1996


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THIAMINE, RIBOFLAVIN, AND SENSORY STABILITY OF IRRADIATED BROWN RICE

A Dissertation

Submitted to the Graduate Faculty of the Louisiana State University and Agricultural and Mechanical College in partial fulfillment of the requirements for the degree of Doctor of Philosophy in

The Department of Food Science

by

Linda Chamberlain Douglas
B.S., University of Southwestern Louisiana, 1991
M.S., Louisiana State University, 1994
December 1996

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DEDICATION

The author wishes to dedicate this dissertation to her husband, Lane, and her children, Jennifer and Mary Kathryn. Thank you for the love, support, and tolerance you have given me throughout this journey.
ACKNOWLEDGMENTS

The author wishes to give thanks to God for all the blessings He has bestowed upon her and her family.

Sincere appreciation is extended to my major professor, Dr. Joseph Liuzzo, for his encouragement and support.

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ABSTRACT

While brown rice is nutritionally superior to white rice, it is not shelf stable, becoming rancid in a matter of weeks. Preservation by refrigeration or freezing is expensive and not universally available. Therefore, other preservation methods, such as aqueous ethanol extraction of lipids, have been studied to overcome this problem. These methods also have limitations.

The effects of gamma-irradiation on the physicochemical properties and lipase activity in brown rice has been well documented. Gamma-irradiation has been shown to reduce lipase activity, presumably preserving sensory quality. A sensory study using two Louisiana grown varieties (‘Mars’ and ‘Lemont’) of irradiated brown rice performed at monthly intervals over a period of 6 months was conducted to provide the definitive data to assess this method of preservation for sensory effects.

Rough rice was de-hulled, irradiated at 0, 1, and 2 kGy, and stored for a period of up to 6 months at ambient temperature in sealed plastic bags. Duplicate samples were taken at monthly intervals starting with time 0 and evaluated for thiamine and riboflavin status using reverse phase HPLC and sensory quality using an experienced 8-person panel, respectively. These procedures were replicated 3 times to ensure scientific and statistical validity.
The 2 kGy treatment level produced the worst hedonic rating for both varieties. The 1 kGy radiation level did not significantly affect the sensory quality of either rice variety. Storage negatively affected nutty and milky attributes, which were stable to irradiation treatment. The rancidity attribute of brown rice samples, as perceived by the sensory panel, was negatively affected by irradiation treatment.

The effect of treatment and storage on thiamine and riboflavin levels of the rice samples was significantly different from the control only at the 2 kGy level Mars variety at 6 months of storage. However, treated samples of the Mars variety stored for a period of 6 months retained nearly 50% of original thiamine and riboflavin levels. Thiamine and riboflavin were essentially unaffected by storage or treatment in Lemont rice samples.
INTRODUCTION

Rice (*Oryza sativa*) is one of the world's major cereal grains, serving as a staple for more than 50% of the world's population. In some areas, rice comprises up to 90% of calories as well as providing a significant portion of protein consumed daily (Juliano, 1985). Unhulled rice, or rough rice, is dehulled and then milled to remove the outer bran layers, thereby reducing nutritional quality while greatly increasing shelf life. Furthermore, while brown rice is nutritionally superior to white rice because of bran layer retention, it is generally not the product of choice. The bran layer of brown rice contains, in addition to beneficial B-vitamins, oils that are subject to rancidity quite quickly after the dehulling process. This leads to stale off-flavors and the general bran flavor of brown rice. Brown rice also requires a much longer cooking time, which can be a major factor in developing countries where cooking fuel costs are critical.

There have been many attempts to stabilize brown rice using different technologies to yield a shelf-stable product. These same technologies have also been explored in order to reduce the cooking time of brown rice. Gamma irradiation is among the techniques which have been reported. The relative safety of irradiated foods has generated great interest. The Joint
FAO/IAEA/WHO Expert Committee has endorsed irradiated foods as being safe for human consumption and nutrition (WHO, 1981). Various physicochemical effects of irradiation on brown rice have been studied and reported in the literature (Sabularse, 1988). However, there is a lack of sensory data to indicate the ultimate desirability of such treatments on brown rice.

In the Fall of 1995, this study was initiated to determine the thiamine and riboflavin stability as well as selected sensory characteristics of brown rice irradiated at the 0, 1, and 2 kGy levels held in storage for a period of six months. Brown rice obtained from the Louisiana Experiment Station in Crowley was dehulled, irradiated, and stored at ambient temperature (25°C) in heat sealed polyethylene bags. Monthly sensory evaluation of samples was conducted and a sample was stored at -20°C for vitamin analysis. Riboflavin and thiamine were analyzed by high performance liquid chromatography.
LITERATURE REVIEW

RICE

Rice, (Oryza sativa) was first grown in the area encompassing central India, Burma Thailand, Laos, Vietnam, and southeastern China. Historical records indicate the independent and simultaneous cultivation of this semiaquatic, annual grass in these areas between 2000 and 1500 B.C. The crop soon spread to the Philippines and northern Australia, before being traded in the inhabited areas of Asia and the Middle East, as well as throughout the European continent (Chang, 1976).

Rice became a commercial crop in the United States in 1686, although it was traded in the New World prior to that time. By 1888, Louisiana and Texas would have an established yearly production of rice, followed by Arkansas in 1904, California in 1912, and Mississippi in 1942. Since that time, Missouri and Florida have also begun rice cultivation on a commercial scale (Dethloff, 1982). It can be assumed that the broad range of conditions acceptable for rice culture have no doubt contributed to its popularity as an agricultural commodity.
Structure and Composition of the Rice Grain

Figure 1 depicts the structure of the rice grain, which consists of the hull, pericarp, testa, embryo, aleurone layer, and endosperm. All parts of the grain, with the exception of the hull are edible, and can make important contributions to human nutrition. The hull, or outer covering of the rice grain is the protective coating against insects and moisture. It consists of 2 parts; the lemma and palea, which cover the dorsal and ventral parts of the grain, respectively. These two portions are joined longitudinally, and contain minute amounts of numerous minerals such as silica, calcium, sodium, magnesium, potassium, manganese, aluminum, iron, copper, and zinc. With the exception of fiber content, this portion of the grain is considered unacceptable as human food (Juliano and Bechtel, 1985).

The pericarp, testa, and aleurone comprise the caryopsis, which is exposed upon dehulling. These layers, along with the germ or embryo are collectively referred to as the bran layer. This bran layer amounts to approximately 8% of the rice grain by weight. The majority of the oil, fiber and protein are to be found in the bran, as well as most of the vitamins and minerals the rice grain has to offer for human nutrition (Lu and Luh, 1991).
Figure 1. Structure of the Rice Grain (Champagne and Hron, 1992a).
However, the milling process removes these layers along with the nutritive value they offer.

The subaleurone layer is subsequently removed by the milling process, accounting for approximately 4% of the rice grain by weight. This layer is commonly known as rice polish, and is rich in protein, but contains less fat than the bran. When the subaleurone layer is polished away, the resulting product is white rice. White rice is composed of starchy endosperm. This endosperm tends to contain more protein in the outer layers, with mostly starch at the core. However, white rice is widely considered to be a good source of carbohydrate and a fair source of protein (de Lumen and Chow, 1991).

World Rice Production and Consumption

Wheat is the only human food crop which has a larger annual production than rice. About 60% of the world depends upon rice as a daily staple and about 90% of this production and consumption occurs in Asia. Figure 2 shows the world rice production and human consumption over a 12-year period. As shown, rice production has increased steadily over time. However, this increase is due to increased yield, as acres planted have not increased significantly. Most of the trade in rice, about 70%, is in the polished, white form.
The United States exports a tremendous amount of its production, amounting to about 20% of the total world trade. This is possibly due to a combination of high yields obtained and low national consumption compared to other rice producing areas of the world (Childs, 1989). Nevertheless, U.S. consumption of rice is rising, due to dietary trends, increasing Asian and Hispanic immigration, and marketing tactics by the Rice Council (USDA, 1992).

![World Rice Production and Consumption Over 12 Years](image)

Figure 2. World Rice Production and Consumption Over 12 Years (Marshall and Wadsworth, 1994).

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Brown Rice

Consumers are shifting toward an increased use of brown rice as a result of health concerns. Brown rice is simply dehulled rice which has not undergone milling and polishing. It retains the nutritive substances found in the bran layers which are normally removed to produce white rice. Brown rice contains approximately 5 times the thiamine, and 3 times the niacin, phosphorous, potassium, iron, sodium, and riboflavin of white rice (Kennedy, 1980). The bran layers have also been found to possess cholesterol-lowering activity (Kahlon et al., 1989; Kestin et al., 1990). However, there are storage problems such as rancidity that limit the shelf life of brown rice after dehulling to little more than three months. This is a contributing factor to the relatively low consumption rate of brown rice, which is about 3% of total rice consumption (Childs, 1991).

Rancidity in Brown Rice

The oil present in brown rice undergoes two types of deterioration: hydrolytic and oxidative. Lipase, whether endogenous or microbial, acts as a catalyst in the hydrolysis of the oil present in the bran. The endogenous lipases are activated upon tissue disruption, as in dehulling. Prior to dehulling, the enzyme and substrate are separate. Lipases are housed in the testa layer,
the oil residing in the aleurone and germ layers. At the time of dehulling, the oil makes contact with the enzymes, thereby initiating the hydrolysis of triglycerides to free fatty acids. Any lipases of microbial origin may also act in the same fashion upon the freed oil (DeLucca et al., 1978). There are many factors which influence the rate of hydrolysis resulting in free fatty acids. Factors such as moisture, temperature, surface area damage, and length of storage are among the most important considerations.

Free fatty acid levels can range from 6 to 25% after 6 months of storage (Sharpe and Timme, 1986). Free fatty acids are important to consider in the sensory quality of brown rice. By themselves, they contribute a soapy flavor to the rice (Barnes and Galliard, 1991). It is the subsequent oxidation of the free fatty acids which constitutes the off-flavors and odors of rancid rice. This oxidation can be catalyzed by enzymes, but this is not always the case.

Lipoxygenase, found in the germ or embryo, catalyzes the oxidation process which yields hydroperoxides from free fatty acids. These hydroperoxides can then further react, yielding such secondary oxidation products as aldehydes and ketones which, in turn, contribute off-odors as well as off-flavors. The amount of free fatty acids in brown rice is critical since the enzyme acts on them preferentially. Therefore, the amount of enzymatic
oxidation products formed is directly related to prior hydrolysis of the rice bran oil (Galliard, 1989). Nonenzymatic oxidation is induced by metals present in bran or metals introduced by equipment. Other factors which contribute to oxidative damage of the oils found in brown rice are light, radiation, heat, and oxygen. These reactions are inhibited by tocopherols present in the bran, which act as antioxidants, playing a protective role for the rice bran oil (Sowbhagya and Bhattacharya, 1976).

Efforts to stabilize brown rice may lead to further rancidity by the redistribution of oil, destruction of natural tocopherols, and increased exposure to oxygen (Eriksson et al., 1969). Figure 3 shows a simplified scheme for the routes of damage to the oil in rice bran.

Stabilization Efforts with Respect to Hydrolysis

Control of hydrolytic rancidity in brown rice has focused on three areas: denaturation and inactivation of lipases using heat, removal of rice bran oil with solvent extraction, and the denaturation and inactivation of lipases with ethanol and ethanol vapors. Processes using heat have included parboiling, soaking, and steaming. These methods produce a quick-cooking brown rice similar to the instant rices on the market today (Ozai-Durrani, 1948, 1956; Miller, 1963; Roberts et al., 1980; McCabe, 1976). Although the lipases are inactivated,
lipases

RICE BRAN OIL \rightarrow \rightarrow \rightarrow \rightarrow \rightarrow \rightarrow \rightarrow FREE FATTY ACIDS

\text{autoxidation} \quad \text{lipoygenase}
\text{photo-oxidation} \quad \text{autoxidation}
\text{photo-oxidation}

HYDROPEROXIDES

\downarrow
\downarrow
\downarrow
\downarrow
\downarrow
\downarrow
\downarrow

SECONDARY OXIDATION PRODUCTS

dipersoxides, aldehydes, semi-aldehydes, semi-aldehydes,
alcohols, ketones, acids, epoxides, cyclic fatty acid monomers,
dimers, polymers, etc.

Figure 3. Routes of Hydrolytic and Oxidative Damage of Rice Bran Oil (Champagne, 1994a).
significant losses of vitamins and minerals occur. Thiamine, riboflavin, and iron losses of up to 67% have been reported (Roberts et al., 1980). Vitamins and minerals are conserved when heat treatments involving hot air are used. However, the resulting brown rice product is puffed and has a toasted flavor (Bardet and Giesse, 1961). Heated gasses have been used to blanch brown rice. This process does not reduce cooking time, but does lower the original free fatty acid content. Total B-vitamins are reduced, but little mineral loss occurs (Bhattacharya and Ali, 1985). The value of these treatments, however, is negatively compensated for by the increased oxidation potential which results from denaturation of hemoproteins, and redistribution of oil (Champagne, 1994b).

Solvent extraction using petroleum ether or hexane reduced free fatty acid levels from 22% to 3.7% over an 80-day storage period (Kester, 1951). Protein, fiber, carbohydrate, and minerals were conserved, although thiamin loss averaged 20%. However, this did not prevent oxidative processes from taking place.

Liquid ethanol and ethanol vapors have been used to stabilize brown rice to hydrolytic rancidity (Champagne et al, 1990; 1991). The stabilizing action of the ethanol is thought to be denaturation and deactivation of lipases and the
killing of lipase-producing microorganisms on the surface of the rice grain. Higher temperatures and longer processing times were associated with lower free fatty acid levels in the rice. Treatment temperatures ranged from 24° C to 70° C and extraction times ranged from 20 minutes to 1 hour. Ethanol vapors produced similar reductions (Champagne and Hron, 1992b).

Brown rice stabilized by the above methods (including ethanol and ethanol vapor processes), is used to produce stabilized rice bran. However, these products are still susceptible to oxidative processes, which are more problematic from a sensory standpoint.

**Stabilization Efforts with Respect to Oxidation**

The absence of oxygen precludes any oxidative damage. Oxygen levels of approximately 1% are required to achieve this effect. This is a difficult task and is usually cost-prohibitive. Therefore, efforts to utilize low temperature storage combined with modified atmospheres has been investigated (Ory et al., 1980). These methods slow oxidative damage, but do not stop it altogether.

The use of antioxidants is effective in preventing nonenzymatic oxidation, but is not useful in enzymatic oxidation. An antioxidant prepared from rosemary at the 300 ppm level demonstrated effectiveness against oxidation in brown rice (Loliger, 1989). Increased consumption of brown rice
and the growing demand for products derived from it, such as rice bran oil require further investigation into prevention of rancidity.

GAMMA IRRADIATION OF FOODS

The process of food irradiation involves exposing foods to ionizing radiation, usually gamma rays or X-rays to achieve specific effects. Among the desired effects are deinfestation, pasteurization, destruction of pathogens and parasites, and inhibition of sprouting and subsequent senescence (Urbain, 1986). Irradiated foods have been exhaustively studied and are considered to be safe for human consumption in all respects (Urbain, 1986).

The first serious application of irradiation to food preservation was executed by Schwartz in 1921, when he patented a process using X-rays to eliminate *Trichinella spiralis* in pork. Other innovations such as irradiation of foods sealed in metallic cans, irradiation of ground beef, seafood, dairy products, vegetables, and fruits soon followed. After this time, experiments on the use of irradiation to sterilize foods and drugs were conducted, including the use of modified atmospheres to protect sensory attributes. These experiments were ill-funded, utilized crude equipment, and usually provided far more questions and problems than answers. The development of food irradiation began in earnest with the U.S. Navy’s increased interest in reducing shipboard
refrigeration leading to a radiation research contract with the Massachusetts Institute of Technology (MIT). The Army Quartermaster Corps soon followed by sponsoring another contract with MIT for deinfestation of several types of rations. Military need has historically resulted in great strides in food processing. Therefore, these and many other irradiation studies funded by the U.S. government were similarly instrumental in the development of irradiation processing techniques commonly used the world over (Josephson and Peterson, 1982).

Since the 1950's, over 9000 research studies have been conducted in the area of food irradiation. Many of these studies have been conducted to determine the safety of irradiated foods for human consumption. The word "safe" in this context refers to toxicological, nutritional, and microbiological safety of the treated food. In addition, the food must also be free of any radioactivity or unusual radiolytic products. Ionizing radiation is legally defined as a food additive by the Food, Drug, and Cosmetic Act of 1958, and is therefore not considered a physical process (Diehl, 1982).

Radiation Sources

The main sources of radiation for food use are radioactive isotopes such as cobalt-60 or mechanical sources such as electron beam generators. These
methods produce predictable, precise ionizing energy suitable to achieve an adequate exposure (WHO, 1994). Gamma sources are usually stored under water for effective shielding of ionizing energy when not in use. For commercial use, the gamma source is held in a concrete shielded area, and the food target is conveyed past the source on a conveyor system. After sufficient exposure, the gamma source is again stored under water. Cobalt-60 does not contribute to radioactive waste of long duration since it has a half-life of 5.3 years and it decays to form nonradioactive nickel (WHO, 1994).

Radiation from electron beam accelerators is produced using electricity, and provides rather shallow penetration. This type of radiation source is limited, but is used extensively on grain and meats for surface decontamination. Converting the electron beams into X-rays provides essentially the same penetrating power of gamma rays, allowing uniform penetration of the target food.

However, there are some advantages to machine generated radiation sources which gamma sources cannot provide. For example, the electric radiation source can be turned off when not in use. It also has the distinct advantage of not having to be replenished as well as being readily available. The established history of usage and high throughput potential of these systems
make them advantageous. However, there are some limitations. The machines used are highly complex and require regular, specialized maintenance. They also require huge amounts of energy for operation and cooling. Still, the process for exposing foods to machine generated ionizing energy is practically identical to that used for irradiation with gamma rays (WHO, 1994).

**Mechanism of Food Irradiation**

Partial destruction of the genetic material of living cells is responsible for the efficacy of the irradiation process. Microorganisms responsible for spoilage as well as disease are effectively destroyed in this manner. This damage is accomplished either through direct effects of the energy on DNA or through secondary effects involving the production of free radicals and ions which also attack DNA. These effects are purely chemical, involving the electrons which surround the atomic nucleus of cells, but never involving the nucleus itself (WHO, 1994).

Primary, or direct effects, are produced by high energy electrons and may induce one or all of the following: The removal of an electron, known as ionization; the loss of hydrogen, or dissociation; and raising the electrons in a molecule to a higher energy level, or excitation. Ionization, dissociation, and
excitation produce a highly reactive pool of free radicals, which then cause secondary, or indirect effects.

Secondary effects resulting from the above factors include dimerization, recombination, and disproportionation. Dimerization, or electron capture, involves other molecules in the food otherwise unaffected by the ionizing treatment. Disproportionation involves the production of a substance which may not have originally resided in the target food (WHO, 1994). These effects are not unusual to any process which significantly alters or denatures food products, such as normal cooking or processing methods.

All cooking and processing methods involve denaturation or the breaking of chemical bonds. Radiolysis is the term used for breaking chemical bonds by the use of ionizing energy. Radiolysis produces unstable products which convert to stable end products. These end products are identical to those found in foods exposed to light or heat (Diehl, 1990).

Temperature has an effect upon the outcome of the irradiation process. Cooler temperatures and the absence of oxygen produce a more desirable product since the radicals produced tend to react with each other rather than with the food (WHO, 1994). Foods processed in the frozen state by ionizing energy retain the highest quality. Proteins, carbohydrates, and fats tend to
undergo oxidative as well as hydrolytic degradation when irradiated. These macronutrients do not undergo significant loss of nutritional value, although sensory effects may be detected (CAST, 1986).

Current Applications of Irradiation

Beneficial effects of food irradiation can be classified in a variety of ways. Functional classifications, with their corresponding doses and target foods are presented in Table 1. Fresh products, such as fruits and vegetables, retain their uncooked characteristics. At the 1 kGy dose level, inhibition of sprouting of tubers, roots, and bulbs is accomplished. Delay of ripening and subsequent senescence of a variety of fruits can be achieved, as well as insect deinfestation is well documented at 1 kGy (WHO, 1994). This level of ionizing energy is considered low-dose. Medium-dose (1-10 kGy) is used for the extension of shelf-life of fish, molluscan shellfish, crustacea, some fruits, raw or frozen meats, and dehydrated vegetables. High dose irradiation of foods is accomplished at the 10 to 50kGy dose level and is used for industrial sterilization, for some meats, poultry, and seafood, and sterilized hospital diets. Some spices and enzyme preparations are also treated at this extreme level.

Another way of classifying ionizing energy doses is in the use of the following terminology, established in 1964, which groups dose levels according
### Table 1: Functions of Food Irradiation (WHO, 1994)

<table>
<thead>
<tr>
<th>Function</th>
<th>Dose (kGy)</th>
<th>Products Irradiated</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Low-Dose (up to 1k Gy)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inhibition of sprouting</td>
<td>0.05-0.15</td>
<td>Potatoes, onions, garlic</td>
</tr>
<tr>
<td>Insect deinfestation and parasite disinfection</td>
<td>0.15-0.5</td>
<td>Cereals, legumes, fruits, meats, fish</td>
</tr>
<tr>
<td>Delay of ripening</td>
<td>0.5-1.0</td>
<td>Fresh fruits and vegetables</td>
</tr>
<tr>
<td><strong>Medium-Dose (1-10 kGy)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Extension of shelf-life</td>
<td>0.5-1.0</td>
<td>Fresh fish, strawberries</td>
</tr>
<tr>
<td>Elimination of microorganisms</td>
<td>1.0-7.0</td>
<td>Seafood, poultry, meats</td>
</tr>
<tr>
<td>Improving technological food properties</td>
<td>2.0-7.0</td>
<td>Grape juice yield, reduced cooking time of dehyr. vegetables</td>
</tr>
<tr>
<td><strong>High-Dose (10-50 kGy)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Industrial sterilization</td>
<td>30-50</td>
<td>Meat, poultry, seafood, sterilized hospital diets</td>
</tr>
<tr>
<td>Decontamination of some food additives and ingredients</td>
<td>10-50</td>
<td>Spices, enzyme preps.</td>
</tr>
</tbody>
</table>

to their microbiological objectives. These terms were established by Goresline (1964). The World Health Organization reported these terms as follows (WHO, 1994):
Radappertization: The treatment of food with a dose of ionizing energy sufficient to prevent spoilage or toxicity of microbial origin no matter how long or under what conditions the food is stored after treatment, provided it is not recontaminated. This is also called sterilization. The required dose is usually in the range of 25-45 kGy.

Radicidation: The treatment of food with a dose of ionizing energy sufficient to reduce the number of viable, non-spore-forming, pathogenic bacteria to such a level that none is detectable in the treated food when it is examined by any recognized bacteriological testing method. Such treatment also inactivates foodborne parasites. The required dose is in the range of 2-8 kGy. The term may also be applied to the destruction of parasites such as tapeworm and Trichinella in meat, in which case the required dose is in the range of 0.1-1 kGy.

Radurization: The treatment of food with a dose of ionizing energy sufficient to enhance its keeping quality by causing a substantial reduction in the numbers of viable spoilage microorganisms. The required dose is in the range of 0.4-10 kGy.

Irradiation of Cereal Grains

The main purpose for the irradiation of cereal grains is for the control of insect infestation. Chemical fumigants have been widely used, but are being viewed with suspicion by the modern consumer and, therefore, falling into disfavor. A number of chemical pesticides used as deinfestation fumigants have been banned. Common problems with these agents are toxicity posing a hazard to the worker using them, development of resistance in the target insects, residues in the grain, and temporary quarantine of the commodity.
Deinfestation requires an ionizing energy dose of 1 kGy or less, and has been used in the former Soviet Union since the early 1980's (Diehl, 1990). This is being accomplished with the use of electron accelerators and minimizes losses due to insect damage. With this method, the grain is moved past the accelerators at high speed through wind tunnels. This is done at the dock during unloading operations, and is cost-effective, and suitable for high-volumes (WHO, 1994). Vitamin retention is a critical factor to consider in the irradiation of cereal grains. Studies have shown a 90% retention of the B-vitamins thiamine, riboflavin, and niacin in wheat irradiated at the 2 kGy dosage level (Vakil et al., 1973). Fat soluble vitamins, such as vitamin E, were retained in oats at greater than the 90% level when irradiated at 1 kGy (Diehl, 1979). Other cereal grains such as corn, sorghum, and millet have been shown to retain vitamins and other nutrient levels with normal irradiation processing doses (Murray, 1983).

Irradiation of Brown Rice

Irradiation studies on brown rice have shown a decreased cooking time, increased water uptake, and increased starch leaching into cooking water (Sabularse et al., 1991). Mars, Tebonnet, and Lemont varieties were irradiated at 1 and 2 kGy, and all responded similarly with regard to physicochemical
properties (Sabularse et al., 1992). Irradiation was also reported to decrease lipase activity as well as free fatty acid content of brown rice (Liuzzo et al., 1996). There have been few studies reporting sensory changes or quality of irradiated brown rice. However, Ismail et al. (1978), and El Saadany et al. (1979), reported decreased eating quality of Egyptian brown rice at the 100 Krad level. Wang et al. (1983), did not find significant changes in the cooking quality of rice irradiated below the 300 Krad level. The effect of cathode ray irradiation on brown rice quality were investigated by Umeda et al. (1968). They found that brown rice irradiated at 30 Krad exhibited an off flavor, which decreased upon storage. Radiation doses below this level had no effect. These studies are somewhat conflicting, and provide rather amorphous information as to the specific changes in the eating quality of irradiated brown rice.

SENSORY ANALYSIS

Analytical methods of assessing sensory quality have evolved from the rudimentary judgement of good or bad edibles and utensils to the systematic assessment of products for consumer trade. The history of modern sensory analysis is based on efforts to provide acceptable food products to American soldiers during war time (Pangborn, 1965). The analyses conducted may include the sensory attributes pertaining to a product’s appearance,
odor/aroma/fragrance, consistency/texture, and flavor. The sensory panelist judges these attributes through the mechanisms of vision, touch, olfaction, chemical factors, gustation, and hearing. These judgements, translated into measurable terms, provide data on which decisions in quality control, product development, and research are made (Meilgaard et al., 1987).

In conducting a sensory study, the investigator must first determine the objective of the study. Overall project needs may include ingredient substitution or product improvement. This definition determines the type of test needed to gather the appropriate data. Test objectives must then be determined. Attribute difference, and overall acceptability are but two of the many parameters which may require determination. Sample screening must be conducted to determine proper handling and to define appropriate sensory terms and test applications. Test design, including selection and training of sensory panelists, design of scoresheet, plans for sample preparation and presentation, as well as data analysis techniques must be determined in advance of actual testing. Conducting the test and subsequently analyzing the data are then made possible through careful consideration of project and test objectives, sample screening, test design, and testing methods (Erhardt, 1978).
Sensory Attributes

Appearance is often the first and only attribute upon which a sample or product is judged. Therefore, it may be vitally important to mask a test sample’s appearance in order to reduce bias. This is done through the use of red, green, or blue lights, covered opaque or translucent containers, or other cloaking methods. Sample odor, which may be detected simultaneously, or in advance of, appearance, is important to overall flavor impression. Sensory terms defining aroma or odor are difficult to standardize, with many interchangeable terms being used for impression description.

The texture of a product is determined in the mouth, and is categorized by the following terms taken from Meilgaard et al. (1987):

- **Viscosity**: refers to homogeneous Newtonian liquids
- **Consistency**: refers to heterogeneous liquids and semi-solids
- **Texture**: refers to solids and semisolids

Viscosity of liquids is the rate at which the liquid flows, usually under the force of gravity and refers to substances with characteristics ranging from water to jelly. Consistency refers to the sensorial impression of substances such as sauces, syrups, and cosmetics. Texture is a sensorial impression of the internal
and external structure of a substance. Texture can be expressed in terms of a substance’s reaction to stress as well as tactile properties.

The flavor attribute is defined as the impressions perceived via the chemical senses from a product in the mouth. Thus, flavor encompasses aromatic sensations perceived by olfaction, soluble substances perceived by the taste buds, and sensations of feeling in the interior of the mouth, throat, and nasal passages (ASTM, 1987).

Sample Handling

Samples for sensory analysis must be weighed or otherwise measured accurately, with adequate and uniform sample size being important in minimizing bias. Care should be taken in container selection, as the containers should be uniform and odorless. Preparation, such as cooking methods should be chosen for suitability, repeatability, timing, and ease. Serving temperature, if important, should be carefully considered, and appropriate timing to achieve temperatures planned. Random coding of samples is normally employed for identification and bias reduction purposes.

Panel Training

At a minimum, panelists require instruction on scoresheet use, sample handling, test procedures, and type of response required. A thorough
familiarity with testing procedures and the nature of panel judgements is crucial to test outcomes. Panel training may also extend to recognition of specific attributes. The training of panelists on attribute recognition also includes terminology development and practice using scales. This may be accomplished individually or in a group setting. Of utmost importance however, is that all panelists adopt common terminology (ASTM, 1993).

**Motivation, Reward, and Feedback**

Rewards for panelists may be short or long term. Short term rewards, such as candy, snacks, and small tokens can provide a small measure of motivation for the sensory panelists. A sense of importance derived from the involvement in meaningful work provides the larger share of motivation, however. For longer-term projects, parties, recognition in the department or company, and other rewards are useful in maintaining active sensory panelists. Feedback from the project leader can be helpful, giving the panelist the sense of “a job well done” is often the best motivator. Feedback from panelists is also useful in the refinement of test procedures and terms. Open communication should be encouraged between panelists and the project leader at all times (Zook and Wessman, 1977).
Measuring Sensory Responses

Sensory analysis utilizes people as instruments to qualify and quantify sensorial parameters of interest in a given product. Sensory data may be characterized in the following manner:

- **Nominal data:** (Latin *nomen* = name): the items examined are placed in two or more groups which differ in name but do not obey any particular order nor any quantitative relationship; example: the numbers carried by football players.

- **Ordinal data:** (Latin *ordinalis* = order): the panelist places the items examined into two or more groups which belong to an ordered series; example: slight, moderate, strong.

- **Interval data:** (Latin: *inter vallum* = space between ramparts): panelists place the items into numbered groups separated by a constant interval; example: first, second, third.

- **Ratio data:** Panelists use numbers which indicate how many times the stimulus in question is
stronger (or saltier, or more irritating) than a reference stimulus presented earlier.

(Meilgaard et al. 1987).

The terms "scaling" refers to the type of sensory measurement in which panelists rate a sample in comparison to a control, or reference, on either a categorical, a linear, or a magnitudinal scale. The simplest measurement methods that will deliver needed data should be employed. Methods simplification saves time and minimizes errors.

**Descriptive Analysis**

Descriptive analyses are frequently used to track sensory changes in a sample over time, and require the use of a trained panel to detect and describe selected sample characteristics. The objective of descriptive tests is to identify attributes which need to be analyzed, and then to determine their intensity (ASTM, 1993). Attributes selected for analysis are introduced to the panelists individually and are represented by single foods or substances. For example, to identify a dairy note in a particular sample, panelists may be given milk or cream as the indicating flavor attribute. Green notes may be represented by celery, bell pepper, or other green vegetable matter which most closely matches the flavor of the sample. Panelists may be directed to choose descriptive terms
from a list, then match them to the representative foods tasting most like the flavors which compose the sample of interest. Linear scales may be used to indicate the intensity of a given attribute. The line scale is usually about 15 centimeters in length, and may be anchored at both ends. The sensory panelist estimates the intensity of a sample attribute corresponding to some point on the line, ranging from 0 (none) to 15 (very), by marking through the line at the desired point. Responses may be measured by some instrument or ruler and expressed as centimeters. These data may then be analyzed statistically (Meilgaard, 1987).

**Hedonic Scale**

The hedonic scale is a type of test which determines the degree of acceptance that a panelist rates a sample. In hedonic scaling, panelists rate a sample according to positive and negative categories provided on the scoresheet. There may be as few as 5 categories to as many as 11. The hedonic tests which give the most information have an equal number of positive and negative responses, with a neutral response in the center. The 9-point hedonic scale is one such balanced scale, with the following descriptive categories: like extremely, like very much, like moderately, like slightly, neither like nor dislike, dislike slightly, dislike moderately, dislike very much, and
dislike extremely. The categories are usually arranged vertically, but may be horizontally oriented, but always in descending order, from like extremely to dislike extremely. The hedonic scale measures a panelist's overall liking for a sample and may be used with experienced or new panelists. In this method, samples are presented for evaluation without any attempt to lead panelist response. Instructions are given regarding use of the scale, but the response must be made by each person according to his own opinion.

THIAMINE AND RIBOFLAVIN

Off-flavor in stored brown rice results from aldehydes, and ketones, as well as lipid, amino acid, and vitamin decomposition products. Moreover, cooked rice flavor has been simulated by exposing an aqueous combination of cysteine, cystine, and riboflavin to sunlight (Obata and Tanaka, 1965). Therefore, vitamin stability may be a critical factor in considering sensory quality of irradiated brown rice.

Vitamin losses resulting from food irradiation has yielded mixed results. Kung et al. (1953), found that 23% of the riboflavin in whole fluid milk was destroyed at the 0.24 Mrad dose level, and 35% was lost at the 0.50 Mrad level. Riboflavin loss in evaporated milk at these same dose levels was 18% and 28%, respectively. The lower water content may play a part in protecting
the vitamin. Ziporin et al. (1957), found that riboflavin losses in turkey and beets was significant at doses 2.79 and 5.58 Mrad, but much less than the 70% to 95% losses in thiamine, respectively. However, Urbain (1986), states that vitamin losses in irradiated foods are no greater than with traditional forms of food processing.

**Thiamine**

Vitamins are substances required by the body in small amounts supplied by the diet. Water soluble vitamins are widely distributed in nature, required daily by humans, and excreted freely in the urine. The B-vitamins, named after the test tube label “B”, were once thought to be one substance, but were later separated into fractions B₁, B₂, and so on (Whitney et al., 1987).

Thiamine, also known as vitamin B₁, was identified through the vitamin-deficiency disease, beriberi. Thiamine is water-soluble, and is not stored in the tissues. Clinical effects of thiamine deficiency on the gastrointestinal system include anorexia, indigestion, severe constipation, gastric atony, and deficient hydrochloric acid secretion. Nervous system effects include depressed neuronal activity, diminished alertness and reflex response, resulting in apathy and fatigue. Continued thiamine deficiency results in demyelinization of the nerves, resulting in nerve irritation, pain, prickly sensations, numbness, and
paralysis. Cardiovascular effects include weakening of the heart muscle and
dilation of peripheral blood vessels, resulting in edema of the extremities and
eventual cardiac failure (Williams, 1989).

Thiamine’s role in metabolism is in the molecule thiamine pyrophosphate
(TPP). TPP is the enzyme responsible for the conversion of pyruvate to acetyl
CoA for entry into either the Krebs cycle to produce energy for cellular
functions or to the lipogenesis pathway through the synthesis of fatty acids.
Carbohydrate intake increases thiamine demand, but fat and protein intake
spares thiamine, with excess being constantly excreted in the urine.

Thiamine is widely available in plant and animal foods including whole or
enriched grains, legumes, eggs and fish. Deficiency states exist under
starvation conditions, alcoholism, and irregular food patterns such as veganism.
Thiamine contents of selected foods are given in figure 4.

Riboflavin

Riboflavin, also known as vitamin B₂, derives the name from its component
sugar, deoxyribose and the from the Latin word flavus, meaning yellow. This
vitamin functions in metabolic pathways, including Krebs cycle, where FAD
(flavin adenine dinucleotide) combines with succinate to eventually form
oxaloacetate, the starter compound for the cycle.
The deficiency state regarding riboflavin, known as arboflavinosis, is rare and mild. Symptoms include tissue inflammation and breakdown. The lips may become swollen, and crack at the corners. This condition is known as

![Thiamine Content of Selected Foods](image)

Figure 4. Thiamine Content of Selected Foods (Watt and Merril, 1963).

Cheilosis. Glossitis, in which the tongue becomes swollen and red is common in arboflavinosis, as is corneal vascularization. The corneas of the eye develop extra blood vessels, resulting in itching, burning and tearing. Seborrheic dermatitis may develop, especially in skin folds. These conditions usually occur in those who suffer from malnutrition and are multiply vitamin as well as protein deficient (Whitney et al., 1987).
Food sources of riboflavin include milk (lactoflavin), organ meats, whole grains, and vegetables. This vitamin is especially sensitive to light and water soluble, so cooking riboflavin-rich foods in open pans with excess water will reduce riboflavin content considerably. Riboflavin contents of selected foods are given in figure 5.

**Rapid Analysis of Thiamine and Riboflavin**

Chromatography is a means of separating a compound into its components. In high performance liquid chromatography, pressure is applied to the column by a pump, forcing the mobile phase through at a much higher rate. This pressure speeds the sample through the column, increasing accuracy and decreasing retention times. The use of reversed-phase high-performance liquid chromatography (RPHPLC), has been widely used in biological and vitamin analyses (Robinson, 1987).

Toma and Tabekhia (1979), used RPHPLC in the analysis of B-vitamins in rice products, including thiamine and riboflavin. Products analyzed were paddy rice, brown rice, and enriched rice. Sample preparation of RPHPLC samples was found to be simple and rapid. Both thiamine and riboflavin could be detected in 20 minutes or less. The investigators also compared the accuracy and speed of RPHPLC and wet chemistry methods, finding that
RPHPLC methods provided accuracy with rapidity in the analysis of water soluble vitamins. Kamman et al. (1980) used the method to compare ultraviolet (UV) and fluorescence (FL) detection methods of thiamine and riboflavin in enriched and fortified foods. The investigators found UV detection suitable for both vitamins as their absorbance wavelength maxima is near 254 nm. With both thiamine and riboflavin being detected under 8 minutes, RPHPLC was found to match the accuracy of AOAC methods and exceeded them for speed and ease of sample preparation.

Chase et al. (1980) tested infant formulas for thiamine, riboflavin, and pyridoxine and found simultaneous RPHPLC analysis to be fast, economical, and simple for the determination of water soluble vitamins. These investigators
also used UV detection for both thiamine and riboflavin determination with good results. Both vitamins were detected in under 15 minutes. Chase et al. (1993) later modified their method for thiamine, riboflavin, and pyridoxine, favoring FL detection for improvement of sensitivity and specificity. The researchers converted thiamine to thiochrome with sodium hydroxide and potassium ferricyanide for FL detection. Elution times of under 26 minutes for all three vitamins were reported. Advantages over the earlier method include smaller sample sizes, increased dilutions, and lower detection limits.

Fellman et al. (1982) analyzed selected foods such as liver, whole wheat flour, and milk for thiamine and riboflavin using simultaneous HPLC methods with both UV and FL detection. Both vitamins eluted in less than 10 minutes, with accuracies comparable with AOAC methods.

Sims and Shoemaker (1993) also used simultaneous RPHPLC methods to determine thiamine and riboflavin in selected foods such as whole grain cereals and wheat flour. The researchers found FL detection to be superior, as riboflavin fluoresces, and thiamine can easily be converted to thiochrome, a fluorescent compound. This methods was reported to rival the accuracy of accepted AOAC methods for thiamine and riboflavin detection, and exceeded it for speed and simplicity of sample preparation.
Riboflavin and lumichrome were measured using RPHPLC and FL detection in enriched macaroni by Woodcock et al. (1982). Both compounds eluted in under 10 minutes. Advantages to this method were reported to be simplicity, elimination of pH adjustments, and elimination of blank determinations.

The use of RPHPLC for the determination of thiamine, riboflavin, and other water-soluble vitamins is well documented and is widely reported to offer several advantages over microbiological and wet chemistry methods. Speed, simplicity, accuracy, and economy are cited as the most important benefits of HPLC analysis, reflecting advances in chemical determination of thiamine and riboflavin.

The major goal of this study, conducted as the final phase of a three-part project, was to assess the overall sensory and vitamin quality of irradiated brown rice, as affected by storage. Specific objectives included:

1. To express brown rice flavor in terms of specific attributes, and to assess sensory changes in these terms.
2. To identify storage, irradiation treatment, and varietal effects on flavor attributes of brown rice.
3. To assess the effect of storage, irradiation treatment, and variety on thiamine and riboflavin levels of brown rice.

Two previous studies were conducted at Louisiana State University to determine the effects of gamma-irradiation, variety, and storage on the physicochemical properties, free fatty acid levels and lipase activity of brown rice (Sabularse, 1988; Ransibrahmanakul, 1991). Sabularse found that irradiation of brown rice reduced cooking time and water uptake. Amylose, moisture, fat and protein content were not found to be affected by irradiation or storage. The color of the rice, however, darkened with irradiation and became lighter with storage. Fat acidity and thiobarbituric acid values (TBA), were lower in irradiated rice samples than in non-irradiated ones. A varietal effect was also observed, with Lemont samples being more stable than Mars samples.

Ransibrahmanakul (1991) observed a reduction in lipase activity of irradiated brown rice samples. This reduction was thought to cause the lowered levels of free fatty acids in irradiated samples. Storage was reported to have no effect on free fatty acid level nor lipase activity of non-irradiated brown rice samples. The current research was conducted to complete the rice
irradiation project at Louisiana State University, and used irradiation levels, storage conditions and rice varieties determined by project originators.
MATERIALS AND METHODS

SAMPLE HANDLING

Rough rice samples of two varieties were obtained from the Louisiana Experiment Station at Crowley, Louisiana from the 1995 harvest. Lemont, a long grain variety, and Mars, a medium grain variety, were each separated into 3 equal 592g portions, sealed in plastic bags (Koch Supplies, Inc., Kansas City, MO), and placed in -20°C storage prior to dehulling. Rough rice was prepared for analysis at the Rice processing Laboratory, Department of Biological and Agricultural Engineering, Louisiana State University, and repackaged. One package of brown rice samples was separated, repackaged, and irradiated at 0, 1, and 2 kGys at a rate of 0.98 kGy/hr, in a Cobalt-60 (60Co) irradiation facility at the LSU Nuclear Science Center. Irradiation levels were chosen on the basis of previous studies at LSU by Sabularse (1988). Treated and control samples were then separated into 6 equal portions and sealed in plastic bags for storage at ambient temperature (24°C). Nonirradiated brown rice was used as the control. Monthly samples were analyzed for sensory characteristics, as well as thiamine and riboflavin content over a 6-month period of time. The entire procedure for both varieties was conducted for 3 complete replications.
Sample Preparation

Rice samples of each variety were dehulled with a Satake Rice Machine No. 18356 (Satake Engineering Co. Ltd., Tokyo, Hiroshima), thereby producing brown rice. Broken rice was separated mechanically with a rice sizing machine, which consists of grading screens with an attached shaking system. This procedure, which allows whole rice grains to flow over the screens while trapping brokens was repeated up to 4 times to remove a sufficient portion of broken rice. Remaining rice was hand-graded to remove remaining broken, cracked, unhulled, chalky, or discolored kernels. This procedure yielded whole brown rice grains suitable for analysis.

Lemont rice was separated, which averaged 13.9% moisture, into 3 equal portions for irradiation at 0, 1, and 2 kGy, respectively. After irradiation, the treated samples were divided into 7 aliquots per treatment level and sealed in polyethylene bags for a storage period of 6 months. A portion of the first aliquot of each treatment level was analyzed for sensory attributes to establish a baseline, and the remainder was stored at -20°C until vitamin analysis could be accomplished. The other 6 aliquots were stored at ambient temperature, pending monthly sampling. This procedure was repeated for the Mars variety.
samples, which averaged 13.2% moisture content. The entire procedure was replicated a total of three times.

SENSORY ANALYSIS

Panel Training

Prior to sensory analysis of rice samples, panelists were given fresh, nonirradiated brown rice samples to evaluate for the purpose of defining appropriate descriptive terms for attribute analysis. Ten potential panelists participated in choosing descriptors and the foods intended to define them. The list of possible descriptors and foods is listed in Table 2.

Table 2. Sensory Descriptors and Their Representative Foods.

<table>
<thead>
<tr>
<th>DESCRIPTOR</th>
<th>REPRESENTATIVE FOOD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dairy/Milky</td>
<td>4% Milk, Butter</td>
</tr>
<tr>
<td>Grain</td>
<td>Whole Wheat Bread</td>
</tr>
<tr>
<td>Nutty</td>
<td>Almonds, Pecans</td>
</tr>
<tr>
<td>Green</td>
<td>Celery, Bell Pepper</td>
</tr>
<tr>
<td>Rancidity/Soapy/Cardboard</td>
<td>Rancid Br. Rice, Rancid Rice Bran</td>
</tr>
<tr>
<td>Starch</td>
<td>Crustless White Bread</td>
</tr>
<tr>
<td>Sweetness</td>
<td>Granulated Sugar</td>
</tr>
<tr>
<td>Water/Metallic</td>
<td>Tap Water</td>
</tr>
</tbody>
</table>

From the above descriptors, 4 were chosen as the most appropriate attributes to represent brown rice flavor. Each panelist was provided a
determine frequency of consumption of brown and polished rice. The final panel consisted of 8 panelists chosen from the original 10.

Table 3 shows the final descriptive terms agreed upon by the panel of 8, and the foods chosen to represent the flavor attributes of interest.

Table 3. Final Descriptors and Representative Foods.

<table>
<thead>
<tr>
<th>DESCRIPTOR</th>
<th>REPRESENTATIVE FOOD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Milky</td>
<td>4% Milk</td>
</tr>
<tr>
<td>Nutty</td>
<td>Almonds</td>
</tr>
<tr>
<td>Rancidity/Cardboard</td>
<td>Rancid Rice Bran</td>
</tr>
<tr>
<td>Water/Metallic</td>
<td>Water</td>
</tr>
</tbody>
</table>

After comparing the brown rice to all representative foods, no "green" notes were detected, "starchy" and "grainy" were determined to be inappropriate because the panelists interpreted their meaning in a negative way, and "dairy" was determined to be too broad a term to be useful. The "milky" descriptor was thought to more adequately describe the creamy starchiness of brown rice. "Water/metallic" notes were detected in the fresh brown rice provided, and chosen as an appropriate descriptor. Almonds were determined to best represent the unique nuttiness brown rice gives as a first impression, with the "sweetness" attribute becoming stronger as chewing progresses.
Further training sessions were conducted in order to reinforce the descriptors with their representative foods. Fresh and rancid brown rice were provided for comparison and identification of descriptive notes in the rice. A sensory score sheet for irradiated brown rice is shown in Figure 6.

Sample Preparation

A 20-gram aliquot was taken from each rice sample to be analyzed for sensory attributes. The method used by Sabularse (1988) was used with the following modifications: the samples were each placed in a flat-topped beaker to which 40 mL of water was added. A watch glass cover was placed on top to limit evaporation. The beakers were then loaded into a perforated metal basket and lowered into a boiling water bath. Samples were held in the boiling water bath, sealed with a lid, for 45 minutes. The samples were then removed by lifting the basket. Samples were allowed to stand 5 minutes in the beakers before being emptied into glass bowls for cooling. Samples were allowed to cool for 5 more minutes for further cooling and firming. Rice was then distributed among sample cups, which were coded with random numbers for sample identification, and capped. Duplicate samples were provided for each panel member for the 0, 1, and 2 kGy treatment levels for one rice variety per sensory panel. Sensory analysis was conducted twice weekly for a period of
6 months. This allowed panelists to sample both varieties in the same week. Panelists were instructed to rinse and expectorate between samples.

RAPID ANALYSIS OF THIAMINE AND RIBOFLAVIN

Analytical System

The liquid chromatograph for riboflavin analysis was a Waters Associates (Milford, MA) M-45 pump, a 715 Ultra WISP injector and a 470 scanning fluorescence detector with excitation at 370 nm, and emission at 520 nm. A Rainin Instrument Co. (Woburn, MA) C-18, 5 μm, 15 cm x 4.6 mm i.d. column was used. For thiamine analysis, a Waters Associates UV detector set at 254 nm was used. Detection levels of thiamine and riboflavin are 0.5 ng. and 1.0 ng., respectively. A mobile phase flow rate of 1.5 mL/minute was used.

Standard Preparation

Originally, thiamine and riboflavin standards were combined in the following manner. Thiamine and riboflavin were each added to a tared 100 mL volumetric flask in 1 gram amounts and brought to volume with 0.1 N hydrochloric acid. This resulted in a stock solution of 20 mg/mL, 10 mg each of thiamine and riboflavin. One milliliter of this solution was then added to a 250 mL volumetric flask, which was then brought to volume with distilled
Rate the product using the scales below. Indicate hedonic rating circling the descriptive term which best suits your impression.

**Water/Metallic:**

I---------------------------------------------------I
none

**Nutty:**

I---------------------------------------------------I
none

**Milky:**

I---------------------------------------------------I
none

**Rancidity/Soapy**

I---------------------------------------------------I
none

**Hedonic:** Circle One

- Like extremely
- Like very much
- Like moderately
- Like slightly
- Neither like nor dislike
- Dislike slightly
- Dislike moderately
- Dislike very much
- Dislike extremely

Figure 6. Sensory Scoresheet for Brown Rice.
water. This yielded an intermediate solution of 8 mg/mL, or 4 mg of thiamine and riboflavin, respectively. This solution was then used to make standard dilutions for preliminary injections in order to achieve the proper mobile phase mixture. The standard solution outlined above was found to be inferior, both in method of preparation, and in using thiamine and riboflavin as a combined standard. There were no repeatable results using the combined standard, therefore, the standard solutions were prepared separately in order to achieve repeatability.

The individual standards were prepared according to the method of Simms and Shoemaker (1993), with the exception that the standards were mixed separately and not derivatized. For thiamine stock solution, 20 mg of powdered standard (Sigma, St. Louis, MO) were weighed into a tared volumetric flask and brought to volume with 0.1 N hydrochloric acid, yielding a 0.1 mg/mL stock solution. For preparation of the intermediate solution, 10 mL of the stock solution was pipetted into a 100 mL volumetric flask and diluted to volume with distilled water to produce a 10 μg/mL solution. From this solution, final standard dilutions of 0.8 μg/mL, 0.6 μg/mL, 0.4 μg/mL, and 0.2 μg/mL, were made by pipetting 8, 6, 4, and 2 mL of the working standard into 4-100 mL volumetric flasks, respectively. After bringing them to volume
with distilled water, the standard dilutions were ready for injection. The same
procedure was used for the preparation of riboflavin standards. The separated
standard solutions gave much more reliable results. All standard preparations
were carried out in subdued light in order to protect the vitamins.

**Mobile Phase**

The mobile phase used by Woodcock et al. (1982) was modified for
HPLC determination of thiamine and riboflavin. Their mobile phase
formulation consisted of 56% distilled water, 43% methanol (Mallinckrodt),
and 1% glacial acetic acid (Fisher Scientific). This formulation was used in the
first standard injections and was found to produce unsatisfactory separation of
the two vitamins. The next attempted mobile phase formulation consisted of
40% distilled water, 59% methanol, and 1% glacial acetic acid. This produced
essentially the same results. Other mixtures consisted of 49:50:1, 60:39:1, and
70:39:1. The most successful formulation was found to consist of 80% distilled
water, 19% methanol, and 1% glacial acetic acid. This formulation allowed
detection and separation of desired peaks, as well as being cost-effective.

**Sample Preparation**

The method described by Kamman et al. (1980) was used to prepare
sample extractions with the following modifications. Samples were diluted
with 25 mL of hydrochloric acid, Tween 20 was added to minimize foaming, and centrifugation was accomplished in 5 minutes at 4000 rpm. Individual rice samples were ground to a 40-mesh particle size. These samples were then accurately weighed into separate centrifuge tubes in 5 gram portions and each diluted with 25 mL of 0.1 N hydrochloric acid. Approximately 0.5 mL of Tween 20 was added to each tube to retard foaming. The centrifuge tube was plugged lightly with cotton, which was held in place with autoclave tape. Samples were then autoclaved for 15 minutes at 15 psi for extraction of thiamine and riboflavin. After cooling, the samples were buffered to pH 6 with 1 mL of sodium acetate buffer added to each tube. The samples were then centrifuged at 4000 rpm for 5 minutes. The clear supernatant liquid was drawn up in a plastic syringe and filtered into an amber autosampler vial with septate snap-cap (Alltech) through a 0.20μm Anotop filter. Ten μL aliquots were used for all injections. All operations were carried out in subdued light.

STATISTICAL ANALYSIS

The general linear model (GLM) was used to evaluate all data for a completely randomized design using Statistical Analysis System Software (SAS Institute, Cary, N.C.). Duncan’s Multiple Range Test was used for pairwise comparison of means at the 5% alpha level.
RESULTS AND DISCUSSION

SENSORY ANALYSIS

The selected sensory attributes of irradiated brown rice for Lemont and Mars varieties are presented in graphical form in Figures 7 and 8, respectively. Cooked rice flavor has previously been evaluated as overall taste and aroma (Juliano, 1982). The sensory attributes selected represent components of brown rice flavor, rather than simply an overall taste, as perceived by an experienced panel. These components, and several others, contribute to the complex of what is brown rice flavor. The relative change in each attribute with respect to storage and treatment may be useful in optimizing processing, stabilization and storage parameters of brown rice. All statistical means represented in the following figures are presented in Appendix F and G.

Water/Metallic Attribute

As shown, the overall effect of irradiation treatment with storage was a slight increase in the water/metallic attribute. This effect was observed in the Lemont as well as Mars variety. This attribute was perceived by some to be a defect in brown rice flavor, even though it was present in untreated samples which were dehulled 24 hours prior to sensory analysis. It is important to note, however, that the water/metallic attribute was not universally reviled. Some
Figure 7. Selected Sensory Attributes of Irradiated Brown Rice (Lemont variety).
Scale: 15 centimeters, anchored at 0 and 15. 0=none; 15=very.

Figure 8. Selected Sensory Attributes of Irradiated Brown Rice (Mars variety).
panelists reportedly enjoyed this flavor note and preferred a stronger water/metallic flavor. Most, however, considered it merely a component of fresh brown rice flavor, since initial water/metallic scores were quite high at the 0 kGy, 0-month level and remained relatively stable throughout the study.

Radiation treatment did slightly increase the water/metallic attribute, especially at the 2 kGy level (p-value 0.02) for Lemont variety samples. While only very marginally significant, this may arise from oxidative products resulting from irradiation treatment, or natural oxidation of the rice bran oil fraction. This effect may also be due to other flavor notes not identified among the attributes of interest. The rancidity attribute, becoming stronger with treatment levels, may be a contributor to the metallic note. Overall, however, no significant varietal effect on the water/metallic attribute was observed (p-value 0.2). This attribute of brown rice was shown to be relatively stable to storage and irradiation treatment.

There were no combination effects observed for the water/metallic attribute. This statistical information, arranged in graphic form, is presented in Appendix A.
**Nutt**y **Attribute**

Sensory panelists universally considered the nutty attribute to be pleasant in the fresh, 0 kGy samples. For both Lemont and Mars rice varieties, the storage effect was significant in the reduction of the nutty attribute, as shown in Figure 9 (p-value 0.0082). The oxidation of free fatty acids may contribute to this effect, directly reducing nuttiness scores, while increasing rancidity scores. Furthermore, an increase in free fatty acid content may also be a contributing factor to the reduction of the nutty attribute. Sharp and Timme (1986) report that free fatty acid content of brown rice (untreated) stored for 6 months can be as high as 25%. Although this is reportedly linked to a soapy taste, it may be sufficient to mask or directly reduce the nutty attribute (Barnes and Gallard, 1991). Varietal effect on nuttiness of irradiated brown rice samples was not found to be significant (p-value 0.3). Irradiation was also not found to be a significant contributor to the reduction of nutty attribute (p-value 0.2). This was surprising, considering the oxidative effects radiation treatment may produce. The gradual reduction in nutty scores may suggest the development of secondary oxidation products, such as aldehydes, carbonyl compounds, and ketones, which are reported to contribute to off-flavors in brown rice.
Figure 9. Storage Effect (0 to 6 months) on Nutty Attribute of Irradiated Brown Rice by Radiation Level.
The nutty attribute, represented in panel training by sliced almonds, may originate from rice bran components, including the oils, which are subject to hydrolysis and oxidation. Reduction in nuttiness scores with increasing storage time has no equivalent in the literature, but is generally in agreement with other reports of reduced quality in stored rice (Barber, 1972; Yasumatsu et al., 1966). This natural degradation of eating quality over time was largely unaffected by radiation level. Since sensory quality of brown rice is known to diminish with storage, this finding may lend some clarification as to which components of rice flavor are adversely affected over time, and if they are stable to radiation treatment.

Combination effects were not observed for the nutty attribute. This statistical information is presented in Appendix B.

Milky Attribute

The milky attribute of brown rice was universally perceived as a positive aspect of brown rice flavor. Milky attribute of both Lemont and Mars rice samples was strongly affected by storage. Months of storage, as shown in Figure 10, exerted a significant negative effect on milkiness attribute. With a p-value of 0.0001, storage time may be the most important parameter of consideration with regard to milky attribute. A varietal effect was noted,
Figure 10. Storage Effect (0 to 6 months) on Milky Attribute of Irradiated Brown Rice by Radiation Level.
however, it was marginal (p-value 0.03). Radiation treatment had no effect on milky attribute. The reduction in milky attribute may be associated with an increase in rancidity arising from secondary oxidation products, as mentioned above. An increase in rancidity may simply mask the milky attribute, which was among the most subtle attributes described by the panelists. If the milky attribute is related to the starch content of brown rice, it may remain unchanged by radiation treatment, as suggested by the lack of radiation effect.

With storage time being the strongest contributing factor identified with reduction in milky attribute, free fatty acid formation and subsequent oxidation may produce quite pronounced off-flavors, effectively obscuring the milky attribute of brown rice. The subtle milky flavor may be acted directly upon, or simply overpowered by rancid off-flavors.

Milky attribute, with respect to brown rice, has not been reported in the literature. However, the observed reduction in this component of brown rice flavor agrees with previously cited findings which describe sensory problems arising from storage. Since rice bran contains large amounts of lipid material, as well as the enzymes to attack it, the deterioration of quality upon storage of rice has been widely reported (Juliano, 1985).
Combination effects were not observed for milky attribute. This statistical information is presented in Appendix C.

**Rancidity Attribute**

Rancidity, one of the major impediments to shelf life of brown rice, was increased in both Lemont and Mars rice varieties. Interestingly, this attribute was perceived in fresh, untreated samples. Storage was not a significant factor in rancidity, as interpreted by sensory panelists, when radiation was involved. The p-value for storage was 0.5, which does not indicate significance. Radiation treatment may influence rancidity attribute in brown rice to such an extent that storage, even for an extended period of time, does not contribute appreciably to poor eating quality of the rice. Radiation treatment, however, did significantly affect rancidity values of brown rice. Figure 11 shows an increasing rancidity value with increasing treatment levels. With a p-value of 0.005, radiation level was found to exert a negative effect on the sensory quality of brown rice with respect to rancidity. The relationship of radiation level to rancidity was shown to be linear (p-value 0.001).

Radiation was shown to be the strongest contributing factor to rancidity in brown rice, as it was perceived by the panel. Storage time was completely negligible as far as rancidity is concerned when brown rice samples were
Figure 11. Radiation Effect on Rancidity Attribute of Irradiated Brown Rice by Variety.
irradiated. This finding is in agreement with other workers who have identified irradiation as a contributor to off-flavors through the formation of compounds such as alcohols, ketones, and lactones (Urbain, 1986).

Combination effects were not observed for rancidity attribute. This statistical information is presented Appendix D.

**Hedonic Rating**

Hedonic ratings, which increased as eating quality decreased, showed an increasing trend in both rice varieties. Storage time was shown to be a factor in poor hedonic ratings (p-value 0.0008). This finding is widely supported by previously cited authors. As stated above, secondary oxidation products arising from free fatty acids may be strong contributors to rancid off-flavors which compromise sensory quality. Varietal effects were not significant, and showed no definite trend over time, with respect to hedonic rating. The most important effect on hedonic rating was irradiation level. This effect, shown in Figure 12, is strongly negative on hedonic rating of treated brown rice. Radiation treatment had a significant effect on hedonic rating at the 2 kGy level. The effect for the 1 kGy level was not significantly different from control samples. While storage time was a factor in the deterioration of
Figure 12. Radiation Effect on Hedonic Rating of Irradiated Brown Rice by Variety.
eating quality, radiation treatment at the highest treatment level was shown to be the most detrimental.

Hedonic rating reflects an overall impression or judgement of a product, rather than specific components of flavor. While hedonic ratings are independent of individual attributes, they may be helpful when used in conjunction with the attributes as the ultimate indicator of important sensory changes. Storage time is independently detrimental to hedonic rating of irradiated brown rice.

Radiation of brown rice, especially at the 2 kGy treatment level seemed to have the most influence on hedonic rating. The reduction in eating quality of brown rice due to radiation treatment was immediately detected by sensory panel members. Some panel members seemed to be particularly sensitive to the off-flavors induced by radiation treatment of the rice samples. This may simply be due to differences in perception and taste acuity among panelists. However, these findings are consistent with those previously reported.

Length of storage was also a factor in the reduction of brown rice quality. The effect of storage on hedonic rating was independent of radiation treatment and variety, however. Combination effects were not observed for hedonic rating. Supporting statistical information is presented Appendix E.
THIAMINE AND RIBOFLAVIN

Thiamine

Thiamine levels in irradiated brown rice were affected by both radiation treatment and storage. Radiation treatment had the strongest effect at the 2 kGy level for Mars variety brown rice (p-value 0.0001). Thiamine levels in Mars rice at the 1 kGy level was 82%, independent of storage. At the 2 kGy level, 80% of the thiamine was retained. Storage was found to be a significant factor in thiamine loss of brown rice of the Mars variety, independent of irradiation (p-value 0.0046). Still, thiamine retention at the 1 kGy level was found to be 88% at 3 months of storage, and 90% at 6 months of storage of Mars variety brown rice.

Lemont variety rice demonstrated better thiamine retention, with respect to both radiation treatment and storage. Thiamine levels remained unaffected by radiation treatment, even at the 2 kGy treatment level. Storage of brown rice over the 6-month period reduced thiamine content to 97% of fresh samples at 3 months of storage. Thiamine levels were found to stay at 97% over the remaining 3-months of storage. Thiamine results are presented for Lemont and Mars rice samples in Figures 13 and 14, respectively.
Figure 13. Thiamine Content of Irradiated Brown Rice (Lemont variety).
Figure 14. Thiamine Content of Irradiated Brown Rice (Mars variety).
Riboflavin

There was no reduction of riboflavin in brown rice samples of the Lemont variety. No radiation or storage effect was detected (p-value 0.7). However, riboflavin levels in Mars brown rice samples showed a significant negative effect with respect to storage and radiation treatment. After 3 months of storage, riboflavin levels in Mars brown rice samples were 84% of fresh samples. This level was reduced to 49% retention after 6 months of storage. Storage was found to be marginally significant (P-value 0.02). Radiation treatment was found to be highly significant for riboflavin values of Mars variety rice samples (p-value 0.0004). Riboflavin retention at the 1 kGy level was 100%, however, this percentage fell to 75% at the 2 kGy level. These findings still suggest fair riboflavin retention in irradiated brown rice.

Thiamine and riboflavin retention in irradiated brown rice is fairly good. The lowest retention is that of riboflavin in Mars variety brown rice (49%). Lemont rice, however, retains thiamine at 97% upon storage for 6 months and 100% at the 2 kGy treatment level. The stated losses of thiamine in irradiated brown rice is marginal and is consistent with nutrient losses found in other processing methods. These findings agree with other reported values for irradiated and processed foods (WHO, 1994).
Riboflavin was very stable in Lemont variety brown rice, showing no detectable losses with irradiation treatment or storage time. In fact, increases in riboflavin levels of Lemont variety samples were detected. This effect has been previously reported in the literature (Barone, 1965, Brooke et al., 1964). This effect has been explained as the enhanced extraction of bound riboflavin resulting from irradiation. Riboflavin reportedly exhibits good retention with respect to irradiation treatment in food matrices (WHO, 1994). Riboflavin was not retained to the same extent in Mars variety brown rice. Moreover, with 75% retention at the 2 kGy treatment level, irrespective of storage, riboflavin displayed more radiosensitivity than thiamine. These findings are also in agreement with the WHO findings previously mentioned.

Storage seemed to have a slightly more detrimental effect with respect to Mars brown rice, with 84% of original riboflavin levels remaining at 3 months and 49% remaining at 6 months of storage. WHO (1994) suggests specific variables affecting vitamin retention such as variety, radiation dose, temperature, hydration, nutrient in question, storage conditions, and food matrix. Some of these factors have been shown to affect thiamine and riboflavin retention of rice samples in this study, which is in agreement with the cited literature.
Thiamine analysis by RPHPLC was difficult in that repeatable results proved troublesome to achieve. The vitamin standard solutions are rather delicate, requiring subdued light, and refrigeration. Even with these precautions, fresh powdered standards had to be obtained during the study, as the original standard was unreliable at delivery. Verbal consultations with other researchers involved in thiamine analysis via RPHPLC confirmed the difficulty of thiamine analysis using this method, which were independent of mobile phase, equipment, and food matrix. These difficulties are not widely reported in the literature, which portrays RPHPLC analysis of thiamine to be a straightforward and simplified method. It was not found to be so in this study, nor by others currently involved similar work. Figures 15 and 16 show riboflavin results for Lernont and Mars brown rice, respectively.
Figure 15. Riboflavin Content of Irradiated Brown Rice (Lemont variety).
Figure 16. Riboflavin Content of Irradiated Brown Rice (Mars variety).
CONCLUSIONS

The research on thiamine, riboflavin, and storage stability of irradiated brown rice was undertaken in an attempt to develop better stabilization techniques while protecting sensory quality and thiamine and riboflavin content. Sensory attributes which contribute to brown rice flavor were identified, and studies were conducted in order to assess changes resulting from irradiation treatment and storage. Studies were also conducted to determine thiamine and riboflavin content of irradiated brown rice samples and how they may change with storage. Two Louisiana rice varieties were used: Mars, a medium grain variety, and Lemont, a long grain variety.

Three phases of the project were included in this research. The first phase involved the determination of appropriate descriptive terms to describe the flavor of fresh, untreated brown rice samples of both Lemont and Mars varieties. Training the panel in attribute recognition and scaling was accomplished through the use of food identifiers. Almonds identified the nutty attribute, tap water the water/metallic attribute, whole milk identified the milky attribute, and rancid rice bran was used for identification of rancidity.

In the second phase, sensory analysis techniques involving scaled attributes were used to define initial quality of fresh rice samples. Sensory
quality of samples at the 0 kGy, 1 kGy, and 2 kGy treatment levels were continued at monthly intervals over a storage period of 6 months to assess the effect of variety, treatment level, and storage time on individual aspects of brown rice flavor, as well as overall hedonic rating. Irradiation was found to decrease eating quality, with hedonic ratings increasing with radiation levels. Irradiation was also important to increased perception of rancidity of brown rice samples. Storage was found to be significant in the reduction of milky attribute, with scores decreasing as storage time increased. The nutty attribute was also negatively affected by storage, decreasing with increasing storage time. No effect was observed for either irradiation or storage on the water/metallic attribute.

In the third phase of the project, effects on thiamine and riboflavin of irradiated brown rice were determined to investigate the changes irradiation and storage may have on rice samples. Thiamine seemed to exhibit more radiosensitivity than riboflavin, especially in the Mars variety. These results seem to indicate a slight varietal effect, which was also seen with riboflavin levels, though not to the same extent.

Storage was also a factor in the reduction of thiamine and riboflavin in the Mars variety. Riboflavin was shown to be less stable in Mars rice samples,
though completely stable to irradiation and storage in Lemont variety rice. No interactions were noted for thiamine or riboflavin in either rice variety.

Further work is necessary to optimize irradiation conditions for brown rice processing, as this method is effective in deinfestation of cereal grains. Modified atmospheres and alternative packaging materials may provide better sensory stability of irradiated brown rice. Some elucidation as to variability of results with respect to varietal response to irradiation would be of interest. Further information as to the cause and control of lipid autoxidation and hydrolysis would also be of interest and may eventually lead to the development of shelf-stable brown rice.

This research marks the culmination of a project started by Sabularse in 1988 in which the physicochemical effects of radiation treatment on brown rice were defined. Ransibrahmanakul (1991) continued the work, studying radiation’s effect on lipase activity and free fatty acid content of brown rice. Both of these researchers noted the need for further work, defining sensory as well as B-vitamin stability of irradiated brown rice, which has been the subject of this body of work.
REFERENCES


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APPENDICES
Appendix A.1. Varietal Effect on Water/Metallic Attribute of Irradiated Brown Rice by Months of Storage (0-6).
Appendix A.2. Storage Effect (0 to 6 months) on Water/Metallic Attribute of Irradiated Brown Rice by Radiation Level.
Appendix B.2. Nutty Attribute of Irradiated Brown Rice (Mars variety).
Appendix B.3. Varietal Effect on Nutty Attribute of Irradiated Brown Rice by Months of Storage (0-6).
Appendix C.1. Milky Attribute of Irradiated Brown Rice (Lemont variety).

Scale: 15 centimeters, anchored at 0 and 15. 0=None; 15=very.
Appendix D.1. Rancidity Attribute of Irradiated Brown Rice (Lemont variety).
Appendix D.2. Rancidity Attribute of Irradiated Brown Rice (Mars variety).

Scale: 15 centimeters, anchored at 0 and 15. 0=none; 15=very.
Appendix D. 3. Storage Effect (0 to 6 months) on Rancidity Attribute of Irradiated Brown Rice by Radiation Level.
Appendix D. 4. Varietal Effect on Rancidity Attribute of Irradiated Brown Rice by Months of Storage (0-6).
Appendix E.1. Hedonic Rating of Irradiated Brown Rice (Lemont variety).
Appendix E.2. Hedonic Rating of Irradiated Brown Rice (Mars variety).
Appendix E.3. Storage Effect (0 to 6 months) on Hedonic Rating of Irradiated Brown Rice by Radiation Level
Appendix E. 4. Varietal Effect on Hedonic Rating of Irradiated Brown Rice by Months of Storage (0-6)
APPENDIX F: STATISTICAL DATA FOR FIGURES 7 THROUGH 16.

Figure 7. Selected Sensory Attributes of Irradiated Brown Rice (Lemont variety).

<table>
<thead>
<tr>
<th></th>
<th>W/MET</th>
<th>NUTTY</th>
<th>MILKY</th>
<th>RANCID</th>
<th>HEDON</th>
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<tr>
<td>0kGy</td>
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<td>1kGy</td>
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<td>2kGy</td>
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Figure 8. Selected Sensory Attributes of Irradiated Brown Rice (Mars variety).

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<thead>
<tr>
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<th>HEDON</th>
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Figure 9. Storage Effect (0 to 6 months) on Nutty Attribute of Irradiated Brown Rice by Radiation Level.

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Figure 10. Storage Effect (0 to 6 months) on Milky Attribute of Irradiated Brown Rice by Radiation Level.

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<td>1 kGy</td>
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<td>2.92</td>
<td>2.69</td>
<td>3.34</td>
<td>2.98</td>
<td>2.18</td>
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Figure 11. Radiation Effect on Rancidity Attribute of Irradiated Brown Rice by Variety.

<table>
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<th>Mars</th>
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<td>3.28</td>
<td>3.36</td>
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<td>1 kGy</td>
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<td>3.49</td>
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<td>2 kGy</td>
<td>4.23</td>
<td>4.14</td>
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Figure 12. Radiation Effect on Hedonic Rating of Irradiated Brown Rice by Variety.

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Figure 13. Mean Thiamine Content (mcg/g) of Irradiated Brown Rice (Lemont variety).

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<td>1 kGy</td>
<td>5.12±0.22</td>
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<td>5.42±0.36</td>
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<td>2 kGy</td>
<td>5.66±0.08</td>
<td>5.73±0.18</td>
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Figure 14. Mean Thiamine Content (mcg/g) of Irradiated Brown Rice (Mars variety).

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<td>2 kGy</td>
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Figure 15. Mean Riboflavin Content (mcg/g) of Irradiated Brown Rice (Lemont variety).

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Figure 16. Mean Riboflavin Content (mcg/g) of Irradiated Brown Rice (Mars variety).

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<td>1.04±0.07</td>
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APPENDIX G: STATISTICAL DATA FOR APPENDIX A THROUGH E.

Appendix A.1. Varietal Effect on Water/Metallic Attribute of Irradiated Brown Rice by Months of Storage (0-6).

<table>
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Appendix A.2. Storage Effect (0 to 6 months) on Water/Metallic Attribute of Irradiated Brown Rice by Radiation Level.

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<td>5.52</td>
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<td>5.45</td>
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<td>5.59</td>
<td>5.22</td>
<td>4.74</td>
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Appendix B.1. Nutty Attribute of Irradiated Brown Rice (Lemont variety).

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Appendix B.2. Nutty Attribute of Irradiated Brown Rice (Mars variety).

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<td>3.82</td>
<td>3.44</td>
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<td>3.31</td>
<td>3.34</td>
<td>3.17</td>
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Appendix B.3. Varietal Effect on Nutty Attribute of Irradiated Brown Rice by Months of Storage (0-6)

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<td>3.39</td>
<td>3.13</td>
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Appendix C.1. Milky Attribute of Irradiated Brown Rice (Lemont variety).

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<td>3.47</td>
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</tr>
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Appendix D.1. Rancidity Attribute of Irradiated Brown Rice (Lemont variety).

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<td>4.44</td>
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Appendix D.2. Rancidity Attribute of Irradiated Brown Rice (Mars variety).

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Appendix D.3. Storage Effect (0 to 6 months) on Rancidity Attribute of Irradiated Brown Rice by Radiation Level.

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<td>3.54</td>
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Appendix D.4. Varietal Effect on Rancidity Attribute of Irradiated Brown Rice by Months of Storage (0-6).

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Appendix E.1. Hedonic Rating of Irradiated Brown Rice (Lemont variety).

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Appendix E.2. Hedonic Rating of Irradiated Brown Rice (Mars variety).

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Appendix E. 3. Storage Effect (0 to 6 months) on Hedonic Rating of Irradiated Brown Rice by Radiation Level.

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Appendix E. 4. Varietal Effects on Hedonic Rating of Irradiated Brown Rice by Months of Storage (0-6)

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<td>5.29</td>
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<td>4.93</td>
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</table>
VITA

Linda Chamberlain Douglas was born in Port Arthur, Texas, on December 25, 1958 to N. Doyle Chamberlain and Virginia Gay Hanson Chamberlain. She lived in Nederland, Texas, until the age of 10, when her family moved to New Iberia, Louisiana. She graduated from New Iberia Senior High School in May, 1977. She entered the University of Southwestern Louisiana in August, 1987, obtaining a bachelor of science degree in Dietetics in December, 1991. She entered graduate studies at Louisiana State University in August 1992, and was granted a master of science degree in Food Science in December, 1994. She is currently a candidate for a doctoral degree in Food Science at Louisiana State University. She is concurrently a dietetic intern through the University of Southwestern Louisiana, completing eligibility requirements for licensure as a Registered Dietitian.

She has been the recipient of the Louisiana State University College of Agriculture Graduate Scholarship, Kiwanis Club Scholarship, and the Food Science Club Scholarship. She was an Honors Convocation Scholar at the University of Southwestern Louisiana, 1989, was a member of Alpha Lambda Delta Freshman Honor Society, and the Gamma Beta Phi Honor Service Society. She is currently a member of the Institute of Food Technologists, the
American Dietetic Association, the LSU Kiwanis Club, and the American Association of University Women.

She is married to William Lane Douglas, III, and they are the parents of two girls, Jennifer Nicole Meyers, and Mary Kathryn Douglas.
DOCTORAL EXAMINATION AND DISSERTATION REPORT

Candidate: Linda Chamberlain Douglas

Major Field: Food Science

Title of Dissertation: Thiamine, Riboflavin, and Sensory Stability of Irradiated Brown Rice

Approved:

[Signatures]

Major Professor and Chairman
Dean of the Graduate School

EXAMINING COMMITTEE:

[Signatures]

Date of Examination: September 27, 1996