animal protein food samples with dose (AB) $= \frac{\text{Mean Square AB}}{\text{Mean Square of Error (b)}}$

$$= \frac{0.830}{0.134} = 6.19$$

Table S: $F(0.05, \nu 1, \nu 2) = F(0.05, 171, 720)$

$$= F(0.05, \sim, \sim) = 1.00$$

F calculated > F table. There is significant interaction between samples and doses.

Have been previously proved statistically that there is a significant difference of irradiated flavor or sensitivity of animal protein food sample to irradiation among each other. To determine the significance of differences between each sample, Duncan's new multiple-range test was applied.

TABLE 1.--Irradiated flavor intensity score of 20 animal protein foods in ten irradiation doses by five trained expert judges.

| Animal Food | "judge" | Dose Krad | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 10 | 50 | 100 | 500 | 1000 | 2000 | 3000 | 4000 | 5000 | Total | Mean |
| Beef | 1 | 1 | 1 | 1 | 2 | 3 | 4 | 3 | 4 | 5 | 5 | 29 | $\frac{141}{50}$ = 2.82 |
| | 2 | 1 | 1 | 1 | 1 | 3 | 3 | 4 | 4 | 5 | 5 | 28 | |
| | 3 | 1 | 1 | 1 | 1 | 3 | 4 | 3 | 4 | 5 | 5 | 28 | |
| | 4 | 1 | 1 | 2 | 1 | 3 | 3 | 4 | 4 | 5 | 5 | 29 | |
| | 5 | 1 | 1 | 2 | 1 | 3 | 2 | 3 | 4 | 5 | 5 | 27 | |
| | | 5 | 5 | 7 | 6 | 15 | 17 | 16 | 20 | 25 | 25 | 141 | |
| Venison | 1 | 1 | 1 | 2 | 1 | 2 | 3 | 3 | 3 | 3 | 4 | 23 | $\frac{119}{50}$ = 2.38 |
| | 2 | 2 | 2 | 1 | 2 | 2 | 1 | 4 | 3 | 5 | 5 | 27 | |
| | 3 | 3 | 1 | 1 | 1 | 2 | 2 | 2 | 3 | 5 | 3 | 23 | |
| | 4 | 2 | 1 | 1 | 1 | 2 | 2 | 4 | 3 | 5 | 3 | 24 | |
| | 5 | 2 | 1 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 3 | 22 | |
| | | 10 | 6 | 6 | 6 | 10 | 10 | 16 | 15 | 22 | 18 | 119 | |
| Lamb | 1 | 1 | 2 | 2 | 1 | 2 | 2 | 3 | 3 | 4 | 3 | 23 | $\frac{116}{50}$ = 2.32 |
| | 2 | 1 | 1 | 2 | 1 | 2 | 2 | 3 | 3 | 3 | 3 | 21 | |
| | 3 | 2 | 2 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 24 | |
| | 4 | 2 | 1 | 1 | 1 | 2 | 2 | 4 | 3 | 5 | 4 | 26 | |
| | 5 | 1 | 1 | 2 | 1 | 2 | 1 | 3 | 3 | 4 | 4 | 22 | |
| | | 7 | 7 | 8 | 5 | 10 | 9 | 16 | 15 | 20 | 19 | 116 | |

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Pork | 1 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 5 | 5 | 30 | $\frac{145}{50}$ = |
| | 2 | 1 | 1 | 2 | 2 | 3 | 3 | 3 | 4 | 4 | 5 | 28 | |
| | 3 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 5 | 4 | 29 | |
| | 4 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 4 | 4 | 28 | 2.90 |
| | 5 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 5 | 5 | 30 | |
| | | 5 | 5 | 10 | 10 | 15 | 15 | 19 | 20 | 23 | 23 | 145 | |
| Hippo-potamus | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 2 | 2 | 3 | 3 | 19 | $\frac{101}{50}$ = |
| | 2 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 2 | 3 | 4 | 20 | |
| | 3 | 2 | 1 | 1 | 1 | 2 | 2 | 4 | 2 | 4 | 4 | 23 | |
| | 4 | 1 | 1 | 2 | 3 | 2 | 2 | 2 | 2 | 3 | 3 | 21 | 2.02 |
| | 5 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 2 | 3 | 3 | 18 | |
| | | 6 | 5 | 6 | 8 | 10 | 10 | 13 | 10 | 16 | 17 | 101 | |
| Horse | 1 | 1 | 1 | 2 | 1 | 2 | 2 | 3 | 4 | 2 | 3 | 21 | $\frac{111}{50}$ = |
| | 2 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 4 | 2 | 4 | 21 | |
| | 3 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 4 | 3 | 4 | 21 | |
| | 4 | 1 | 1 | 1 | 1 | 2 | 1 | 4 | 4 | 4 | 5 | 24 | 2.22 |
| | 5 | 1 | 1 | 1 | 2 | 2 | 2 | 3 | 4 | 3 | 5 | 24 | |
| | | 5 | 5 | 6 | 6 | 10 | 9 | 15 | 20 | 14 | 21 | 111 | |
| Bear | 1 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 4 | 4 | 4 | 23 | $\frac{120}{50}$ = |
| | 2 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 4 | 4 | 4 | 23 | |
| | 3 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 4 | 4 | 5 | 24 | |
| | 4 | 1 | 1 | 1 | 1 | 2 | 2 | 4 | 4 | 5 | 5 | 26 | 2.40 |
| | 5 | 1 | 1 | 1 | 1 | 2 | 2 | 4 | 4 | 4 | 4 | 24 | |
| | | 5 | 5 | 5 | 5 | 10 | 10 | 17 | 20 | 21 | 22 | 120 | |

Legend: 1 = no irradiation flavor; 2 = slight irradiation flavor; 3 = moderate irradiation flavor; 4 = strong irradiation flavor; 5 = very strong irradiation flavor.

| Animal Food | "Judge" | Dose Rad | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 10 | 50 | 100 | 500 | 1000 | 2000 | 3000 | 4000 | 5000 | Total | Mean |

TABLE 1.--Cont.

| Animal Food | "judge" | Dose Krad | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 10 | 50 | 100 | 500 | 1000 | 2000 | 3000 | 4000 | 5000 | Total | Mean |
| Elephant | 1 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 2 | 3 | 3 | 18 | $\frac{100}{50}$ = |
| | 2 | 2 | 1 | 1 | 1 | 2 | 2 | 3 | 2 | 4 | 4 | 22 | |
| | 3 | 1 | 1 | 1 | 1 | 2 | 3 | 2 | 2 | 3 | 5 | 21 | |
| | 4 | 1 | 1 | 1 | 1 | 2 | 1 | 3 | 2 | 4 | 3 | 19 | 2.00 |
| | 5 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 2 | 4 | 3 | 20 | |
| | | 6 | 5 | 5 | 5 | 10 | 10 | 13 | 10 | 18 | 18 | 100 | |
| Whale | 1 | 2 | 3 | 1 | 2 | 2 | 2 | 3 | 4 | 4 | 4 | 27 | $\frac{121}{50}$ = |
| | 2 | 2 | 2 | 1 | 1 | 2 | 3 | 3 | 4 | 3 | 4 | 25 | |
| | 3 | 2 | 2 | 2 | 2 | 2 | 3 | 4 | 4 | 5 | 4 | 30 | |
| | 4 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 4 | 3 | 3 | 20 | 2.42 |
| | 5 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 4 | 3 | 2 | 19 | |
| | | 8 | 9 | 6 | 7 | 10 | 12 | 14 | 20 | 18 | 17 | 121 | |
| Beaver | 1 | 2 | 1 | 1 | 3 | 2 | 2 | 2 | 3 | 3 | 3 | 22 | $\frac{107}{50}$ = |
| | 2 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 22 | |
| | 3 | 1 | 1 | 1 | 1 | 2 | 3 | 2 | 3 | 3 | 4 | 21 | |
| | 4 | 1 | 1 | 2 | 1 | 2 | 1 | 4 | 3 | 5 | 2 | 22 | 2.14 |
| | 5 | 1 | 1 | 1 | 1 | 2 | 3 | 2 | 3 | 4 | 2 | 20 | |
| | | 6 | 5 | 6 | 7 | 10 | 11 | 13 | 15 | 19 | 15 | 107 | |

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Rabbit | 1 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 3 | 2 | 3 | 18 | $\frac{125}{50}$ = 2.50 |
| | 2 | 2 | 2 | 1 | 1 | 2 | 1 | 5 | 3 | 5 | 3 | 25 | |
| | 3 | 2 | 1 | 4 | 4 | 2 | 3 | 2 | 3 | 5 | 5 | 31 | |
| | 4 | 2 | 1 | 2 | 1 | 2 | 2 | 4 | 3 | 5 | 5 | 27 | |
| | 5 | 2 | 1 | 1 | 1 | 2 | 1 | 3 | 3 | 5 | 5 | 24 | |
| | | 9 | 6 | 9 | 8 | 10 | 9 | 16 | 15 | 22 | 21 | 125 | |
| Opossum | 1 | 1 | 1 | 2 | 3 | 2 | 2 | 3 | 3 | 2 | 3 | 22 | $\frac{102}{50}$ = 2.04 |
| | 2 | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 3 | 3 | 3 | 20 | |
| | 3 | 1 | 1 | 2 | 1 | 2 | 2 | 3 | 3 | 3 | 3 | 21 | |
| | 4 | 2 | 1 | 1 | 1 | 2 | 1 | 2 | 3 | 4 | 2 | 19 | |
| | 5 | 1 | 1 | 1 | 1 | 2 | 1 | 3 | 3 | 3 | 4 | 20 | |
| | | 6 | 5 | 7 | 7 | 10 | 8 | 14 | 15 | 15 | 15 | 102 | |
| Chicken | 1 | 1 | 2 | 1 | 3 | 2 | 1 | 4 | 4 | 4 | 5 | 27 | $\frac{134}{50}$ = 2.68 |
| | 2 | 1 | 3 | 2 | 1 | 2 | 2 | 5 | 4 | 4 | 4 | 28 | |
| | 3 | 2 | 2 | 1 | 1 | 2 | 3 | 4 | 4 | 5 | 5 | 29 | |
| | 4 | 2 | 1 | 1 | 2 | 2 | 1 | 4 | 4 | 4 | 4 | 25 | |
| | 5 | 1 | 2 | 2 | 1 | 2 | 2 | 4 | 4 | 3 | 4 | 25 | |
| | | 7 | 10 | 7 | 8 | 10 | 9 | 21 | 20 | 20 | 22 | 134 | |
| Turkey | 1 | 1 | 1 | 1 | 2 | 3 | 3 | 3 | 4 | 4 | 4 | 26 | $\frac{136}{50}$ = 2.72 |
| | 2 | 1 | 1 | 1 | 2 | 3 | 3 | 2 | 4 | 3 | 4 | 24 | |
| | 3 | 1 | 1 | 2 | 2 | 3 | 4 | 4 | 4 | 5 | 5 | 31 | |
| | 4 | 1 | 2 | 1 | 2 | 3 | 4 | 2 | 4 | 4 | 5 | 28 | |
| | 5 | 1 | 1 | 1 | 1 | 3 | 3 | 3 | 4 | 5 | 5 | 27 | |

TABLE 1.--Cont.

| Animal Food | "judge" | Dose Krad | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 10 | 50 | 100 | 500 | 1000 | 2000 | 3000 | 4000 | 5000 | Total | Mean |
| Turtle | 1 | 2 | 1 | 2 | 2 | 2 | 2 | 3 | 3 | 3 | 3 | 23 | $\frac{113}{50}$ = 2.26 |
| | 2 | 1 | 2 | 2 | 1 | 2 | 1 | 3 | 3 | 3 | 4 | 22 | |
| | 3 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 3 | 3 | 4 | 20 | |
| | 4 | 1 | 2 | 1 | 2 | 2 | 3 | 4 | 3 | 3 | 4 | 25 | |
| | 5 | 2 | 2 | 1 | 1 | 2 | 2 | 4 | 3 | 3 | 3 | 23 | |
| | | 7 | 8 | 7 | 7 | 10 | 10 | 16 | 15 | 15 | 18 | 113 | |
| Frog | 1 | 1 | 2 | 2 | 3 | 2 | 3 | 2 | 3 | 3 | 4 | 25 | $\frac{121}{50}$ = 2.42 |
| | 2 | 1 | 2 | 1 | 2 | 2 | 1 | 2 | 3 | 4 | 4 | 22 | |
| | 3 | 1 | 1 | 3 | 2 | 2 | 2 | 2 | 3 | 4 | 4 | 24 | |
| | 4 | 2 | 1 | 2 | 1 | 2 | 2 | 4 | 3 | 5 | 4 | 26 | |
| | 5 | 1 | 1 | 1 | 1 | 2 | 2 | 4 | 3 | 5 | 2 | 24 | |
| | | 6 | 7 | 9 | 9 | 10 | 10 | 14 | 15 | 21 | 20 | 121 | |
| Halibut | 1 | 1 | 1 | 1 | 2 | 2 | 3 | 2 | 3 | 3 | 4 | 22 | $\frac{119}{50}$ = 2.38 |
| | 2 | 1 | 2 | 2 | 1 | 2 | 2 | 4 | 3 | 5 | 4 | 26 | |
| | 3 | 2 | 1 | 1 | 1 | 2 | 3 | 2 | 3 | 5 | 5 | 25 | |
| | 4 | 1 | 1 | 3 | 1 | 2 | 2 | 2 | 3 | 4 | 4 | 23 | |
| | 5 | 1 | 1 | 1 | 2 | 2 | 1 | 4 | 3 | 3 | 5 | 23 | |
| | | 6 | 6 | 8 | 7 | 10 | 11 | 14 | 15 | 20 | 22 | 119 | |

| | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Trout | 1 | 1 | 1 | 1 | 1 | 2 | 3 | 3 | 4 | 3 | 5 | 24 | $\frac{129}{50}$ = |
| | 2 | 2 | 1 | 1 | 2 | 2 | 3 | 3 | 4 | 4 | 4 | 26 | |
| | 3 | 2 | 1 | 1 | 3 | 2 | 2 | 5 | 4 | 4 | 5 | 29 | |
| | 4 | 2 | 1 | 1 | 1 | 2 | 1 | 3 | 4 | 4 | 5 | 24 | 2.58 |
| | 5 | 2 | 1 | 1 | 1 | 2 | 3 | 3 | 4 | 4 | 5 | 26 | |
| | | 9 | 5 | 5 | 8 | 10 | 10 | 17 | 20 | 19 | 24 | 129 | |
| Shrimp | 1 | 1 | 1 | 1 | 1 | 2 | 3 | 2 | 3 | 4 | 4 | 22 | $\frac{122}{50}$ = |
| | 2 | 2 | 1 | 1 | 2 | 2 | 3 | 3 | 3 | 5 | 4 | 26 | |
| | 3 | 1 | 1 | 2 | 2 | 2 | 4 | 3 | 3 | 4 | 5 | 27 | |
| | 4 | 2 | 1 | 1 | 2 | 2 | 2 | 1 | 3 | 2 | 3 | 19 | 2.44 |
| | 5 | 1 | 2 | 2 | 3 | 2 | 3 | 3 | 3 | 5 | 4 | 28 | |
| | | 7 | 6 | 7 | 10 | 10 | 15 | 12 | 15 | 20 | 20 | 122 | |
| Lobster | 1 | 1 | 2 | 1 | 3 | 2 | 3 | 2 | 3 | 3 | 4 | 24 | $\frac{122}{50}$ = |
| | 2 | 1 | 1 | 1 | 3 | 2 | 4 | 3 | 3 | 2 | 5 | 25 | |
| | 3 | 1 | 1 | 2 | 2 | 2 | 2 | 3 | 3 | 5 | 4 | 25 | |
| | 4 | 1 | 1 | 1 | 3 | 2 | 2 | 3 | 3 | 4 | 5 | 25 | 2.44 |
| | 5 | 1 | 1 | 1 | 2 | 2 | 2 | 2 | 3 | 4 | 5 | 23 | |
| | | 5 | 6 | 6 | 13 | 10 | 13 | 13 | 15 | 18 | 23 | 122 | |
| Treatment total | | 130 | 122 | 136 | 151 | 215 | 227 | 303 | 330 | 387 | 403 | 2404 | |

TABLE 2.--Judges' scores of irradiated flavor for samples evaluated.

| Judge's no. | Total score |
|---|---|
| 1 | 468 |
| 2 | 481 |
| 3 | 506 |
| 4 | 480 |
| 5 | 469 |
| Overall total | 2404 |

TABLE 3.--Degree of Freedom, Mean Square, and F values for Analysis of Variance.

| | DF | SS | Mean Square | F |
|---|---|---|---|---|
| Judge | 4 | 4.7 | 1.175 | |
| Animal Food Samples (A) | 19 | 60.7 | 3.195 | 6.63 |
| Error (a) | 76 | 36.6 | 0.482 | |
| Dose (B) | 9 | 1058 | 117.555 | 877.27 |
| Interaction (AB) | 171 | 141.9 | 0.830 | 6.19 |
| Error (b) | 720 | 96.9 | 0.134 | |

Note: Derivation of Degree of Freedom

Error (a) = (a-1)(r-1)

Interaction AB = (a-1)(b-1)

Error (b) = a(r-1)(b-1)

TABLE 4.--Interpolated value of SSR.

| Value of p | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| SSR | 2.82 | 2.97 | 3.07 | 3.13 | 3.19 | 3.23 | 3.27 | 3.30 | 3.31 | 3.34 | 3.37 | 3.39 | 3.40 | 3.42 | 3.43 | 3.44 | 3.45 | 3.46 | 3.47 |
| LSR | 0.28 | 0.29 | 0.31 | 0.31 | 0.32 | 0.32 | 0.33 | 0.33 | 0.33 | 0.33 | 0.34 | 0.34 | 0.34 | 0.34 | 0.34 | 0.34 | 0.35 | 0.35 | 0.35 |

$$S_{\bar{x}} = \sqrt{\frac{\text{error mean square}}{r}} = \sqrt{\frac{0.482}{50}} = \sqrt{0.00964} = 0.0981 = 0.10$$

$$LSR = SSR \times S_{\bar{x}}$$

Source: Steel and Torrie: Principles and Procedures of Statistics 1960, Table A7.

Duncan's new multiple-range test applied to determine irradiated flavor differences among the animal protein food samples.

TABLE 5.--Ranking of animal protein food samples according to their means of irradiated flavor for overall dose evaluation based on flavor intensity scale of one to five (none to very strong).

| Ranking no. | 1 | 2 | 3 | 4 |
|---|---|---|---|---|
| Animal Food | Elephant | Hippopotamus | Opossum | Beaver |
| Irrad. Flavor Mean | 2.00 | 2.02 | 2.04 | 2.14 |

| 5 | 6 | 7 | 8 | 9 | 10 | 11 |
|---|---|---|---|---|---|---|
| Horse | Turtle | Lamb | Halibut | Venison | Bear | Frog |
| 2.22 | 2.26 | 2.32 | 2.38 | 2.40 | 2.40 | 2.42 |

| 12 | 13 | 14 | 15 | 16 | 17 | 18 |
|---|---|---|---|---|---|---|
| Whale | Shrimp | Lobster | Rabbit | Trout | Chicken | Turkey |
| 2.42 | 2.44 | 2.44 | 2.50 | 2.58 | 2.68 | 2.72 |

| 19 | 20 |
|---|---|
| Beef | Pork |
| 2.82 | 2.90 |

Determination of significant or not significant difference between the animal food samples: based on the difference between their respective means and compared with the SSR (Significant Studentized Ranges, Table 4).

Pork - Elephant = 2.90 - 2.00 = 0.90 > 0.35 (table): Significant

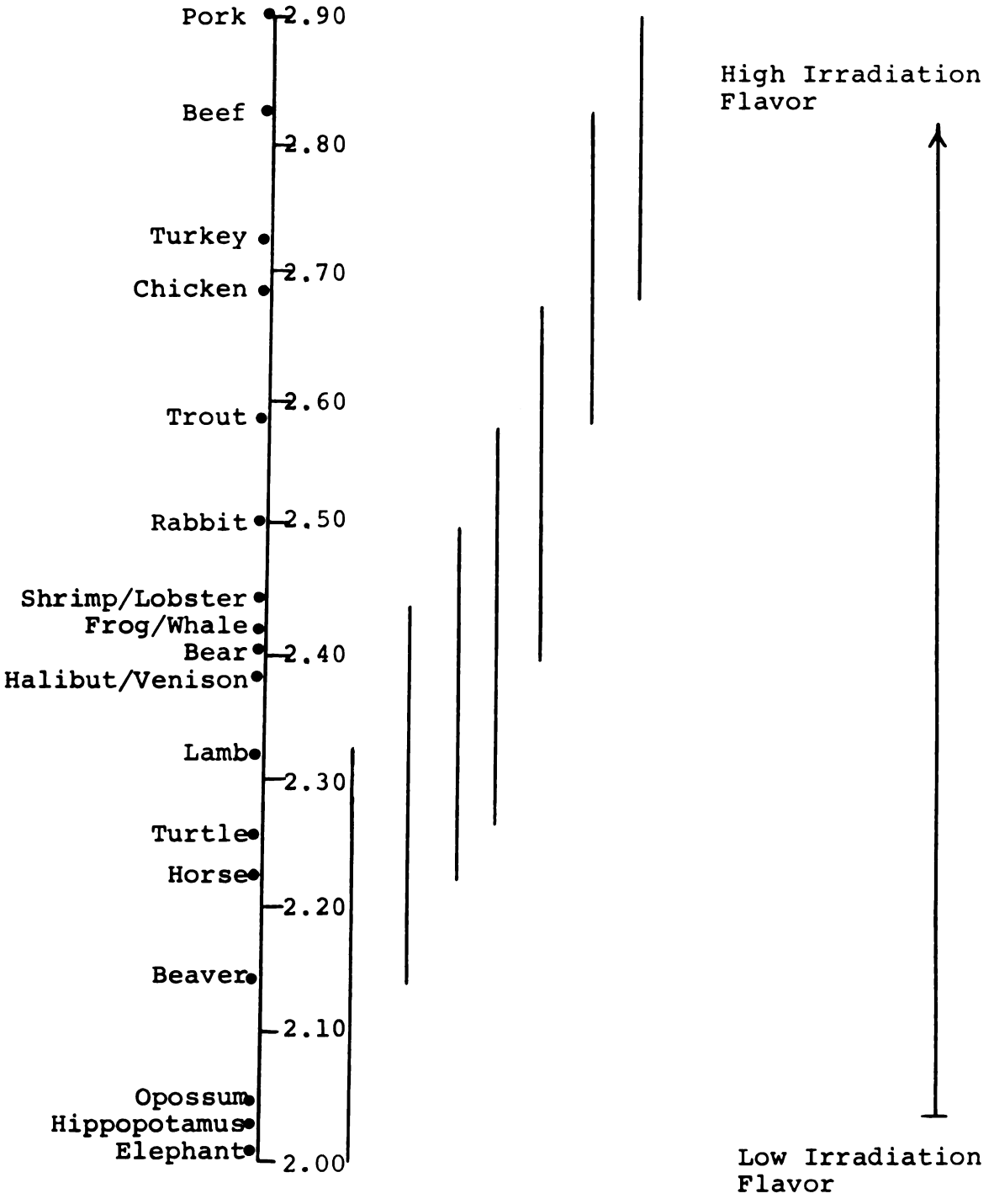
Pork - Hippopotamus = 2.90 - 2.02 = 0.88 > 0.35: Significant

. . . . . .

Pork - Chicken = 2.90 - 2.68 = 0.22 < 0.31: not significant

Determination of significant or not significant difference between two samples was continued until covering all twenty food samples. The results of differences among the animal protein food samples are summarized on the following Figure 2. Any sample means not covered by the same vertical line are significantly different. Otherwise, any means covered by the same vertical line are not significantly different.

It has been proved statistically that there was a significant interaction between kind of food and dose. If this interaction is large enough to be of biological importance, the effect of radiation on irradiated flavor produced in the foods should be discussed for each level of radiation, not on the basis of differences among the kinds of food averaged over all doses. Despite the evidence obtained from the statistical analysis of the interaction between kinds of food and dose, an inspection of the data reveals that this interaction is not of great biological significance. In general a food showing greater development than another displayed this greater sensitivity at all doses (Figure 3), and as a consequence, one can refer to this food as more sensitive to radiation than the other.




The numbers are overall means of samples for whole dose irradiated flavor scores.

Figure 2. Significant or not significant differences among the animal protein food samples. The samples are arranged as they are on Table 5.

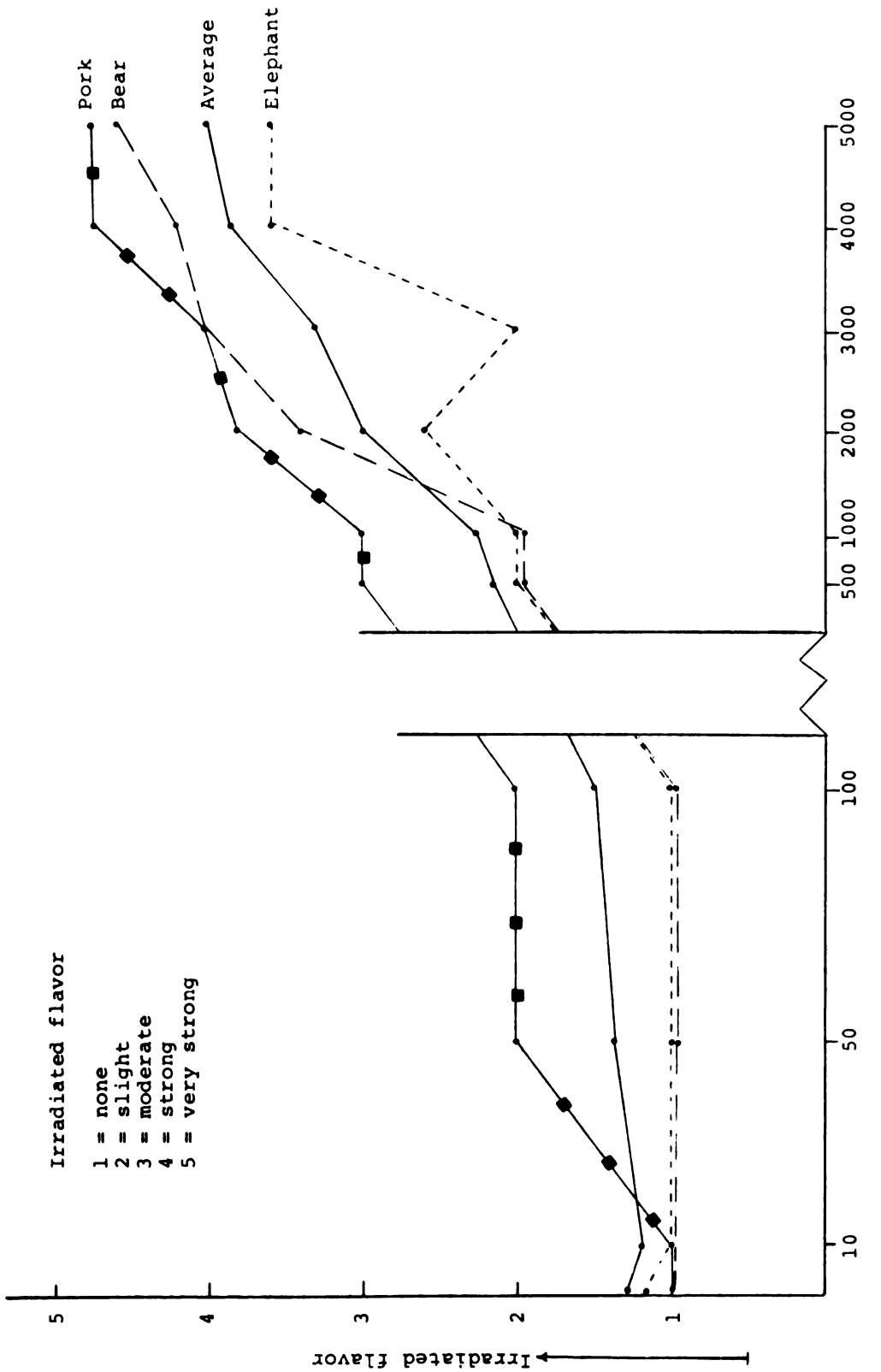



Figure 3. Curves showing irradiated flavor vs dose for pork (most sensitive to irradiation), elephant (least sensitive), bear (medium sensitive) and the average of 20 food samples.

To illustrate the way each animal protein food responded to irradiation, Figure 3 shows the relationship between irradiated flavor intensity vs. irradiation dose (Krad).

Table 6 gives the mean score of all doses of each animal protein food, mean of all foods and the standard error of the means.

Table 7 shows the means flavor intensity scores for all foods at each dose studied, and clearly shows increasing flavor intensity with increasing dose.

The data shown in Figure 2, indicate there is no evidence of a relationship between flavor sensitivity to irradiation of a food and the biological classification of the animal from which it is derived. For example, beef, deer, lamb, swine and hippopotamus belong to the same order of Artiodactyla in the class of Mammalia, but the flesh foods obtained from these animals were scored in a scattered manner over the flavor sensitivity range.

On the other hand, all the fish and sea foods investigated and the amphibian (shrimp, lobster, halibut, trout, whale and frog) have no significant difference in irradiated flavor intensity with respect to each other. As a group they fall in the middle of the flavor sensitivity scale.

Arranging the animal food samples in order from the lowest to the highest mean values of the irradiated flavor

TABLE 6.--Irradiated flavor means of animal protein food samples and their standard error.

| Food | Mean Score of All Doses |
|---|---|
| Elephant | 2.00 |
| Hippopotamus | 2.02 |
| Opossum | 2.04 |
| Beaver | 2.14 |
| Horse | 2.22 |
| Turtle | 2.26 |
| Lamb | 2.32 |
| Halibut | 2.38 |
| Venison | 2.38 |
| Bear | 2.40 |
| Frog | 2.42 |
| Whale | 2.42 |
| Shrimp | 2.44 |
| Lobster | 2.44 |
| Rabbit | 2.50 |
| Trout | 2.58 |
| Chicken | 2.68 |
| Turkey | 2.72 |
| Beef | 2.82 |
| Pork | 2.90 |
| Mean of all foods | 2.40 |
| Standard Error of each food mean | 0.0981 |

TABLE 7.--Irradiated flavor score means for all animal protein food samples at each dose.

| Dose, Krad | Means of Irradiated Flavor[a] |
|---|---|
| 0 | 1.30 |
| 10 | 1.22 |
| 50 | 1.36 |
| 100 | 1.51 |
| 500 | 2.15 |
| 1000 | 2.27 |
| 2000 | 3.03 |
| 3000 | 3.30 |
| 4000 | 3.87 |
| 5000 | 4.03 |

[a]Irradiated flavor intensity score range from one to five: 1 = none; 2 = slight; 3 = moderate; 4 = strong; 5 = very strong.

scores, or in other words from the least sensitive to the most sensitive in flavor change due to irradiation, as shown in Figure 2, reveals that the most intensively domesticated animals such as swine, beef, turkey and chicken yield foods that are the most sensitive to irradiation. Less domesticated or unusual animals such as elephant, hippopotamus and opossum give foods that are among the least sensitive of all observed.

Possible reasons for this finding may be:

1. Animals, such as elephant and hippopotamus ordinarily are wild (game) animals and the particular foods derived from them used in this study probably had such an origin. This is in contrast with the highly domesticated popular animals such as swine, beef, turkey and chicken. It is possible that the mode of living, environmental living condition, physical activity, variety of feeds, or even psychological condition of the animal, will have some effects on the animal sensitivity of flavor change due to irradiation. This is supported by the fact that all aquatic animals (sea foods and amphibian frog) have no significant difference in sensitivity to irradiation among each other. The effects of those factors to the physical or chemical nature of the animal foods which will contribute to the development of irradiated flavor are not exactly known.

2. Another possibility is the subjectivity of the sensory panel procedure. It is likely that lack of familiarity with the unusual animal foods may have affected the judgements on the irradiated flavor.

Somewhat contrary to the reports (Hannan and Thornley, 1957; Huber, et al., 1953; Coleby, 1959), in this present work no significant evidence observed in sensitivity difference among beef, chicken, turkey and pork. As shown in Figure 2, pork was the most sensitive to irradiation among all the animal foods observed. There is the possibility that this result which differs from that of other investigators may be due to differences in quality of the meats or part of the animal used as sample. More work is indicated to clear up this disagreement, such as the application of procedures and statistical analyses which will be more sensitive in determination of the flavor change due to irradiation among beef, pork, chicken and turkey. For example the evaluation of irradiated flavor intensity among these animal foods at the same dose at one time can be suggested. Although there was no significant difference in sensitivity among the common kinds of meats, from Figure 2 in this present work it can be observed that beef is more sensitive than chicken and turkey as Hannan and Thornley (1957) and Coleby (1959) have reported.

The contribution of animal fat to the irradiated flavor development seems not to be justified. From a

visual observation, opossum and beaver were among the fattest by the fact that thick layer of fat surrounded small piece of lean meat, but they belong to the least sensitive among other animal protein foods on the irradiated flavor scale. A report by Huber, et al. (1953) apparently supports this present observation. They stated that there was no significant difference of irradiated flavor between lean and fat beef. A report by Groninger, et al. (1953) stated that beef was more sensitive to irradiation than pork, regardless of the belief that peroxides were produced in higher quantities in pork due to the higher unsaturated fatty acid content which could contribute more in overall irradiated flavor development (Hannan and Thornley, 1957; Huber, et al., 1953; Coleby, 1959). Once again in this present study, the role of fat and lipids as important contributors to the development of irradiated flavor and odor is in question. It is indeed, true that the chemical nature and the role of lipids in the development of irradiated flavor in animal protein foods is not quite understood yet.

Because of the amino acid composition of the animal foods in this work were not known, we have no background to explain the distribution of the sensitivity of animal protein foods to radiation in relation to their amino acid composition.

Approach for Determination of "Threshold Dose" for Each Animal Protein Food

The term "threshold dose" occurs in the literature. This is considered to be the dose at which an irradiated flavor can be just detected. The data obtained in this study indicate a correlation of flavor intensity with dose. The intensity score scale of one to five which was used is arbitrary and not necessarily linear. There are some difficulties of interpretation of the data in defining a threshold dose for each food. Since there is interest, both in the threshold dose level itself for a given food and in the comparative value of threshold doses, there may be justification in the following effort.

The score value of "one" indicates absence of any irradiated flavor or odor. The score value of "two" indicates a level of irradiated flavor or odor just detected. One may consider a score just less than two to be the upper limit of absence of irradiated flavor. Above this value a flavor or odor is detectable (by definition). By plotting for each food flavor intensity scores vs. dose, and noting the dose corresponding to a 2.0 score, one can assign a dose value as the threshold dose. This is illustrated in Figure 4 for two meats, pork and bear. These estimates are more area estimates than precise point estimates since the volume of the data did not justify determining precise response curves. Appropriate response curves were drawn and threshold values made from these curves.

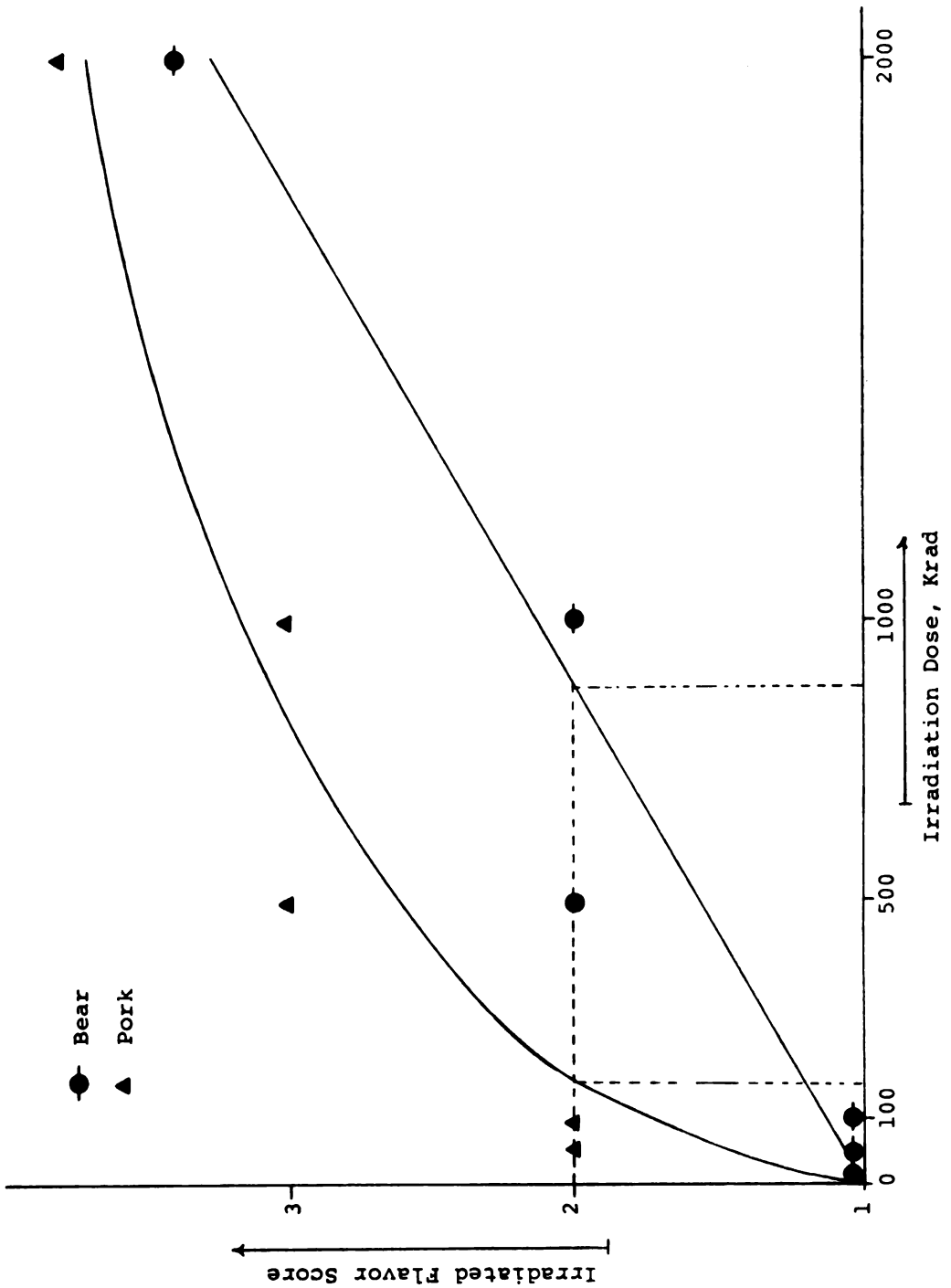



Figure 4. Irradiated flavor score vs irradiation dose. Threshold dose was determined at flavor intensity score of 2.

Hence the threshold values given in Table 8 should be recognized as only approximate. The threshold levels so determined for all protein foods investigated are listed in Table 8.

The data shown in Table 8 show the threshold dose range from 150 Krad (for turkey) to 875 Krad (for bear). In overall picture, the threshold dose is higher for the less sensitive animal food. But the correlation between threshold dose and the sensitivity is not necessarily linear, since the threshold doses were derived from plotting the score values of irradiated flavor of lower doses only (0 to 2000 Krad), and the sensitivity was determined from all dose treatments from 0 to 5000 Krad.

In 1962 it was reported that no off odor could be detected in beef up to 93 Krad but at 470 Krad it could be detected readily (Anonymous, 1962). Hannan and Thornley (1957) suggested the threshold dose of beef was about 100 Krad. The figure in this present work (Table 8) of 250 Krad for threshold dose for beef probably is greatly different from the dose range of the previous investigators. The deviation probably is due to sampling methods, statistical or cooking methods especially in picking the score value of taste threshold.

For lean pork Hannan and Thornley (1957) reported 250 Krad as its threshold dose. That figure apparently is fairly close to the figure of 175 Krad obtained in this study.

TABLE 8.--The "threshold dose" for each animal protein food investigated, determined at flavor intensity score value of 2 (slight irradiation flavor).

| Animal Food | Threshold Dose Krad |
|---|---|
| Pork | 175 |
| Beef | 250 |
| Turkey | 150 |
| Chicken | 250 |
| Trout | 450 |
| Rabbit | 350 |
| Shrimp | 250 |
| Lobster | 250 |
| Frog | 400 |
| Whale | 400 |
| Bear | 875 |
| Halibut | 500 |
| Venison | 625 |
| Lamb | 625 |
| Turtle | 450 |
| Horse | 650 |
| Beaver | 550 |
| Opossum | 500 |
| Hippopotamus | 525 |
| Elephant | 650 |

Coleby (1959) and Thornley (1957) both reported the same threshold dose of 250 Krad for chicken cooked by steaming, the same as obtained in this present work.

In 1967 Holston, et al. reported the optimum dose range for irradiation of fish and shellfish is 150-450 Krad. The data in Table 8 of this present study indicate a threshold dose range of 250-500 Krad for fish and seafoods (including whale, sea turtle and frog).

No data were available for other animal foods for comparison with this present study of threshold dose of irradiated flavor in animal protein foods.

Clearly more work is needed to obtain an accurate scientific explanation of the irradiated flavor and odor development in animal protein foods.

# SUMMARY AND CONCLUSIONS

The effect of gamma irradiation on changes of flavor in twenty selected animal foods was studied. The animals selected as the source of protein foods were beef, deer, lamb, swine, hippopotamus, bear, elephant, rabbit, whale, beaver, opossum, horse, chicken, turkey, sea turtle, frog, halibut, trout, shrimp and lobster.

In general, the most intensively domesticated animals such as swine, beef, turkey and chicken yield foods that are the most sensitive to irradiation. Less domesticated or unusual animals such as elephant, hippopotamus and opossum give foods that are among the least sensitive of all foods observed.

The importance of the contribution of animal fat to irradiated flavor development seems doubtful. Opossum and beaver meats were among the fattest, but were among the least sensitive to irradiation.

The "threshold dose" for each food was determined from the dose corresponding to a 2.0 flavor intensity score. As is to be expected, the threshold dose is higher for the less sensitive animal food.

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