EFFECT OF RICE BRAN OIL SPREAD (RBOS) ON THE PHYSICAL AND SENSORY QUALITY OF BAKED PRODUCTS

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B.TECH (Food Technology)

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BY

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THESIS SUBMITTED TO THE PROFESSOR JAYASHANKAR TELANGANA STATE AGRICULTURAL UNIVERSITY IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF

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2016

DECLARATION

I, **RIZWANA SHAIK,** hereby declare that the thesis entitled **"EFFECT OF RICE BRAN OIL SPREAD (RBOS) ON THE PHYSICAL AND SENSORY QUALITY OF BAKED PRODUCTS"** submitted to the **Professor Jayashankar Telangana State Agricultural University** for the degree of **Master of Science in Food Technology** is the result of original research work done by me. I also declare that no material contained in the thesis has been published earlier in any manner.

Place: **(RIZWANA SHAIK)**

Date: **I.D. NO. FST/2014-007**

CERTIFICATE

Mrs. RIZWANA SHAIK has satisfactorily prosecuted the course of research and that thesis entitled **"EFFECT OF RICE BRAN OIL SPREAD (RBOS) ON THE PHYSICAL AND SENSORY QUALITY OF BAKED PRODUCTS"** submitted is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination. I also certify that neither the thesis nor its part thereof has been previously submitted by her for a degree of any university.

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Date:

Chairperson

CERTIFICATE

This is to certify that the thesis entitled **"EFFECT OF RICE BRAN OIL SPREAD (RBOS) ON THE PHYSICAL AND SENSORY QUALITY OF BAKED PRODUCTS"** submitted in partial fulfillment of the requirements for the degree of 'Master of Science in Food Technology' of the Professor Jayashankar Telangana State Agricultural University, Hyderabad is a record of the bonafied original research work carried out by **Mrs. RIZWANA SHAIK** under our guidance and supervision.

No part of the thesis has been submitted by the student for any other degree or diploma. The published part and all assistance received during the course of the investigations have been duly acknowledged by the author of the thesis.

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Date:

Place: Hyderabad

(RIZWANA SHAIK)

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LIST OF SYMBOLS AND ABBREVIATIONS

%	: Per cent
°C	: Degree Celsius
µg	: Microgram
µmol	: Micromole
ANOVA	: Analysis of Variance
CHD	: Coronary Heart Disease
CLA	: Conjugated Linoliec Acid
CLFA	: Cholesterol-Lowering Fatty Acids
CRFA	: Cholesterol-Raising Fatty Acids
CVD	: Cardiovascular Disease
et al.	: and other people
etc.	: and so on; and other people/things
FAO	: Food and Agricultural Organization
FA	: Fatty Acid
FAME	: Fatty Acid Methyl Esters
FDA	: Food and Drug Administration
FFA	: Free Fatty Acid
FTIR	: Fourier Transform Infrared Spectroscopy
g	: Gram
GC	: Gas Chromatography
GC-FID	: Gas Chromatography with Flame Ionization Detector
GLC	: Gas Liquid Chromatography
HF	: Hydrogenated Fat
i.e.	: *id est*; that is; in other words
IIRR	: Indian Institute of Rice Research
I-TFA	: Industrial Trans Fatty Acids
LDL-C	: Low-Density-Lipioprotein Cholesterol
Lp	: Lipoprotein
mcg	: Microgram
Mha	: Million hectare
mg	: Milligram
min	: Minute

:

MUFA	: Mono Unsaturated Fatty Acids
NIN	: National Institute of Nutrition
No.	: Number
PHVO	: Partially Hydrogenated Vegetable Oils
PJTSAU	:Professor Jayashankar Telangana State Agricultural University
PUFA	: Poly Unsaturated Fatty Acids
RBO	: Rice Bran Oil
RBOS	: Rice Bran Oil Spread
SFA	: Saturated Fatty Acids
SFO	: Sun Flower Oil
TFA	: Trans Fatty Acids
UFA	: Unsaturated Fatty Acids
USDA	: United States Department of Agriculture
viz.	: Namely
vs.	: Versus; against
WHO	: World Health Organization

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ABSTRACT

Baked foods serve as one of the main staple food sources for consumers and lipids play an important role in the majority of baked products. When choosing a lipid source for baking, the nutritional characteristics of lipid and the ability of the lipid to impart a desired physical quality to the finished product need to be considered. Recently consumers have become more concerned about the health implications of trans fats which are produced in the baked products due to usage of hydrogenated fat at high baking temperatures. Studies have shown that Rice Bran Oil semisolid Fraction (RBOF) can also be incorporated into baked food formulations with improvement in oxidative stability in baked foods. Therefore, a study was planned to develop and standardize baked products with Rice bran oil and 2 varieties of Rice Bran Oil Spread (RBOS) to evaluate the physico-chemical and sensory properties of the developed products, as well as to determine the consumer acceptability of the best acceptable product. The best accepted product was subjected to scanning electron microscopic study and fatty acid estimation with special reference to trans fatty acids.

Five different baked products namely cakes, cookies, piecrusts, muffins and doughnuts were developed and standardized by using Hydrogenated fat (HF), Rice bran oil (RBO) and two varieties of Rice bran oil spread (RBOS1 and RBOS2). The five standardized products with HF (control) & RBO, RBOS1 and RBOS2 (experimental) were subjected to physico-chemical analysis and sensory evaluation by 15 semi-trained panel members using a 9-point hedonic scale.

The results of physical parameters revealed that the moisture content was highest in HF cake (16.035±0.36) and least in RBOS1 cake (8.924±0.43). The cake density was statistically similar in HF cake (0.330±0.01) and RBOS1 cake (0.280±0.01); RBO cake (0.397±0.01) and RBOS2 cake (0.390±0.01). In cookies the moisture content and cookie spread were highest in RBO cookies, whereas cookie dough density was highest in RBOS1 cookie. In piecrust, moisture, height point, crust height were highest in HF pie crust compared to other pie crusts. The oiliness was least

in RBOS2 (0.063±0.02) pie crust indicating the healthiness of piecrust made with RBOS2. In muffins moisture content was highest in RBOS1 muffin (17.546±0.18) and least in RBOS2 muffin (13.874±0.70), where as the bulk density was highest in RBOS2 muffin (0.423±0.01) and least in RBOS1 muffin (0.257±0.01), indicating an inverse association between the two parameters. In doughnut, the moisture content was least in RBOS2 doughnut (11.197±0.10), whereas the volume was also highest (44.333±0.41) in the same. The specific volume of doughnut was highest in RBOS1 (8.400±0.10) and least in RBO (7.427±0.12) doughnut.

Results of sensory evaluation showed that there is a significance difference among the five products made with HF, RBO, RBOS1 and RBOS2 in the sensory attributes like colour, texture, taste and overall acceptability. According to the results of overall acceptability, it was observed that the cake made with RBOS2 was highly acceptable compared to other products and hence the same product was used to carry out consumer acceptability study, scanning electron microscopic study and fatty acids estimation with special reference to trans fatty acids.

Consumer study was done on a total of 100 subjects in the age group of 18-60 years by using a consumer acceptability questionnaire consisting of two sections describing the demographic profile and the hedonic scoring (5-point hedonic scale) attributes like appearance, color, odour, flavour, texture, taste and overall acceptability of the product. Results of the study revealed that there was no significant difference in the sensory attributes of the cake made with hydrogenated fat and RBOS2. The cake made with RBOS2 was well accepted by the respondents selected for consumer acceptability study.

The scanning electron microscope results indicated that the macro size pores of the RBOS2 cake was much better than the control cake made with HF. The results of SEM are similar with the results of sensory evaluation (textural attributes of cake), indicating that RBOS2 as an ingredient instead of hydrogenated fat can contribute to better texture and loaf volume in cake.

Fatty acid analysis results showed that the total fat content in cake was highest in RBOS1 cake (17.15) and lowest in RBOS2 cake (14.95). The saturated fat content was highest in HF cake (44.55) and lowest in RBO cake (24.08). The total amount of unsaturated fat was highest in RBO cake (72.36) and lowest in HF cake (39.99). Total trans fat content was highest in HF cake (15.46) and lowest in RBOS2 cake (15.46). The TFA content assumes significance in terms of their ill effects on the health of consumers, only if fat content is also high. Consumption of the HF products might prove to be harmful if consumed in large amounts and at higher frequencies due to the presence of trans fatty acids. Though there is presence of trans fatty acids in RBOS2 cake, it does not attain significance due to the quantity present as a whole in comparison to the total fat content. Hence, RBOS2 can be promoted as a healthy alternative to hydrogenated fat for preparation of baked products.

Chapter I

INTRODUCTION

Rice (*Oryza sativa*) belongs to the family Graminecae that includes 20 wild and two cultivated species, viz. Asian rice (*Oryza sativa*) and African rice (*Oryza glaberrima*). *O. sativa* is the most commonly grown species throughout the world today (Oko and Ugwu, 2011). Rice has been considered the best staple food for over three billion people, constituting over half of the world's population (Cantral and Reeves, 2002). Rice has been adapted and consumed by humans for almost 5,000 years (Zhou *et al.*, 2002) and is ranked as world's number one human food crop (Itani *et al.*, 2002). At global level, rice occupies an area of about 153.77Mha which is 30% of the total area under food grain cultivation. India occupies the highest area of 45Mha under rice with a production of 91.0 metric tons next only to china accounting for 46% area under cereals and 42% of total grains (Anonymous, 2007). Rice production in India has witnessed a spectacular increase in the recent past and is approximately one-third of the total rice production of the world (Singh *et al.*, 2005). Rice (*O. sativa*) provides about 75% of the protein in the average daily diet of people (Anonymous, 2002). Over two billion people in Asia alone derive 80% of their energy needs from rice. Rice is the only cereal crop cooked and consumed mainly as whole grain and quality considerations are much more important than for any other food crop (Hossain *et al.*, 2009).

Brown rice grains contain more nutritional components, such as dietary fiber, E and B vitamins and gamma aminobutyric acid (GABA), than ordinary milled rice grains. These bio-functional components exist mainly in the germ and bran layers that are removed by polishing or milling (Champagne *et al.*, 2004). The brownish portion of rice which is taken out in fine grain form, during de-husking and milling of paddy is the rice bran (Hernandez *et al*., 2000). The bran is the hard outer layer of rice consisting of aleurone and pericarp. Rice bran contains an array of micronutrients like oryzanols, tocopherols, tocotrienols, phytosterols, 20% oil, 15% protein, 50% carbohydrate (majorly starch), dietary fibers like beta-glucan, pectin and gums (Jiang *et al*., 2005).

Rice bran, which was earlier used primarily as animal feed, is now finding major application in the form of rice bran oil (Hoed *et al*., 2006). India and Thailand have been the most successful countries in rice bran oil production. In India the solvent extraction process of 40 lakh tons rice bran yields about 6.5 lakh tons of rice bran oil

(Anonymous *et al.*, 2009). Rice bran oil refining industry produces residues such as wax sludge, gum sludge and soap stock that are a rich source of many nutraceuticals like oryzanols, tocopherols, tocotrienols, ferulic acid, phytic acid, lecithin, inositol and wax (Patel and Naik, 2004).

India is the second largest producer of rice in the world and has the high potential to produce rice bran oil, a byproduct of rice milling industry. Since rice bran oil is unconventional oil, its chemical composition, nutrient evaluation and toxicological safety were assessed. Experimental studies have demonstrated the hypolipidaemic effect of rice bran oil. It was established that minor constituents present in the unsaponifiable fraction of rice bran oil were responsible for its hypolipidaemic effect. Nutritional and toxicological studies had shown that there were no abnormalities in animals fed with either rice bran oil or groundnut oil. In view of its safety and hypolipidaemic activity, rice bran oil was considered as an alternative source of edible oil (Raghuram and Rukmani, 1995).

Rice bran oil is extensively used in Japan, Korea, China, Taiwan and Thailand as "Premium Edible Oil". In Japan, Rice Bran Oil is more popularly known as a "Heart Oil". In Western countries, Rice Bran Oil has acquired the status of a "Health Food". The oryzanol present in rice bran is reported to have functions similar to vitamin E in promoting growth, facilitating capillary growth in the skin, and improving blood circulation along with stimulating hormonal secretion (Luh *et al.*, 1991). Rice bran oil is an excellent cooking medium because it is nutritionally superior, contains more micronutrients, longer shelf life, more stable at higher temperature, gives better taste & flavor to food items; frying takes less time, so saves energy and economic due to 15% less absorption of oil during frying (Sharma, 2002).

Rice bran oil has garnered attention from consumers in recent years, owing to its high concentrations of health-promoting compounds, ranging from tocopherols and tocotrienols, phytosterols and γ-oryzanol (Sen *et al.*, 2006). Various distributions of tocopherols are found in vegetable oils and multiple tocopherols contribute to the oxidative stability of these oils (Warner *et al.*, 2008). Tocotrienols also act as antioxidants and display anticancer, cholesterol lowering, and neuroprotective properties that are distinct from tocopherols (Sen *et al.*, 2007). γ-Oryzanol has been shown previously to act as an antioxidant (Miller and Engel, 2006; Juliano *et al.*, 2005) and is also effective at lowering cholesterol (Yokoyama, 2004). Rice bran oil specifically has been shown to protect against lipid peroxidation in vitro (Lee *et al.*,

2005). Many of the same molecules in rice bran oil that impart antioxidant activity in biological tissues also inhibit lipid oxidation on the shelf: rice bran oil is relatively resistant to oxidative degradation at both ambient and elevated temperatures, leading to longer product life of foods made with rice bran oil. While commodity oils have traditionally been used in the manufacture of spreads, rice bran oil itself is now being explored, for use in spreads, as cooking oil and as bakery fat (Eady *et al.*, 2011).

In the baking industry, a number of functions are induced by lipids (Ghotra *et al.*, 2002; Rogers, 2004). They include tenderization, mouthfeel, structural integrity, lubrication, air incorporation, heat transfer and shelf life extension. Because of their functional properties (e.g. creaming ability), plastic shortenings, which are often made by partial hydrogenation, are commonly used in the baking industry (Hernandez, 2007; Zhou *et al.*, 2011). However, trans fat generated during the hydrogenation process increases the risk of coronary heart disease (Dhaka *et al.*, 2011). The United States Department of Agriculture (USDA) and the Food and Drug Administration (FDA) recommend consuming less than 10% of calories from saturated fatty acids by replacing them with monounsaturated and polyunsaturated fatty acids, and to reduce trans fat intake as much as possible (US Department of Agriculture and US Department of Health and Human Services 2010). In recent years, the food industry has been making efforts to reduce trans fat by blending oils (high oleic and low linolenic) with fully hardened oils (palm), or by randomizing through interesterification (Wassell and Young, 2007; Jeyarani *et al.*, 2009; Sahri and Dian, 2011; Musavi *et al.*, 2011). However, because of its supreme characters such as plasticity, which leads to desired physical qualities of baked products, lipids containing trans fat are still widely used in food industry (Jeyarani *et al.*, 2009).

Recently, consumers have become more concerned about the health implications of Trans Fatty Acids (TFA) consumption. Ascherio and Willet (1997), suggested that TFA intake can be reduced by lowering total fat intake. However, there is a concern that, this will lead to an insufficient intake of essential fatty acids (n-3 and n-6). In 2006, the FDA issued the final rule on TFA labeling, requiring companies to declare the content of TFA (0.5g or more per serving) on the nutrition facts panel of food products in a separate line below SFA. Industry has responded by finding alternative ways to minimize TFA content in food products. Current process approaches include: genetic modification of edible oil seeds (Tarragotrani *et al.*, 2006), modification of the hydrogenation process and interesterification. Other TFA alternatives include: the use of

fractions high in solids from natural oils (i.e., coconut, palm kernel oils) and the use of blended oils (Wassell and Young, 2007) for production of baked food products with less trans fats.

Baked foods serve as one of the main staple food sources for consumers and lipids play an important role in the majority of baked products. When choosing a lipid source for baking, the nutritional characteristics of lipid and the ability of the lipid to impart a desired physical quality to the finished product need to be considered. Usually, animal fats, such as lard, are more saturated than plant oils such as soybean oil and their polyunsaturated fatty acid (PUFA) vs. Saturated fatty acids ratio is much lower (Rogers, 2004). In addition to the nutritional profiles, lipids should be selected according to their specific performance in the finished product (Wainwright, 1999). In general, lipids in baking contribute to products quality characteristics such as tenderness, moist mouthfeel, lubricity, flavor, structure and shelf life. During processing, lipids affect air incorporation, lubrication and heat exchange in the baked products (Ghotra *et al*., 2002).

Rice bran oil semisolid fraction (RBOF) can also be incorporated into food formulations (Bakota *et al.,* 2014). Lipid ingredients serve a number of essential functions in baked goods, including acting as a moisture barrier, aiding air incorporation, contributing to the desired structure of the product matrix, providing lubrication for the product in the mouth and enhancing palatability. Lipid properties also contribute to the shelf life of the finished product. In the past, commercially prepared baked goods have relied on the use of hydrogenated or partially hydrogenated oils to provide the desired product attributes (Cheong *et al.,* 2011). A study by Bakota *et al.* (2014) on usage of rice bran oil derived spread as an antioxidant source in frying oils and as fat replacer in baked goods reported that, use of rice bran oil spread imparted oxidative stability to the oil and when incorporated into two baked goods, there was consistently high acceptability ratings for the baked goods tested.

Hence a study was taken up to understand the effect of hydrogenated fat versus rice bran oil and rice bran oil spreads on various baked products, their sensory acceptability and consumer acceptability. For different physical qualities of baked products, extensive literature can be found for cake (Berglund and Hertsgaard, 1986; NorAini *et al*., 1992; Dogan *et al*., 2007; Rutkowska and Zbikowska, 2010; Zhou *et al*., 2011), cookies (Berglund and Hertsgaard, 1986; Jacob and Leelavathi, 2007) and pie crusts (Matthews and Dawson, 1963; Darweesh *et al*., 1991) made with various lipid sources. Very little information is available that correlates the physicochemical properties of lipids with the

sensory qualities of baked products and their final consumer acceptance. Consequently, understanding the physicochemical properties of lipid that in turn influence the sensory properties of baked products and the relationship to their consumer acceptance is very important to both food scientists and food producers. However, the application is worthless to industry without consumers approval. Although consumers desire healthier alternatives, they are unlikely to accept large sensory changes in food products. Therefore, consumer sensory studies are needed to evaluate differences in consumer acceptance between baked products made with rice bran oil spread. Taking the above criteria into consideration a study titled "Effect of Rice Bran Oil Spread (RBOS) on the physical and sensory quality of baked products" was planned with the following objectives:

1. To develop and standardize baked products with RBOS.
2. To perform sensory evaluation studies on the developed products and select best acceptable product.
3. To study the physical quality parameters of developed baked products.
4. To study the consumer acceptance of the selected product made with RBOS

Chapter II

REVIEW OF LITERATURE

The study "Effect of Rice bran oil spread (RBOS) on the physical and sensory quality of baked products" was conducted in the Department of Foods and Nutrition, Post Graduate and Research Centre, Professor Jayashankar Telangana State Agricultural University, Rajendranagar, Hyderabad. Different research workers have carried out studies on lipids in human diet, importance and consumption of baked products, hydrogenated fats and their application in baked products and health benefits of rice bran oil. Literature available relevant to present study has been reviewed under the following headings.

2.1 Importance of fats and oils in human diet & food industry

2.2 Functional role of fats/lipids in baking

2.3 Importance and consumption of baked products

- 2.3.1 Cake
- 2.3.2 Cookies
- 2.3.3 Piecrusts
- 2.3.4 Doughnuts
- 2.3.5 Muffins

2.4. Hydrogenated fats and their application in baked products

- 2.4.1 Trans fatty acids in processed foods
- 2.4.2 Alternatives to Trans fatty acids in processed foods

2.5. Rice Bran Oil

- 2.5.1 Production and consumption of rice bran oil globally
- 2.5.2 Production and consumption of rice bran oil in India
- 2.5.3 Properties of Rice bran oil and gamma oryzanol
- 2.5.4 Health benefits of Rice bran oil
- 2.5.5 Rice bran oil in baked products

2.6. Novel cooking alternatives to saturated fats in baked products

- 2.6.1 Rice bran oil products as alternatives in baked products

2.1 Importance of fats & oils in human diet & food industry

Fats and oils have been an essential element in human diet since prehistoric times as they are a rich source of energy (9 kcal/g) and essential fatty acids. Approximate fatty acid composition of dietary fats and oils consumed in India as recommended in Dietary guidelines for Indians by National Institute of Nutrition (2010) is given in Table 2.1. Furthermore, fats and oils help carry important water-insoluble substances in the human body, such as fat-soluble vitamins, phytosterols, beta-carotene, lutein and many other substances. (FAO/WHO, 1993) Nowadays, fats and oils play an important role in the food industry as their physical properties impact food products; they contribute to the flavor, texture, lubricity, and satiety of food (Rios *et.al.*, 2014; Mc Clements and Decker, 2010). Artificial *trans* fats are inexpensive and provide long shelf life, stability of food and desirable textures. Artificial *trans* fatty acids and saturated fats are mostly found in partially hydrogenated oils, vegetable shortenings, stick margarine, fast food, ready-to-eat frosting, spreads, savory snacks and frying and baking products (American Heart Association, 2014). To avoid *trans* fats and minimize the intake of saturated fats, the concept of structuring oils that can carry the characteristics of solid crystallized fats has been very active since the last recent years (Marangoni and Garti, 2011)

2.2 Functional role of fats/lipids in baking

Baked foods serve as one of the main staple food sources for consumers and lipids play an important role in the majority of baked products. When choosing a lipid source for baking, the nutritional characteristics of lipid and the ability of the lipid to impart a desired physical quality to the finished product need to be considered. Usually, animal fats, such as lard, are more saturated than plant oils such as soybean oil, and their polyunsaturated fatty acid (PUFA) vs. Saturated fatty acids (Rogers, 2004). In addition to the nutritional profiles, lipids should be selected according to their specific performance in the finished product (Wainwright, 1999). In general, lipids in baking contribute to products' quality characteristics such as tenderness, moist mouth feel, lubricity, flavor, structure and shelf life. During processing, lipids affect air incorporation, lubrication and heat exchange in the baked products (Ghotra *et al.*, 2002).

In addition to providing energy for the human body, fats have functional properties that shape the color, flavor, texture, tenderness, and emulsification of food as well as provide a cooking medium (McWilliams, 2008).

Butter provides a pleasant creamy yellow color to food products, and many brands of margarines have added beta carotene to simulate the desired yellow color. Fats contribute a rich flavor to food that is seldom replicated. Butyric acid in butter marks the distinctive taste of butter; "olive oil and lard are examples of other types of fats that contain distinctive flavor components" (McWilliams, 2008). Butter is creamed with sugar "to obtain a very fine cell structure of great uniformity in a shortened cake" (McWilliams, 2008). This produces a fluffy and light texture in baked products.

Fat is a tenderizing agent, and it is important in the creation of a tender baked product. Because of their "ability to interfere with the development of gluten, the structural protein complex in wheat flour products," fats are essential to baked goods. This is referred to as shortening power, the "ability of a fat to cover a large surface area to minimize the contact between water and gluten during the mixing of batters and doughs" (McWilliams, 2008).

Flakiness in pastries is achieved when fat in the dough "melts during the baking period and flows, leaving a hole where steam collects and pushes upward against the upper surface of the resulting cell" (McWilliams, 2008). This is highly desirable of biscuits with thin, flaky layers. Emulsification of foods result in uniformly dispersed ingredients, which are mostly oil-in-water emulsions. Fat as an emulsifying agent is used to aid in the formation of stable emulsions, as shortenings are produced today "with added mono and diglycerides, so that the fat and milk in cake batters are emulsified, resulting in a fine-textured cake" (McWilliams, 2008). Finally, fats and oils are used as a cooking medium in deep-fat frying and stir-frying.

2.3 Importance and consumption of baked products

2.3.1 Cakes

Zhou *et al.* (2011) studied that shortening is a major ingredient used in high-ratio layer cakes. Plastic shortenings are most commonly used by the U.S. baking industry, but high levels of trans or saturated fats cause health concerns. Compared to plastic

shortenings, liquid shortenings could significantly reduce the dependence on high melting point fats and the emulsifiers used would enhance the shortening's functionality. Cakes baked with soybean oil to evaluate the function of three emulsifiers (PGMS, GMS and Lecithin) on layer cake quality, including volume, cake score, interior visual texture (C-Cell), and firmness (Zhou *et al*., 2011) was studied. An optimum emulsifier combination was chosen (PGMS 1.8%, GMS 1.0% and Lecithin 0.8%) for addition to the liquid oil. Four groups of layer cakes were baked using: plastic shortening, liquid shortening, liquid oil, or liquid oil plus emulsifier combination. Cake performance and firming over-time were evaluated. The liquid shortening provided the best fresh cake characteristics and cake firmness performance. Liquid oil combined with a combination of added emulsifiers performed very similar in terms of firmness, as did the liquid shortening. This indicated that emulsifiers played an important role on the improvement of cake firmness shelf-life.

In cakes, lipids play a predominant role in determining the final cake structure (Hartnett and Thalheimer, 1979). This affects the volume and texture of the finished cake (Stauffer 1998). A homogeneous cake crumb that contains small aerated pores is considered a desirable cake quality (Rutkowska and Zbikowska, 2010). Air incorporation is equally important in cookies (Rogers, 2004) and lipid type influences cookie spread (Wainwright, 1999).

Zhou *et al*. (2011) studied that as a porous food, cake quality largely depends on air incorporation. Plastic lipids contain solid crystals that can adsorb to air bubbles incorporated into batter during mixing and stabilize them. Previous research suggests that, along with the initial air incorporation, final cake density depends on the stability of air bubbles during baking. As the oven is heated up, bubbles tend to migrate and expand, while lipid crystals (absorbed to the air bubbles during mixing) melt and form a uniform layer over the inside surface of the bubble, allowing bubble expansion without rupture. Melting that occurs too early or too late during this process will lead to aggregation and loss of air bubbles and a denser cake.

Rutkowska and Zbikowska (2010) determined the effects of fatty acid composition of liquid margarines (LM) on the sensory properties of sponge cakes and compared it with cakes containing solid margarines and butter. Fats were different in principal groups of fatty acid and were of the following composition: SFA (9.3-66.4), TFA (1.2-10.3), PUFA

(1.9-53.9%). The taste and aroma of cakes scored lower with increasing content of unsaturated FA, especially PUFA and higher with increasing content of SFA and TFA combined. With respect to taste and aroma, cakes containing LM scored lower in comparison to cakes containing butter, while the texture of such cakes was rated as high as cakes containing butter. Approximately 34-65% of young consumers perceived such cakes as "like very much" and "like moderately", however, 5% of consumers described them as "dislike a lot". The results of this study indicate that liquid margarines could be recommended for cakes in large-scale manufacture. Regarding recommendations of reducing the intake of TFA in processed foods, it is justified to replace hydrogenated vegetable oils by liquid fats for manufacture of bakery products.

Jeyarani *et al*. (2009) reported that shortenings containing *trans* fat usually have more plastic character, higher melting points and greater stability against oxidative rancidity. In cake production, these characters are highly important in initial air incorporation and air bubble protection during baking.

2.3.2 Cookies

Cookie preparation starts with mixing of the ingredients into “short” dough. In the first step, a semi-stiff white cream is prepared by mixing the sugar, shortening and water (from egg). In the second step, the flour is added with minimal mixing resulting in cookie dough with minimal gluten development if any.

One major role of shortening in cookie dough is to eliminate gluten development by breaking the continuity of the protein and starch structure (Ghotra *et al*., 2002). This effect varies with different shortening level and different liquid oil portion within a shortening. If the total shortening level or liquid oil portion within the shortening is high, water or sugar solution would be coated by shortening, resulting in less chance to interact with flour protein in the second step, therefore little gluten will be formed in final cookie (crisp or crumbly texture). On the other hand, with a low total shortening level or low liquid oil portion, more gluten will be formed and therefore resulting in a hard or chewy cookie texture. Hence for different texture requirements, different shortening will be selected.

A second function of shortening in cookies is aeration (less so than in cake making). Plasticity in shortening is required for this function since during the first creaming process,

a plastic shortening can entrap and retain air that has a leavening effect (Jacob and Leelavathi, 2007).

Bakota *et al.* (2013) developed a new solvent fractionation procedure for the production of a spreadable product derived from rice bran oil. A study by Jacob and Leelavathi (2007) reported that cookies containing rice bran oil had relatively higher spread value, while the cookies containing the non-emulsified hydrogenated fat (Dalda) had the least spread.

Sharif *et al.* (2005) investigated application of rice bran oil in baked products such as cookies at various levels i.e. 0, 25, 50, 75 and 100% by gradually replacing normal shortening to improve the quality of cookies in term of shelf life due to natural antioxidants present in RBO. Five treatments of RBO and normal shortening (NS) were used to prepare cookies and 45 days' storage study was conducted to investigate improvement in shelf life. Statistical analysis on chemical characteristics of cookies indicated that both treatments and storage have highly significant effect on moisture content, fat content and NFE content of cookies while ash content and crude fiber showed non significant change. Storage also had significant effect on protein content of cookies, but it remains unaffected by changing treatments. During 45 days' storage, moisture content, protein content, fat content and NFE content decreased significantly. The thiobarbituric acid number (TBA no.) was calculated after each storage interval (fortnightly). Both treatments and storage showed significant effect on TBA no. of cookies. Treatment T5 (100% RBO) exhibited TBA no. (0.03), while T1 (100%NS) showed (0.05) mg of malenaldehyde/Kg at 0 day. There was an increase in TBA no. during storage, but Treatment T5 (100%RBO) showed the minimum increase (0.05) followed by T4 (0.06) and T3 (0.08). Based on the results of proximate analysis, sensory evaluation and TBA number, it was concluded that by increasing the percentage of rice bran oil (RBO), the TBA number decreases and the onset of rancidity is delayed. Moreover, the study suggests that T3 (50%RBO + 50%NS) can produce superior quality cookies to prove effectiveness of RBO as bakery shortening.

Pareyt and Delcour (2008) reported that the degree of spread resulting in the final cookie diameter is controlled by the spread rate and set time of the dough. Shortening functions as a lubricant in cookie dough, thus affecting spread rate: more shortening/more liquid oil portion gives a larger spread.

2.3.3 Piecrust

In piecrust production, shortening and flour are first pinched together. Then water is added to form the crust dough. During this process, the role of lipid is to prevent excessive gluten formation and create pockets in the dough (Stauffer, 1998). Gluten in dough is formed by the interaction of flour and water (Patient, 1994).

Pyler and Merriam (1988) studied that lipids with a more fluid character tend to limit gluten development, since flour particles are covered by lipid and have less chance to interact with water. However, solid lipids are more likely to be coated by flour particles than vice versa, and form pockets of lipid within the dough. Therefore, flour proteins are able to interact with water, leading to increased gluten development and tougher pie crusts.

Ghotra *et al.* (2002) studied that in pie crusts lipids with a proper solid character and melting point help contribute to desirable flakiness and tenderness.

2.3.4 Doughnuts

Tan and Mittal (2006) studied the effects of vacuum frying (up to 9kPa of vacuum) on cake doughnut properties and found that volume increased, firmness decreased, and density decreased with increasing vacuum. Fat content was found to be significantly affected by both frying temperature and amount of vacuum. Lower frying temperatures and higher vacuum levels resulted in an increased fat content. Some properties of finished doughnuts have been shown to be a function of cooking time. Volume, chewiness, cohesiveness, gumminess, springiness, force to break crust and force to compress crumb increase and density were shown to decrease as frying time increased.

Oil temperature, cooking time and atmospheric pressure have been found to affect more than oil content. Velez and Sosa (2003); Tan and Mittal (2006), using oil temperature ranges of 180°C to 200°C and 165°C to 180°C, respectively, found that increased oil temperature resulted in decreased volume, increased density, increased firmness, increased crust darkening, and increased thermal diffusivity in both cake and yeast doughnuts.

Doughnuts with low initial moisture content tended to have cracks in the surface of the dough, enabling oil to flow easily into the doughnut while frying and increasing oil uptake (Tan and Mittal, 2006).

Velez and Sosa (2003) studied that in yeast doughnuts, oil absorption tends to decrease with increasing oil temperature, but increases with increasing cook time. However, the oil content of the doughnuts was shown to decrease after a certain frying time. The frying time at which the oil content began to decrease was temperature dependent and the decrease in oil content was lower with increased oil temperature. Velez and Sosa (2003) attributed these effects to a desorption phenomenon; hypothesizing that the oil desorption was due to structural changes in the doughnuts. Yeast doughnut crumb was found to become more compact after prolonged cooking; the denser crumb could have prevented absorption of more oil.

Velez and Sosa (2003) also found the heat capacity and thermal conductivity of doughnuts to be independent of oil temperature over the range studied. The necessary frying time, or the time needed for the doughnut center to reach 99°C, and doughnut crust color change appeared to be independent of level of vacuum; moisture content appeared to be independent of both level of vacuum and frying temperature (Tan and Mittal, 2006).

Shih *et al*. (2001**)** studied the effects of modified and unmodified rice flours mixed with wheat flour on various doughnut properties and compared the results to those of doughnuts made with only wheat flour. Oil uptake decreased by up to 64%, depending on the rice flour type used and the level of wheat flour replacement. Final moisture content also depended on flour type, although all doughnuts had a final moisture content of 18-21%.

Velez and Sosa (2003) found that yeast doughnut volume reached a maximum, then decreased after a certain cooking time. The frying time at which the maximum volume was reached was temperature dependent and decreased with increased temperature.

2.3.4 Muffins

Kaur *et al.* (2014) studied replacement of bakery shortening with refined rice bran oil in the preparation of muffins. Physico-chemical properties of bakery shortening and rice bran oil were studied. Rice bran oil was found to have a higher content of the essential fatty acid linoleic acid (34.98%) as compared to that of bakery shortening (5.14%). Chemical composition of wheat flour used was also studied. Muffin samples were prepared by replacing bakery shortening with rice bran oil at 0, 25, 50, 75 and 100% levels.

Muffins were examined for quality that is weight, volume, specific volume and organoleptic quality that is appearance, colour, texture, flavour and overall acceptability on a 9 point hedonic scale. Statistical analysis revealed that muffin making and organoleptic quality of muffins prepared after replacing rice bran oil at the 50% level or greater varied significantly, which is desirable from that of control. Statistically significant variations were observed in the texture of muffins prepared with shortening alone from that prepared after replacing bakery shortening with rice bran oil at 50% level.

A study on quality characteristics of muffins in steam assisted and conventional ovens by Yilmazer *et al*. (2011) reported that steam assisted baking was a better choice over natural convection or forced baking in terms of moisture content, height, bulk density and browning index of muffins. It was also reported that convention baking resulted in lower acrylamide content.

2.4 Hydrogenated fats & their application in baked products

With recent advances in food technology, food manufacturers have utilized the process of hydrogenation of polyunsaturated fats to include in everyday food items. Hydrogenation is the "addition of hydrogen to an unsaturated fatty acid in the presence of a catalyst to reduce the unsaturation of the molecule and raise the melting point" (McWilliams, 2008). This process started in the early 20th century that led to the introduction of commercial *trans* fats into the American diet. It was first described by Paul Sabatier, a French chemist and "uses a nickel catalyst to hydrogenate – or saturate – double bonds in vegetable oils" (Remig *et al*., 2010). In between hydrogenation and unsaturated fatty acids, partial hydrogenation of vegetable oils produces both *cis* and *trans* fatty acids.

Partially hydrogenated oils also withstand repeated heating without breaking down or going rancid, making them ideal for frying fast foods (Frenk, 2012). Partially hydrogenated oils became popular in restaurants and the food industry for many uses, including frying, baking and processing snack foods and margarines.

Along with the functional aspects of hydrogenated oils, the food industry wanted to find a healthier substitute for using animal fats in food products. Because of the health concerns of saturated fats and cholesterol, food manufacturers wanted to find a replacement to use in food products that would reduce saturated fats and cholesterol content. Unsaturated vegetable oils are usually liquid at room temperature, but with this

hydrogenation process, the unsaturated vegetable oils can be modified to solids and used as margarines and shortenings. Hydrogenation is also used to process peanut butter from its original state of oil separation to remain "homogenous even during extended shelf storage" (McWilliams, 2008). With this hydrogenation process, food manufacturers believed that it would be "healthier" to suit the needs of people trying to reduce their saturated fat and cholesterol intake while providing the same qualities as the saturated fat counterparts. Although some undesirable results occur with partially hydrogenated fats because "some unsaturated fatty acids undergo isomerization, which results in trans configuration of some double bonds rather than the *cis* form" (McWilliams, 2008). Completely hydrogenated oils are sometimes used since they do not contain double bonds

2.4.1 Transfatty acids in processed foods

In biochemistry, saturated fats have C-C single bonds. This means that all the carbons on the fatty acid are "saturated" with hydrogen, giving the fatty acid stability. Unsaturated fats have C=C double bonds. The *cis* forms of polyunsaturated fatty acids are oils which are liquid at room temperature. Trans fatty acids "produce more rigid configuration that requires much less space than the *cis* double bond, resulting in a melting point around room temperature" (Remig *et al.*, 2010). This provides a desirable texture and mouth-feel along with enhancing product shelf life. Dietary *trans* fats, first of all, are formed "naturally by bacteria present in the rumens of ruminant animals" (Remig *et al.*, 2010).

Generally speaking, dairy and meat products have trace amounts of *trans* fats. In a man-made process of hydrogenation or partial hydrogenation, *trans* fats are formed from liquid vegetable oils high in unsaturated fatty acids. These sources of *trans* fat include but not limited to stick margarines, soft or tub margarines, whipped margarines, and shortening. Stick margarines are spreads "made by hydrogenating plant oils and adding water, milk solids, flavoring, and coloring to achieve a produce similar to butter" (McWilliams, 2008). Soft or tub margarines are similar spreads, but with lower melting points than those of stick margarines due to more polyunsaturated fatty acids. These margarines are less hydrogenated, so not as many saturated C-C bonds form, giving them a less solid structure than harder, more hydrogenated margarines. Whipped margarines have been whipped mechanically into foam; therefore, the increased volume gives whipped

margarines less calories per volume in comparison to the rest. Shortening is the "hydrogenation of vegetable oils to achieve the desired consistency" (McWilliams, 2008).

In batters and doughs, mono and diglycerides are added to shortening for forming better emulsions. Commercial hydrogenation generates the majority of the *trans* fats found in the American diet today. However, the negative effects of *trans* fatty acids in the diet have brought about changes to produce healthy spreads with reduced amounts of *trans* fatty acids (McWilliams, 2008).

Research by Kong (2012) on health implications and subsequent policy changes of the trans fatty acids, such as listing trans fat content on food label nutrition facts, led the same food companies to re-evaluate the hydrogenation process, or production of trans fat and begin using alternatives to partially hydrogenated oils. Fifteen food companies were contacted through their websites and surveyed about what kinds of oils and/or methods they used to replace the previously used partially hydrogenated oils in their food products to maintain standards for taste and texture. This research and literature review reveals that food companies are back to using saturated fats and tropical oils, especially coconut oil, instead of the cheap trans fats in their products.

Semma (2002) reported that trans fatty acids have several beneficial aspects for processed foods owing to their characteristic structures. These very characteristic structures, in turn, have been suspected to be associated with the possibility that trans fatty acids affect the development of several health problems, including coronary heart disease, fetal and infant neurodevelopment, growth, and childhood allergies.

Trans fatty acids (TFA) have similar melting points to those of the corresponding saturated fatty acids and are extremely important contributors to the functional properties of hydrogenated products (Ghotra *et al.,* 2002).

2.4.2 Alternatives to Trans fatty acids in processed foods

Alternatives to *trans* fatty acids are becoming more available and widespread. Food companies are exploring a number of options for replacing trans-fat-containing partially hydrogenated oils. These options include "changes in oils used, extent of hydrogenation, interesterification, alterations in diacylglyceride levels, adding short- and

medium-chain fatty acids and changing the fatty acids in plants through selective breeding and genetic alterations" (McWilliams, 2008).

Many food companies are going back to butter and animal fat, what trans fatty acids were supposed to replace in the first place (Frenk, 2010). Although saturated fats and cholesterol in these products concern the public, recent research has shown that saturated fats are not the enemy in chronic diseases, such as cardiovascular disease, cancer and obesity.

Coconut and palm oils were "more widely used in prepared food until 1988" when Americans began to worry that high-saturated fat oils were the cause of chronic disease (Frenk, 2010). This caused food companies to replace these tropical oils with partially hydrogenated oils made from soy, corn, sunflower and rapeseed. Recent research shows that coconut oil strongly increases HDL cholesterol, which may make it a good choice when hard fat is needed (Norton *et al*., 2004).

Another alternative to partially hydrogenated oils is to use fully hydrogenated vegetable oils. In exposing liquid oils rich in unsaturated fats to hydrogen gas for a longer time, a hard, waxy, full hydrogenated fat is produced. This means the oil has become a saturated fat and is *trans*-fat free. Combining fully hydrogenated vegetable oils and liquid vegetable oils "yields a semi-soft fat that is *trans* fat free and can be used for margarines, baking and other types of cooking" (Frenk, 2010).

Using traditional liquid vegetable oils is another healthy alternative to using trans fats. Olive, canola, corn and soy oils spoil faster than partially hydrogenated oils and don't have a long shelf life. They don't hold up as well as partially hydrogenated oils to the high temperatures needed for commercial deep drying. However, "the food industry is rapidly changing its practices and food formulations so that more vegetable oils can be used successfully in foods" (Frenk, 2010) by substituting partially hydrogenated oils for pure vegetable oil. However, this is more expensive to using *trans* fats, and the unsaturated oils do not produce the same texture and quality in food products due to being liquid at room temperature.

Another alternative is to use new liquid vegetable oils from new seeds. Linolenic acid, an omega-3 fatty acid is most responsible for spoilage in oils because it is easily

oxidized. Alpha-linolenic acid is a carboxylic acid with an 18-carbon chain and three *cis* double bonds, an essential fatty acid only acquired through diet. Iowa State University and other universities started selective breeding programs to create soybeans and other seeds with very low levels of linolenic acid (Frenk, 2010). High levels of linolenic acid will make products more susceptible to oxidation, therefore becoming rancid quicker. However, this process eliminates healthful alpha-linolenic acid, one of the healthiest components of many vegetable oils, so it will be important to get omega-3 fats from other sources, such as fatty fish or walnuts.

Remig *et al*. (2010) states that food manufacturers use genetically modified plants that produce low-linoleic, mid-oleic, or high-oleic oils as another option. "Frito-Lay (Pepsico, Purchase, NY), for example, now uses mid-oleic sunflower oil for its potato chips" (Remig *et al*., 2010). However, developing a reliable supply of these oils is a challenge due to lower crop yields.

Interesterification, a process that repositions the fatty acids on triglyceride molecules, can replace partially hydrogenated oils used in food production. According to McWilliams (2008), interesterification is "treatment of a fat, usually lard, with sodium methoxide or another agent to split fatty acids from glycerol and then to reorganize them on glycerol to form different fat molecules with less tendency to form coarse crystals." This does not reduce trans fats if they are already present before the treatment, and health implications of this new commercial fat are not known. Interesterification can produce shortenings and margarines with higher melting points and good spreading characteristics without partially hydrogenating the oils. Nova Lipid, based on interesterification, uses "a mix of approximately 1 part fully saturated soybean oil (high in stearic acid with a melting point of approximately 70°C) and 3 parts liquid soybean oil" (McWilliams, 2008). This fat is solid at room temperature and used in baking. Stearic acid is useful in this product because it does not appear to influence LDL blood levels along with contributing a high melting point.

Stanol and sterol esters have emerged as the effective compounds in alternative spreads. Phytosterols (beta-sitosterol) have similar cholesterol structures but with different side chains that alter their action in the body (McWilliams, 2008). Esterifying phytosterols with a fatty acid and hydrogenating them is necessary for incorporation into margarines. On

the other hand, phytostanols (beta-sitostanol or campestanol) are known as “tall oils” since they are derived from pine tree pulp. Like phytosterols, phytostanols have to be esterified with a fatty acid, though hydrogenation is not required (McWIlliams, 2008). Some products that use stanol and sterol esters include “Take Control and Benecol”. “Take Control”, a spread developed by Lipton, Unilever’s subsidiary, contains a plant sterol ester derived from soybeans (McWilliams, 2008). “Benecol” (marketed by Nc Neil Consumer Healthcare) is a plant stanol ester. These products work by preventing the absorption of LDL cholesterol, which helps reduce the risk of coronary heart disease (McWilliams, 2008). LDL cholesterol is synthesized in the liver and circulates through the bloodstream, depositing in the walls of arteries. Fortunately, two servings of these spreads a day is effective since these products are expensive compared with regular margarines.

Eady *et al.* (2011) analysed the effectiveness of a new phytosterol-containing spread derived from rice bran oil (RBO), in a randomised, double-blind, cross-over human clinical trial conducted over 12 weeks. A total of eighty mildly hypercholesterolaemic (total blood cholesterol level ≥ 5 and ≤ 7·5mmol/l with a serum TAG level of ≤ 4·5mmol/l) individuals were randomized into two groups (n=40). Group 1 consumed spread only daily for 4 weeks. They were randomized to consume 20g RBO spread (RBOS), 20g standard spread (SS) or 20g phytosterol-enriched spread (PS). After a 4-week period, individuals changed to the next randomized treatment until all three treatments had been consumed. Group 2 consumed spread plus oil daily for 4 weeks. They consumed 20g RBOS plus 30ml RBO, 20g SS plus 30ml sunflower oil or 20g RBOS. Blood samples were collected for the analysis of lipid parameters, and 3 day (3d) diet records were collected. Compared with SS, RBOS significantly reduced total cholesterol by 2·2% (P =0·045), total cholesterol: HDL by 4·1% (P = 0·005) and LDL-cholesterol by 3·5% (P=0·016), but was not as effective overall as PS, which reduced total cholesterol by 4·4% (P=0·001), total cholesterol: HDL by 3·4% (P=0·014) and LDL-cholesterol by 5·6 % (P=0·001). In group 2, the addition of RBO to the RBOS produced no differences in cholesterol levels. These results confirm that RBOS is effective in lowering serum cholesterol when consumed as part of a normal diet

Sharma *et al*. (2006) studied the thermal behavior of pure rice bran oil, sunflower oil and their model blends during deep fat frying. Model blends prepared from pure rice bran oil (RBO) and sunflower oil (SFO) were examined for change in their physico-chemical parameters (acid value, iodine value, colour value, peroxide value and fatty

acids). Repeated deep fat frying processes were carried out using dried potato chips in pure rice bran oil, sunflower oil and their model blends, in order to study the thermal behaviour of pure rice bran oil, sunflower oil and their model blends. Pure rice bran oil and sunflower oil showed good thermal stability during the repeated deep fat frying cycles. Although all the blended oils used in the study showed good thermal stability during repeated deep fat frying cycles, model blends consisting of 60%RBO + 40% SFO showed better suitability during repeated deep fat frying than the remaining blended oils.

The work of Debnath *et al.* (2012) deals with the effect of frying cycles on physical, chemical and heat transfer quality of rice bran oil (RBO) during the preparation of *poori* (an Indian traditional fried food) by deep-fat frying. The frying was carried out in intermittent mode (5 batches each for 3 min in a day without any time lag) and repeated for 6 frying cycles. Result indicated that in first two cycles, free fatty acid content, peroxide value and total polar materials increased, while radical scavenging activity decreased. Further increase in frying cycles did not result in any significant changes in these parameters ($p > 0.05$). Similar trends were observed for these parameters in case of heating. Relative amounts of total saturated fatty acids increased due to marginal decrease in total unsaturated fatty acids content during frying cycles, however, no significant change was observed during heating. The convective heat transfer coefficient was found to decrease with an increase in frying cycle due to increase in kinematic viscosity of RBO for every frying or cycles. Despite the marginal changes in physical and chemical properties, there was no significant difference ($p > 0.05$) in the sensory characteristics of *poori* prepared in oil subjected to different cycles of frying.

2.5 Rice Bran Oil

Chotimarkorn and Silalai (2008) studied the effect of proportion of soybean oil to rice bran oil on lipid peroxidation inhibition in fried dough with rice flour during storage. There was no significant difference in decreasing gamma-oryzanol contents of fried dough during storage ($p > 0.05$). These results demonstrated the effect of soybean oil mixtures with 50% and 75% rice bran oil on retardation of oxidative rancidity and hydrolytic rancidity in fried dough during storage.

Tahira *et al.* (2007) studied the characterization of rice bran oil, which was taken from the Pattoki Rice Mills, Jaranwala and was stabilized to inactivate lipase activity. The oil was extracted through solvent extraction. The extracted oil was subjected to refining process. Different physico-chemical parameters were characterized. The refractive index, peroxide value, iodine value, and free fatty acid value were recorded as 1.4792, 0.92meq/kg, 105, and 0.07% (as oleic acid), respectively. The fatty acid profile showed palmitic acid (16.74%), stearic acid (1.9%), oleic acid (42.79%), linoleic acid (34.65%) and linolenic acid (0.19%) as major fatty acids.

Singh *et al*. (2007) studied the thermal behavior of pure RBO, safflower oil and their model blends during deep fat frying. Model blends prepared from pure rice bran oil (RBO) and safflower oil (SFO) were explored for changes in the physico-chemical parameters (acid value, iodine value, specific gravity, refractive index, colour value, peroxide value and fatty acids of triglycerides). Pure RBO and SFO had shown good thermal stability during the repeated deep-fat frying cycles. Although, all the blended oils, used in the study showed good thermal stability during repeated deep-fat frying cycles yet model blends constituting 20% RBO + 80% SFO showed better suitability during repeated deep fat frying than other blended oils.

Rangaswamy and Nasirullah (2011) studied the physico-chemical changes in rice bran oil during heating at frying temperature. Rice bran oil was subjected to static heating at 180±2°C in a domestic fryer for 8h. In this process, 150ml of the heated oil samples were drawn, at intervals of every 2h, to study the changes in the physico-chemical characteristics. Results indicated that the peroxide value and free fatty acid content increased gradually from 0.2 to 2.9meq. O2/kg of oil and 0.25 to 0.63% respectively. The oil became darker as given by the colour value (5R + Y) 63 Lovibond units. Tocopherol content was found to decrease from 48mg/100gram to 5mg/100gram at the end of 8h of heating whereas, oryzanol was fairly stable (1.59 to 1.40%). The p-anisidine value and Total polar compound (TPC) increased from 5.04 to 18.30 and 1.0 to 1.8% respectively, showing the formation of secondary oxidation products. Rice bran oil is a non-Newtonian fluid having shear thinning behavior. Heating was found to cause an increase in the flow behavior index. Fatty acid composition did not show significant changes except for the linoleic acid content which decreased from 29.4 to 27.1%.

The effect of sesame oil (SEO) and rice bran oil (RBO) on the rancidity of canola oil (CAO) during the process of frying potato pieces at 180ºC was investigated by (Farhoosh and Kenari, 2009). The SEO and RBO were added to the CAO at levels of 3 and 6%. Frying stability of the oil samples during the frying process was measured on the basis of total polar compounds (TPC) content, conjugated diene value (CDV), acid value (AV), and carbonyl value (CV). In general, frying stability of the CAO significantly ($P < 0.05$) improved in the presence of the SEO and RBO. The positive effect of the SEO on the stability of the CAO was more than that of the RBO. Increasing the amounts of SEO and RBO from 3 to 6% led to decreases in the TPC and AV, and increases in the CDV and CV of the CAO during the frying process. The best frying performance for the CAO was obtained by use of 3% of both SEO and RBO together.

Zigoneanu *et al*. (2008) studied that rice bran oil, extracted by microwave-assisted extraction with isopropanol and hexane using a solvent-to-rice bran ratio of 3:1 (w/w). The increase in total vitamin E with temperature from 40 to 120°C was 59.63% for isopropanol and 342.01% for hexane. Isopropanol was the best solvent for the extraction of γ-tocopherol and γ-tocotrienol as compared with hexane for both microwave-assisted and conventional solvent extraction. Isopropanol was better for oil yield extraction at high temperatures. Samples extracted with isopropanol at 120°C had higher antioxidant activity. No differences in oil yield, total vitamin E and antioxidant activity of oil was noticed between the two methods (microwave-assisted and solvent extractions), at 40°C. No degradation of α-tocopherol was noticed during the process.

Hoed *et al*. (2006) investigated the effects of each individual step of the chemical refining process on major and minor components of rice bran oil. In comparison with common vegetable oils, rice bran oil contains a significantly higher level of several bioactive minor components such as γ-oryzanol, tocotrienols and phytosterols. Alkali treatment or neutralization results in a significant loss of oryzanol. In addition, it gives rise to a change in the individual phytosterol composition. After bleaching, some isomers of 24-methylenecycloartanol were detected. Because of their relatively high volatility, phytosterols and tocotrienols are stripped from the rice bran oil during deodorization and concentrated in the deodorizer distillate. At the same time, oryzanol is not volatile enough to be stripped during deodorization; hence, the oryzanol concentration does not change after deodorization. Complete refining removed 99.5% of the FFA content. Depending on

the applied deodorization conditions, trans FA can be formed, but the total trans content generally remains below 1%.

Danielski *et al.* (2005) reported that rice bran, a by-product of rice processing, obtained through the polishing of the rice grain presents considerable high oil content (between 20 and 25%) and is considered an excellent source of nutritionally beneficial compounds, such as tocotrienols, tocopherols and sterols.

Sharif *et al*. (2005) reported that the vegetable oils are generally excellent sources of polyunsaturated fatty acids. Although general consumption of fats and oils has increased, recent trends in the baking industry are to reduce total usage of fats and oils and to replace plastic fats with liquid vegetable oils.

Shiela *et al*. (2004) investigated the storage stability of blends of groundnut oil, sunflower oil and mustard seed oil with plam olein, rice bran oil and sesame oil. Physicochemical properties and levels of minor and potentially health-promoting components of the blended oils packaged in pouches were studied during storage for 6 months at 27°C and 65% RH (relative humidity) or 40°C and 30-40% RH. Colour, peroxide value (PV; 0.6-20.7meq O2/kg), and contents of free fatty acids (0.08-2.1%), tocopherol (360-1700 ppm), oryzanol (460-2000mg %) and sesame antioxidants (400-2000mg %) did not change in either unblended or blended oils. Oils and oil blends containing a higher initial PV (18.9-20.7meq O2/kg) showed a slight reduction in PV at 40°C, whereas oils having lower PV (5-10meq O2/kg) showed slight increase during storage. The β-carotene contents decreased by 8.9-60.2% at 27°C and 48-71% at 40°C. Results suggest that the packaged oil blends tested were stable under the conditions used.

Pandey *et al*. (2004) analysed edible oils such as coconut, groundnut, hydrogenated vegetable, linseed, mustard, olive, palm, refined vegetable, rice bran, safflower, sesame, soybean and sunflower for the presence of light and heavy polycyclic aromatic hydrocarbon (PAH) residues using liquid-liquid extraction, cleanup on a silica gel column, and resolution and determination by HPLC using a fluorescence detector. Analysis of 296 oil samples showed that 88.5% (262) samples were contaminated with different PAH of 262 contaminated edible oil samples, 66.4% of the samples showed PAH content of more than the 25micro g/kg recommended by the German Society for Fat Science. The total PAH content was highest in virgin olive oil (624micro g/kg) and lowest in refined

vegetable oils (40.2micro g/kg). The maximum content (265micro g/kg) of heavy PAH was found in olive oil and the minimum (4.6micro g/kg) in rice bran oil. The intake of PAH was highest through olive oil (20.8 micro g/day) followed by soybean oil (5.0micro g/day) and lowest through refined vegetable oil (1.3micro g/day).

Mellema (2003) reported that, it is necessary to understand the mechanism for oil uptake during immersion frying to reduce the amount of oil uptake in fried foods. As a food is fried, the surface begins to dry due to evaporation of moisture. This surface drying causes a porous crust to form as the drying continues during the frying process, the crust thickens (Farkas *et al*., 1996; Mellema, 2003) and the pores in the crust become larger, especially at high rates of drying (Mellema, 2003). Oil migration into the food occurs when the pressure from water vapor migrating from the core to the crust is not sufficient to prevent oil from entering, either due to low moisture content in the core or an inability of moisture to migrate from the core to the crust (Mellema, 2003). It has been found that most of the oil uptake occurs after frying. As the food cools, evaporation of vapor from the food ceases the pressure in the pores in the crust decreases and oil on the surface of the food moves into the pores via suction (Vitrac *et al.,* 2002; Baik and Mittal, 2005; Vliet *et al*., 2007). Interestingly, the majority of oil absorbed during and after frying is located in the crust of the food (Farkas *et al.,* 1996; Normen *et al.,* 1998; Mellema, 2003).

The type of oil used in frying (Wheeler and Stingley, 1963; Rimac *et al*., 2004) and the batter temperature do not appear to affect oil absorption (Wheeler and Stingley, 1963).

Sundararaj *et al.* (2002) reported an increase in color, refractive index, free fatty acid, peroxide value, anisidine value, total oxidation value and decrease in iodine value after 3, 6, 9 months of storage of rice bran oil at 7°C and 38°C in transparent PET bottles. Fatty acid composition of oil was not affected by storage conditions, while α-tocopherol had been totally destroyed after storage for 9 months, minimal decreases were observed in γ-Oryzanol content. The frying stability of rice bran oil was assessed by measuring the oxidative deterioration undergone by the oil and was compared with groundnut oil (GNO). There was an increase in color, % free fatty acid, peroxide value, anisidine value and total oxidation value of both oils following deep fat frying procedure. Rice bran oil showed less oxidative deterioration as compared to groundnut oil.

Sharma *et al.* (2002) studied that the rice bran oil is an excellent cooking medium because it is nutritionally superior, contains more micronutrients, longer shelf life, more stable at higher temperature, gives better taste & flavor to food items, frying takes less time so saves energy and economic due to 15% less absorption of oil during frying.

Joo *et al*. (2001) investigated effects of rice bran oil on the oxidative stability and nutritional properties of restricted beef roasts and concluded that addition of 2% rice bran oil (W/W) is effective in improving both oxidative stability and vitamin E levels of restructured beef roasts.

2.5.1 Production and Consumption of Rice bran oil globally

RBO is extensively used in Japan, Korea, China, Taiwan and Thailand as premium edible oil. It is the conventional and the most favorite cooking medium of the Japanese, the longest living human beings on this earth and is popularly known as a "Heart Oil" in Japan. In recent years, U.S. Scientists have also shown a tremendous interest in the cholesterol lowering properties of RBO and this oil has acquired the status of "Health Food" with Americans. (Azam *et al*., 2015).

Rice bran and its oil contain large concentrations of compounds that include unsaturated fatty acids, triterpene alcohols, phytosterols, tocotrienols and α-tocopherol which have the potential to prevent chronic disease through their demonstrated ability to favourably alter lipid profiles (Cicero and Gaddi, 2001).

Traditionally, consumption of rice bran oil (RBO) has been limited due to problems with rancidity caused by high lipase activity in the bran which affects the stability of the oil (Rukmini, 1988; Deckere and Korver, 1996). However, methods of extraction have evolved to increase stability and in Asian countries such as Japan, China, Korea, Pakistan, Taiwan and Thailand, RBO is extracted for edible purposes and is in demand as a "healthy oil" (Rukimini, 1988; Sugano and Tsuji, 1997).

Rice bran oil has many favorable characteristics including a mild taste and high smoke point making it suitable for most culinary uses including frying. Rice bran is produced as a byproduct of the rice milling industry obtained during polishing when the bran is separated from between the paddy husk and endosperm (Lee *et al*., 2005).

Rice bran contains 10-25% oil consisting of saturated fatty acids (palmitic acid 17%) and oleic acid (40%). It also contains linoleic acid (40%) and a detectable amount of α-linolenic acid (2%), along with negligible amounts of water-soluble β glucans and larger amounts of insoluble dietary fibre (Sugano and Tsuji, 1997). Of greater interest is the large amount of unsaponifiable material (4.2% in crude oil and up to 2.6% in refined oil) which is much higher than that found in other vegetable oils and is characterised by several compounds including gamma oryzanol and phytosterols (Lee *et al*., 2005).

2.5.2 Production and Consumption of Rice bran oil in India

India produced 158.00Mt of paddy during 2013-14, which is equivalent to 106Mt of rice (67% recovery), and this could yield 9.48Mt of rice bran (6% recovery) which has the potential to yield 1.61 of RBO (17% recovery). However, the country could produce only 0.93Mt of RBO during 2013-14, which is just 58% of the potential. Untapped potential for RBO was 0.68Mt (Azam *et al*., 2015). Rice bran oil production in India from 2004-05 to 2011-12 along with estimated production for 2012-13 is given in Figure 2.1.

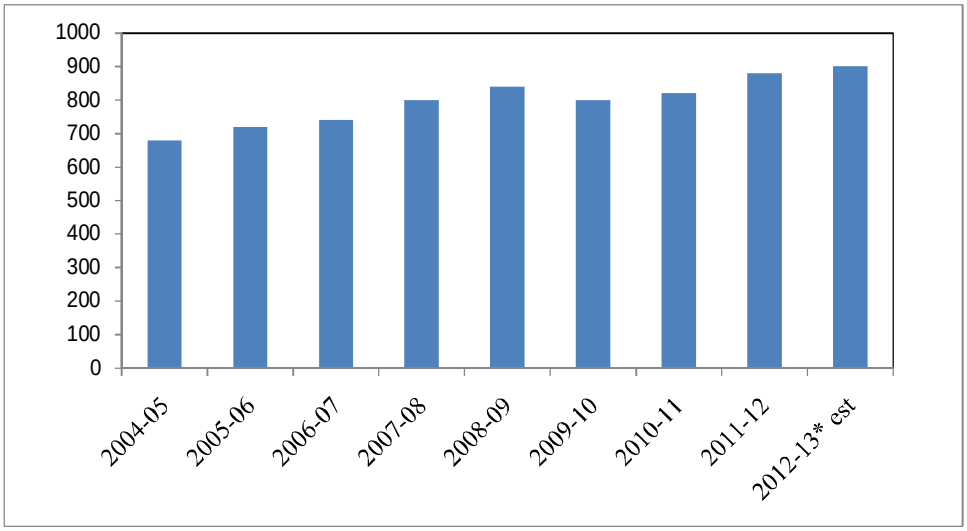


Figure 2.1Rice Bran oil production in India (2004-13)
Source: (Azam *et al*., 2015).

One of the reasons for this wide production-potential gap is the scattered presence of rice mills and transport time is crucial to maintain the quality of rice bran required to

prepare edible oil. A major constraint of RBO extraction is the practice of one stage milling which results in a mixture of hulls and bran with an oil content too low for economic oil extraction. The use of two stage mills provides bran with economic oil content. Another reason which inhibits the full utilization of the bran is its rapid deterioration due to enzymatic hydrolysis. Rice bran contains a powerful lipolytic enzyme system which causes a rapid release of free fatty acids (FFA), up to 30% of the oil being converted to FFA in one week under tropical storage conditions. Freshly milled bran has a FFA of 3%. Oil with more than 10% FFA cannot be economically refined. FFA can be lowered by controlling the lipase activity in bran i.e. by bran stabilization which can be carried out in static and fluidized bed. Another option for keeping the FFA content low is the immediate extraction of the oil from bran. This is possible only when milling and oil extraction units are in the same campus. Wax present in the crude RBO creates problem in refining and high refining losses occur. Conventional methods of degumming/dewaxing are not very economical in rice bran. Physical process like solvent membrane filtration, separation by ion exchange resins will be more suitable (Azam *et al*., 2015).

2.5.3 Properties of Rice bran oil and gamma oryzanol

Rice Bran Oil as a Healthy Cooking Medium: RBO indeed has the most desirable fat composition, better frying stability and offers unique health due to the presence of phytosterols and oryzanols. Its neutral taste makes it great for frying and its chemical make-up means it generally does not burn. It is favourless and has stable shelf-life (Azam *et al*., 2015).

Ideal Fatty Acid Composition: The ratio of SFA: MUFA: PUFA of any oil is very important and as per the latest recommendations by National Institute of Nutrition (NIN) and Indian Council of Medical Research (ICMR) the ideal fatty acid composition of an oil should be 27-33% (SFA) : 33-40% (MUFA) : 27-33% (PUFA). Fatty acid composition of RBO comes closest to these recommendations with the percentages at 24: 42: 34 (Azam *et al*., 2015).

High Smoking Point: The smoking point of a fat is a desirable quality for a cooking medium. When oils are heated beyond their smoking point, they develop free radicals which contribute to risk of cancer. A fat is no longer fit for consumption after it has exceeded its smoke point as it begins to break down. Oils with low smoking point cannot

be used for high temperature cooking / frying. RBO has a smoking point of 254°C. None of the cooking oils have such a high smoking point. High smoking point makes RBO perfect for deep frying, pan or stir frying and is a premium choice for replacement of hydrogenated oil containing trans fat now being used in deep fryers. This oil retains its antioxidant stability even at high temperature (Azam *et al*., 2015).

RBO a rich source of phytonutrients: RBO has about 4% of unsaponifiable fraction. RBO unsaponifiable fraction is rich in vitamin E complex, tocopherols and tocotrienols, a unique antioxidant known as gamma oryzanol, high quantities of phytosterols, polyphenols and squalene. Another antioxidant, oryzanol, makes RBO an unique cooking oil (Azam *et al*., 2015).

Oryzanol is a powerful antioxidant found only in RBO in the order of 2000 ppm . It is more active than Vitamin E in fighting free radicals. Oryzanol is effective in lowering cholesterol level in the blood, reducing liver cholesterol synthesis and treating menopausal disorders. RBO has highest total antioxidant amount (2447 ppm) which makes RBO an oil of choice for possibly improving serum cholesterol levels and helping prevent cardiovascular diseases. These antioxidant and other micronutrients help fight free radicals and combat the effects of aging. RBO has more of these components than other cooking oils. These components of RBO give it an outstanding shelf life. Phytosterols reduce cholesterol, provide anti-inflammatory effects, growth of cancer cells, improve the immune system and have other health benefits. (Azam *et al*., 2015).

Rice bran oil has excellent fatty acid profile. It has oleic acid (38.4%), linoleic acid (34.4%) and linolenic acid (2.2%) as unsaturated fatty acids while palmetic acid (21.5%) and stearic acid (2.9%) as saturated fatty acids (Rukmini and Raghuram, 1991). The saturated, monounsaturated and polyunsaturated fatty acids are in the ratio of approximately 1: 2.2: 1.5 (Shin and Chung, 1998). Three major fatty acids, palmitic, oleic and linoleic make up 90% of the total fatty acids of the rice bran oil (Amarasinghe and Gangodavilage, 2004).

2.5.4 Health benefits of Rice bran oil

Antioxidant compounds in rice bran oil have professed health benefits such as improving the storage stability of foods (Chotimarkorn and Silalai, 2008). Also, it has been

determined that the amount of cholesterol lowering occurs to a greater extent than expected from the fatty acid composition of the oil, suggesting that besides fatty acids, other components in the oil were responsible for the cholesterol lowering effect. Oryzanol has been reported to be used in the cure of nerve imbalance and disorders of menopause (Iqbal *et al.,* 2005).

Valantina *et al*. (2010) reported the antioxidant stability in palm oil and rice bran oil at different times of heating using the parameters like density, viscosity, adiabatic compressibility and acoustic impedance of different oils. The antioxidant stability is resolute at every time of heating. It was recommended that rice bran oil can be used for frying without adverse effect preventing the incidence of malignancy and coronary heart diseases

There is evidence (Mozaffarian *et al*., 2006) that saturated fat is a dietary component with the greatest influence on total blood cholesterol and LDL cholesterol levels, but there are differences in cholesterol-raising effects among the fatty acids. The effects are generally greater for fatty acids with medium chain lengths, e.g. palmitic (C16:0), than for those with longer chain. This study showed that from a nutritional point of view liquid margarines were healthier than solid fats.

Crevel *et al*. (2000) reported that RBO does not produce any allergenic reactions when ingested, unlike several vegetable oils.

RBO lowered cholesterol despite having a large component of saturated fatty acids which are known to be detrimental to cholesterol profiles (Most *et al*., 2005), which supports the theory of the unsaponifiable fraction being the active component of RBO. Whilst these studies show rice bran and RBO to be effective at lowering cholesterol, an earlier study by (Lichtenstein, 1994) showed that RBO was no more effective than other commonly consumed oils. Fifteen moderately hypercholesterolaemic individuals were studied in a randomized double blind trial comparing canola oil, olive oil, corn oil and RBO on plasma lipid concentrations for a period of 32 days. Plasma total cholesterol and LDL-C concentrations were similar and statistically indistinguishable between all treatments. However, the diets were not matched for fatty acid composition which could have been a confounding factor in this study and overwhelmingly the evidence available

supports the hypocholesterolaemic properties of RBO (Deckere and Korver, 1996; Ciero and Gaddi, 2001).

The ability of RBO to lower serum cholesterol levels is one of the proposed health benefits providing some protection against cardiovascular disease. Other possible mechanisms contributing to the positive effect on cardioavascular health include the antioxidant activity relating to γ-oryzanol. Oxidative damage, particularly to LDL-C, has a pivotal role in the development of plaque formation, an initiation event in atherosclerosis (Lee *et al.*, 2005). Many research studies have proven the hypocholesteromic properties of rice bran and rice oil as presented in the Table 2.2

Anti-cancer benefits: These benefits relate to the presence of plant sterols which has been previously discussed and also to the presence of ferulic acid and high levels of vitamin E isomers in rice bran oil. Vitamin E has been shown to prevent or cause regression of certain cancers in animal models and in vitro systems and to a lesser extent in limited large scale intervention trials. Conversely other studies have observed no beneficial effect of vitamin E supplementation in populations of cancer patients (Iqbal *et al*., 2004; Bramley *et al*., 2000). Both tocotrienols and tocopherols are thought to have some anti-cancer activity although tocotrienols are thought to be more effective (Wada *et al*., 2005; Sen *et al*., 2007). Kilne *et al*. (2004) have demonstrated that synthetic vitamin E analogues are capable of including apoptosis through restoration of pro-death signaling pathways in breast, ovarian and prostate cancer cells and Galli *et al*. (2004) showed that tocopherol and tocotrienol metabolites were also capable of inhibiting prostate cancer cell growth in tissue culture. Furthermore, Wada *et al*. (2005) showed that oral supplementation of tocotrienol rich vitamin E prevented lung and liver carcinogenesis in mice and inhibited proliferation of human liver cancer cells in vitro.

Immune Modulation: Sierra *et al*. (2005) conducted a study in mice, feeding them RBO or sunflower oil enriched diets for four weeks and demonstrated that the mice fed RBO had increased stimulation of the immune system with proliferation of the T and B lymphocytes and an enhanced primary macrophage response. Additionally, the mice demonstrated an increase in the stimulation of TH1- type cytokines (IL-2 and TNF-α) whilst having a decrease in the TH2 cytokine, IL4 and Ige suggesting a possible anti-allergenic role for RBO. Furthermore, this study also fed refined RBO and oryzanol enriched sunflower oil to

the mice and found that although the mice demonstrated an increase in TH1 lymphocyte response and reduction in TH2 lymphocyte response, there was no increase in lymphocyte proliferation or IL-4 expression. The authors concluded that the immune modulation effect was due in part to the fatty acid composition rather than simply the unsaponifiable material of RBO. RBO is rich in linoleic acid, an n-6 polyunsaturated fatty acid and precursor to the eicosanoids which play an important role in the regulation of immune and inflammatory response (Calder, 2003).

Neuroendocrinological effects: γ-oryzanol is thought to have a modulatory effect on anterior pituitary hormone secretion with results suggesting that it can inhibit luteinizing hormone, thyroid stimulating hormone, prolactin releasing hormone and growth hormone. Limited evidence supplied from early animal studies provide some support, but human studies are lacking and further research is required in this area (Cicero and Gaddi, 2001).

Osteoporosis: Godber *et al*. (2002) showed some evidence in rats that feeding a 7% RBO concentrate after an ovariectomy reduced the observed amount of bone loss. However other research in this area has not been presented and this effect remains unsubstantiated. Like plant sterols, RBO is considered safe for human consumption and work has found it to have no acute or chronic activity, no mutagenicity or carcinogenicity and no side effects (Ciecero and Gaddi, 2001)

2.5.5 Rice bran oil in baked products

Rice bran oil semisolid fraction (RBOF) can also be incorporated into food formulations (Bakota *et al.,* 2014). Lipid ingredients serve a number of essential functions in baked goods, including acting as a moisture barrier, aiding air incorporation, contributing to the desired structure of the product matrix, providing lubrication for the product in the mouth, and enhancing palatability. Lipid properties also contribute to the shelf life of the finished product. In the past, commercially prepared baked goods have relied on the use of hydrogenated or partially hydrogenated oils to provide the desired product attributes (Cheong *et al.,* 2011).

Sharif *et al*. (2003) investigated usage of Rice bran oil (RBO) into baked products such as cookies at various levels i.e. 0, 25, 50, 75 and 100% by gradually replacing normal shortening to check its effectiveness in extending the shelf life of product due to its natural antioxidants by using thiobarbituric acid number (TBA number) test with the help of

spectrophotometer. Five treatments of RBO and normal shortening (NS) (T1=100 NS+0% RBO, T2=75% NS+25% RBO, T3=50% NS+50% RBO, T4=25% NS+75% RBO, and T5=0% NS+100% RBO) were used to prepare cookies and 45 days' storage study was conducted. TBA number was calculated after each storage interval. There was an increase in TBA number during storage of 45 days. Treatment T5 (100% RBO) showed the minimum increase (0.05) followed by T4 (0.06) and T3 (0.08). It was evident from the results that by increasing the percentage of rice bran oil (RBO), the TBA number decreases and the onset of rancidity is delayed.

2.6 Novel cooking alternatives to saturated fats in baked products

2.6.1 Rice bran oil products as alternatives in baked products

A study by Bakota *et al.* (2014) on usage of rice bran oil derived spread as an antioxidant source in frying oils and as fat replacer in baked goods reported that, use of rice bran oil spread imparted oxidative stability to the oil and when incorporated into two baked goods, there was consistently high acceptability ratings for both baked goods tested.

Kaur *et al.* (2014) studied on replacement of bakery shortening with refined rice bran oil in the preparation of muffins. Physico-chemical properties of bakery shortening and rice bran oil were studied. Rice bran oil was found to have a higher content of the essential fatty acid linoleic acid (34.98%) as compared to that of bakery shortening (5.14%). Chemical composition of wheat flour used was also studied. Muffin samples were prepared by replacing bakery shortening with rice bran oil at 0, 25, 50, 75 and 100% levels. Muffins were examined for quality that is weight, volume and specific volume and organoleptic quality that is appearance, colour, texture, flavour and overall acceptability on a 9 point hedonic scale. Statistical analysis revealed that muffin making and organoleptic quality of muffins prepared after replacing rice bran oil at the 50% level or greater varied significantly which is desirable from that of control. Statistically significant variations were observed in the texture of muffins prepared with shortening alone from that prepared after replacing bakery shortening with rice bran oil at 50% level.

Amarjeet *et al*. (2012) replaced bakery shortening with refined rice bran oil in bread preparation. Physico-chemical properties of bakery shortening and rice bran oil were studied. Rice bran oil was found to have a higher content of essential fatty acid linoleic acid (34.98%) as compared to that of bakery shortening (5.14%). Chemical composition of

wheat flour used was also evaluated. Bread samples were prepared by replacing bakery shortening with rice bran oil at 0, 25, 50, 75 and 100% levels. Breads were examined for bread making quality i.e. loaf weight, loaf height, loaf volume and specific volume and organoleptic quality i.e. appearance, crust colour, crumb colour, aroma, texture, taste and overall acceptability on a 9 point hedonic scale. Statistical analysis regarding bread making quality and organoleptic quality of bread revealed that bread making and organoleptic quality of breads prepared after replacing bakery shortening with rice bran oil at 50% level varied significantly from that of control. Statistically significant variations were observed in the texture of breads prepared with shortening from that prepared after replacing bakery shortening with rice bran oil at 50% level.

Table 2.1 Approximate fatty acid composition of dietary fats and oils consumed in India. (Azam *et al*., 2015)

Fats/oils	SFAs (27-33%)	MUFAs (33-40%)	LA	ALA	Predominant Fatty Acids
High SFAs					
Coconut	92	6	2	-	SFA
Palm Kernal	83	15	2	-	SFA
Butter/Ghee	68	29	2	1	SFA
Palmolein	39	46	11	<0.5	SFA &MUFA
High MUFAs & Moderate LA & ALA					
Groundnut	19	41	32	<0.5	MUFA, Close to recommendations
Rice bran	17	43	38	1	MUFA, Closest to Recommendations
Sesame	16	41	42	<0.5	MUFA, Close to recommendations
Olive	13	76	10	<0.5	MUFA, Close to Recommendations
High MUFAs & High ALA					
Canola	6	60	22	10	MUFA & Omega 3, Close to recommendations
Mustard/rapeseed	4	65	15	14	MUFA & Omega 3, Contains undesirable erucic acid
High LA					
Cotton seed	24	29	48	1	LA/Omega 6
Corn	12	35	50	1	LA/Omega 6
Safflower/Kardi	9	13	75	-	LA/Omega 6
Sunflower	12	22	62	-	LA/Omega 6
High ALA					
Soybean	14	24	53	7	ALA/Omega 3
Flaxseed	10	21	16	53	ALA/Omega 3
High TFAs					
Vanaspati	46	49	4	-	Contains undesirable Trans Fatty acids

Note: SFA- Saturated Fatty Acids, MCT- Medium chain triglycerides, MUFA- Monounsaturated Fatty acids, PUFA- Polyunsaturated Fatty acids, LA- Linoleic acid, ALA- Alpha linolenic acid, TFAs- Trans Fatty acids.

Table 2.2 Hypocholesterolemic properties of rice bran and rice bran oil

Author	Subjects	Treatment	Finding
Gerhart and Gallo,1998	52 moderately hypercholesterolaemic men and women	6wk randomized double blind trial comparing hypocholesterolaemic properties of full fat rice bran, oat bran and rice starch as an addition to normal diets	Rice bran and oat bran produced a significant reduction in serum cholesterol with rice bran reducing LDL-C by 13% and oat bran educing LDL-C by 17.1%. Compared to baseline values. No change to HDL-C or triglycerides was reported
Vissers *et al.*, 2000	28 men and 32 women. Healthy normolipaemic volunteers	3wk randomized double blind crossover trial looking at the effects of plant sterols from rice bran oil and triterpene alcohols from shea nut oil on lipid profiles	2.1g plant sterols from RBO lowered serum cholesterol by 5% and LDL-C by 9%. Triterpene alcohols from shea nut oil did not affect lipoprotein concentrations
Queshi *et al.*, 2002	90 hypercholesterolemic men and women	Randomised cross over trial of 3 phases each for 35 days evaluating the effects of different doses of a tocotrienol rich fraction (TRF25) of stabilized and heat treated rice bran on cholesterol parameters. Dosages of 25, 50, 100 and 200mg/d were administered to participants following the restricted AHA step 1 diet.	Dosages of 100mg/d of TRF25 produced a 20% decrease in total cholesterol, 25% decrease in LDL-C,14% decrease in Apolipoprotein B and 12% reduction in triglycerides compared to baseline values.
Berger *et al.*, 2005	30 mildly hypercholesterolaemic men	Evaluation of RBO with high (0.8g.d) and low (0.05g/d) gamma oryzanol compared to peanut oil in a randomized controlled parallel design study	Low and high gamma oryzanol RBO lowered total plasma cholesterol by 6.3%, LDL-C: HDL-C ratio by 18.9% and were not significantly different from each other. Peanut oil with a similar FA profile did not alter lipid parameters
Most *et al.*, 2005	27 healthy men and women	Randomised cross-over 10 week feeding study to assess the effects of defatted rice bran oil in an average American diet on blood lipids in midly hypercholesterolaemic individuals.	RBO did not alter lipid concentrations compared with a low fibre control diet but RBO decreased LDL-C by 7% without affecting HDL-C when compared to a diet of similar fat content.

Chapter III

MATERIAL AND METHODS

The present study "Effect of Rice Bran Oil spread (RBOS) on the physical and sensory quality of baked products" was conducted in Department of Foods and Nutrition, Post Graduate and Research Center, Professor Jayashankar Telangana State Agricultural University, Rajendranagar, Hyderabad during the year 2014 - 2016.

This chapter includes detailed description of experimental procedure of the study under the following heads.

3.1 Procurement of raw material

3.2 Standardization and development of baked products

3.3 Analysis of physical quality characteristics

3.4 Estimation of fatty acids and trans fatty acids

3.5 Microstructure analysis of cake by using Scanning Electron Microscope (SEM) imaging

3.6 Sensory evaluation

3.7 Consumer evaluation

3.8 Statistical analysis

Protocol followed for the study is represented below

3.1 Procurement of raw material

Raw materials such as hydrogenated fat, rice bran oil, refined wheat flour, sugar were procured from the local market. Equipments required for preparation of the baked products were utilized from the bakery laboratory of Post Graduate & Research Centre, PJTSAU, Rajendranagar, Hyderabad. Two types of Rice Bran Oil Spread viz. RBOS1 and RBOS2 were procured from the Indian Institute of Rice Research (Formerly Directorate of Rice Research Centre) for the product development of baked products. Rice bran oil spread (RBOS) used in the present study was prepared by mixing selected edible grade gelator with 100g of rice bran oil. The resulting mixture was heated in a

microwave for five minutes. When the gelator got dissolved in the oil, it was allowed to cool at room temperature for 24 hours which yielded transparent gel. Two different RBOs were prepared with two compositions of edible grade gelator and were used in the study as RBOS1 & RBOS2. RBOS1 was made with higher concentration of edible grade gelator which yielded a hard gel, where as RBOS2 was made with lower concentration of edible grade gelator which yielded a soft gel.

(a) RBOS1 **(b) RBOS2**

Figure 3.1 Images of Rice Bran Oil Spread (RBOS)1 and 2

3.1.1 Chemicals and glassware

All chemicals used in the investigation were of analytical grade. Chemicals and glassware were utilized from the laboratory of Post Graduate and Research Centre, PJTSAU, Rajendranagar, Hyderabad.

3.2 Standardization and development of baked products

Various baked products like cakes, cookies, pie crusts, muffins and doughnuts were prepared with hydrogenated fat (Dalda), Rice Bran Oil (RBO) and 2 variations of Rice Bran Oil Spread (RBOS1 and RBOS2). Products prepared from hydrogenated fat served as control; products made by incorporating RBO, RBOS1 and RBOS2 served as experimental samples. The ingredients, processing conditions, process parameters were standardized after several trails in the bakery laboratory of the department.

Table 3.1 Preparation of cake

Ingredients	Quantity
Refined wheat flour	115g
Sugar	115g
Hydrogenated fat/RBO/RBOS1/RBOS2	60g
Baking powder	1/4tsp
Essence	2-4 drops
Eggs	2
Milk	To make up the consistency

Method:

- Flour and baking powder were sieved together.
- Fat and sugar were creamed till light and fluffy.
- Eggs were beaten and essence was gradually added to the mixture, mixing all the time with a rubber scoop.
- The above mixture was folded in sieved flour very lightly and a little milk was added to make batter consistency.
- This mixture was spread on the sprayed sheet and tapped it little to spread evenly and baked in a preheated oven for 22 to 25 minutes and at 190ºC.

Figure 3.2 Cake prepared with HF (control), RBO, RBOS1 and RBOS2

Table 3.2 Preparation of cookies

Ingredients	Quantity
Refined wheat flour	200g
Sugar powder	80g
Hydrogenated fat/RBO/RBOS1/RBOS2	120g
Baking powder	1/4tsp
Custard powder	10g
Milk powder	10g
Milk	20ml
Cardamom Powder	1/2tsp

Method:

- Flour and baking powder were sieved together.
- Fat, sugar and milk were creamed till light and fluffy.
- Refined flour, custard powder, milk powder, cardamom powder and baking powder were added together and it was mixed well with above mixture and a pliable dough was made.
- The dough was rolled out and cut with a biscuit cutter and baked in a preheated oven at 195ºC for 15 minutes.

(a) Cookies prepared with HF **(b) Cookies prepared with RBO**

(c) Cookies prepared with RBOS1 **(d) Cookies prepared with RBOS2**

Figure 3.3 Images of control (HF) cookies, RBO, RBOS1 and RBOS2 cookies.

Table 3.3 Preparation of Piecrust

3.3 a Preparation of Crust

Ingredients	Quantity
Refined wheat flour	350g
Hydrogenated Fat/RBO/RBOS1/RBOS2	226g
Sugar	30g
Ice cold water	100ml
Salt	1teaspoon

Method:

- The flour, salt and sugar were placed in a bowl and processed with a hand blender until combined. Butter was added and processed until the mixture resembled coarse meal (about 15 seconds).
- 60ml of ice cold water was added to the above mixture until the dough just holds together when pinched and it was not processed more than 30 seconds.
- The dough was divided in half, flattening each half into a disk, covered with plastic wrap, and refrigerated for about one hour before using. This chilled the butter and relaxed the gluten in the flour.
- After the dough had chilled sufficiently one portion of the dough was removed from the fridge and placed it on a lightly floured surface. The pastry was rolled

into a circle. The dough was folded in half and gently transferred to a pie pan. The edges of the pastry were trimmed to fit the pie pan. It was covered with plastic wrap and placed in the refrigerator.

- The second round of pastry was removed and rolled it into circle. It was transferred to a parchment lined baking sheet, covered with plastic wrap and placed in the refrigerator.

(a) Piecrust prepared with HF

(b) Piecrust prepared with RBO

(c) Piecrust prepared with HF

(d) Piecrust prepared with RBO

Figure 3.4 Images of control (HF) piecrust, RBO, RBOS1 and RBOS2 piecrust

3.3 b Preparation of Apple pie

Ingredients	Quantity
Apples(sliced)	1.1kg
Sugar	1/4gm
Lemon juice	1 table spoon
Cinnamon	1 tea spoon
Salt	¼ tea spoon
Corn starch	15gm

Apple Filling:

- The sliced apples with the sugar, lemon juice, ground cinnamon, nutmeg and salt were combined in a large bowl. It was kept at room temperature for 30 minutes.
- The top pastry crust from the refrigerator was removed and kept at room temperature for about 10 minutes. Apples slice mixture was transferred to a large bowl and mixed them with the cornstarch (corn flour). Then the reduced syrup was poured over the apples and tossed to combine. The apples and their syrup was poured into the chilled pie crust. The edges of the pie shell was moistened with a little water and then the top crust was placed over the apples. The edges were crimped using a fork. Slits were made by using a knife from the center of the pie out towards the edge of the pie to allow the steam to escape. The pie was covered with plastic wrap and placed in the refrigerator to chill the pastry.
- The oven was preheated to 220° C and the pie was baked for about 45 to 55 minutes until the juices start to bubble through the slits and the apples felt tender (not mushy) when a toothpick or sharp knife was inserted through one of the slits. The pie was removed from the oven and placed on a wire rack to cool for about 3-4 hours.

Table 3.4 Preparation of muffins

Ingredients	Quantity
Refined wheat flour	100g
Sugar Powder	100g
Hydrogenated fat/RBO/RBOS1/RBOS2	100g
Baking powder	1/4tsp
Vanilla essence	2-4 drops
Eggs	2
Tutti-fruity	10g

Method:

- Preheat the oven to 375°F.
- Flour and baking powder were sieved together
- The flour, baking powder and sugar were mixed in a large bowl.

- Eggs, fat and vanilla essence were stirred only enough to dampen the flour and spooned into the muffin pans, filling each cup about two-thirds full.
- Tutti fruity was added to the above mixture and was baked for about 20 to 25 minutes each

(a) Muffin prepared with HF **(b) Muffin prepared with RBO**

(c) Muffin prepared with RBOS1 **(d) Muffin prepared with RBOS2**

Figure 3.5 Images of control (HF) muffin, RBO, RBOS1 and RBOS2 muffins

Table 3.5 Preparation of Doughnut

Ingredients	Quantity
Refined wheat flour	200g
Sugar	40g
Hydrogenated fat/RBO/RBOS1/RBOS2	30g
Fresh yeast	10g
Salt	2.5g
Eggs	1
Milk	100ml
Oil	500ml

Method:

- Yeast, salt, sugar and milk were put in an enamel bowl.
- Egg was beaten into the above mixture and mixed it well.
- Flour and butter were added and mixed well.
- The dough was kneaded well till smooth.
- An enamel bowl was greased and placed the dough in it. The bowl was covered with a wet cloth and kept for fermentation for half an hour. After fermentation, the dough was made into smooth balls and rolled into a disc of ¼ inch thickness; then cut into circles with a doughnut cutter and kept them for proofing for 15-20 minutes.
- Doughnuts were deep fried in smoking oil till golden brown.
- The fried doughnuts were rolled in ground sugar.

(a) Doughtnut made with HF

(b) Doughnut made with RBO

(c) Doughnut made with RBOS1

(d) Doughnut made with RBOS2

Figure 3.6 Images of control (HF) doughnut, RBO, RBOS1 and RBOS2 doughnuts

3.3 Analysis of physical quality characteristics

3.3.1 Estimation of Moisture

The moisture content of the samples was determined by the method of AOAC (2005). A petri dish was weighed with lid (W_1) and approximately 10g of sample was weighed into it and weighed again (W_2). Petri dish along with sample was dried in an oven at 100-105ºC for 15-17hrs and cooled in a desiccator and weight was noted (W_3). The same process was repeated till constant weight was obtained.

Calculation:

$$\textbf{Moisture content, \% by mass} = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

Where

W_1 = mass of empty petri dish (grams)

W_2= mass in of petri dish with the material before drying (grams)

W_3 = mass in of petri dish with the material after drying (grams)

3.3.2 Estimation of Cookie height

Cookie height was taken in the center of each cookie using a vernier calipers.

3.3.3 Estimation of Percentage Shrinkage

Percent shrinkage was determined based on the area change in each pastry strip as measured before and after baking (Dreher *et al*., 1983).

3.3.4 Estimation of Crust height

Crust height and highest point were taken as an average of 4 readings (taken at the center points on each side of the crust strip) and the highest point on the same crust with a digital caliper (Dreher *et al*., 1983).

3.3.5 Estimation of Oiliness

Oiliness of the crust was taken as the weight difference of the parchment paper liner before baking and after baking, where 7-8 strips were baked on a single pan.

3.3.6 Estimation of Density (g/cm^3)

A representative sample of the finished cake (approximately $1cm^3$) was cut from the center of the loaf and weighed to determine its density.

3.3.7 Estimation of Bulk density (g/cm^3)

- The bulk densities of baked muffins were calculated by taking the ratio of a known amount of muffin to its volume.
- The volume of baked muffin samples was determined by the rape seed displacement method as described by Rahman (1995).
  - The volume of a container was measured by filling the container using the rapeseeds.
  - Then, the cake was placed into the container and followed by refilling the rapeseed into the container.
  - The cake displaced the volume of seed which is equivalent to its own volume

3.3.8 Estimation of dough density

- Dough density was measured by weighing an individual portion of dough (W) (a scoop of approximately 1 ml).
- The exact volume of each portion was determined by reading the volume of water that was displaced in a graduated cylinder.
- It was then calculated from these measurements as g/cm^3

3.3.9 Estimation of Cookie spread

- The spread of the finished cookies were measured using AACC Official Method 10-50 D (1983).
- Cookies were allowed to cool thoroughly for about 2min, then centered over a template with concentric rings placed 2mm apart.
- The spread was calculated as the average distance from the center as read in each of 4 quadrants.

3.3.10 Estimation of volume and specific volume

- Volume of the doughnut was measured using rapeseed displacement method.
- Doughnut samples were put in a metallic box with known volume (V).

- The box was topped up with rapeseed, the doughnuts were removed and the volume of the rapeseed was noted (VR). Volume (VL) was then calculated according to the formula given below.

  Calculation

$$VL\ (ml) = V - VR.$$

- Then doughnut was weighed, W (g).
- Specific volume (Vs) of doughnuts was calculated according to the formula given below:

$$Vs\ (ml/g) = VL/W$$

3.4 Estimation of fatty acids and trans fatty acids

3.4.1 Estimation of Total fat

Total Fat content of samples was estimated by following standard procedure of AOAC, 19th Edition (2003.06) using Petroleum Ether for the extraction of fat by Gerhardt soxtherm fat analyser.

Procedure

- The empty fat extraction beaker was weighed along with boiling stones (W_1)
- Approximately 2g of samples were weighed and packed into a filter paper thimble, and placed the thimbles in extraction coloumns provided with the instrument.
- 150ml of petroleum ether (60-80°C) was taken in a beaker
- Required conditions for soxtherm extraction were maintained.
- The thimbles in which sample was placed was inserted into the slot provided for thimbles.
- Thimble containing the test portion was immersed into the boiling solvent. The intermixing of the matrix with hot solvent ensured rapid solubilization of extractables.
- In the 2nd Step, the thimble was raised above the solvent and the test portion was further extracted by a continuous flow of condensed solvent. The solvent was evaporated and recovered by condensation.
- The resulting crude fat residue was determined gravimetrically after drying.
- Extraction cups were dried at 102°C ± 2°C in a hot air oven for 30min to remove moisture and cooled in a dessicator. Excessive drying was avoided, which can oxidize fat and give erroneous results.

- After cooling in a dessicator the beakers were weighed to the nearest 0.1mg (F).
- Fat/oil extracted by soxhlet method described in this section was used for methyl ester preparation.

Calculation

$$\% \text{ Crude fat, petroleum} = \frac{F-T}{S} \times 100$$

Where

F= Weight of cup + fat residue (g)

T= Weight of empty cup (g)

S= Test portion weight (g)

3.4.2. Preparation of fatty acid methyl esters

Fatty acid methyl ester was prepared by following procedure given below

- 18.00g of KOH was weighed and added to 10ml of water, then diluted with methanol to 500ml, and allowed to stand for 24hours, and filtered into a polyethylene bottle.
- 50mg of oils/ lipids that were extracted as described in section 3.4.1 were reconstituted in 1ml of 0.5M methonolic KOH and hydrolyzed at 80°C for 1 hour in a water bath.
- 1ml of fresh 10% BF3 in methanol was added and *trans* esterification performed at 100°C for 20minutes in a water bath.
- After trans esterification, 2ml distilled water and 1ml hexane were added to the sample to quench the reaction.
- The organic phase was recovered by pippeting out the upper layer, pooled and spiked with the methyl ester.

3.4.3 Estimation of fatty acids and trans fatty acids

Fatty acids were estimated by GC, 7890B of Agilent Technologies with 7693 auto sampler, equipped with Flame Ionization Detector (FID) and split injector. Injector temperature was set at 260°C and samples were injected (1µl) with split ratio of 10:1 by the auto sampler. Carrier gas (Nitrogen) flow rate was set at 30ml/min. Column used was Agilent - DB-FFAP which is a nitroterephthalic-acid-modified polyethylene glycol

(PEG) column of high polarity for the analysis of volatile fatty acids and phenols, with a length 30m X 250µm, 0.25mm diameter, 0.25µm film thickness. The temperature program was at set with an initial temperature of 100°C, hold time, 5min, rising at an increasing rate to 240°C at the rate of 4°C /min and held for 5min. Total run time was 45min. Nitrogen was used as carrier gas at a column flow rate of 1.0ml/min. Detector temperature was at 280°C. EZ Total Chrome software was used for running the GC and calculation of fatty acid composition. FID Hydrogen gas flow rate was 30ml/min. Zero air flow was 300ml/min and make up flow was 25ml/min. The fatty acid content was measured based on area normalization.

Standards used were 4*7885-U* Supelco® 37 Component FAME Mix, 10mg/ml in methylene chloride (varied), analytical standard. For individual trans-fatty acids standards, Supelco trans-9-Eliadic methyl ester, 10mg/ml in heptane, trans-9, 12-Octadecadienoic (linoleliadic) methyl ester and trans-11-Vaccenic methyl ester, were used. After injecting the 37-FAME standard, individual trans-fatty acid methyl ester standards were also injected and the retention times were compared under standard conditions described above to ascertain that the individual standard peaks were coinciding exactly with the peaks in the combined standard.

Samples were processed as described in section 3.4.2 and injected as for standards. Sample fatty acid composition was compared with standard fatty acid composition and percentages calculated by normalization of peak areas

3.5 Microstructure analysis of cake by Scanning Electron Microscope (SEM) imaging

The most accepted cake sample (RBOS2) and the control cake sample (hydrogenated fat) were viewed and photographed on a Scanning Electron Microscope (SEM) (JOEL model JSM 6360LV) operating with magnification from 100× to 1000× to study the porosity and internal appearance of cake which related to expandable characteristics of products. For microstructure study, cake samples were mounted over the stubs with double-sided conductivity tape and applied a thin layer of gold over the samples using an automated sputter coater for about 3 min (Bozzola and Russell, 1999). The samples were scanned in a scanning electron microscope at various magnifications at RUSKA Lab, College of Veterinary Sciences, PV Narsimha Rao Veterinary University, Hyderabad, India.

3.6 Acceptability of the product

The acceptability of the developed products were evaluated by sensory evaluation (Meilgard *et al.*, 1999).

3.6.1 Sensory evaluation

The sensory assessments were conducted in a purpose-built, six-booth sensory evaluation laboratory. A panel of 30 members consisted of staff and graduate students of the Department of Foods and Nutrition, Post Graduate and Research Centre, Professor Jayashankar Telangana State Agricultural University and Indian Institute of Rice Research, Rajendranagar, Hyderabad. The panelists had no knowledge of the project objectives. All the five samples i.e. cake, cookies, muffins, piecrusts and doughnuts (prepared with hydrogenated fat, RBO, RBOS1 and RBOS2) were coded using random three-digit numbers and served with the order of presentation counter-balanced. Panelists were provided with a glass of water and instructed to rinse and swallow water between samples. They were given written instructions and asked to evaluate the products for acceptability based on its appearance color, texture, taste, flavor, and overall acceptability using nine point hedonic scale (0 = Dislike extremely to 9 = Like extremely as described by (Meilgard *et al.*, 1999). The score card developed is presented in Appendix A.

Figure 3.7 sensory evaluation

Figure 3.8 Consumer evaluation

3.7. Consumer evaluation

According to the results of sensory evaluation, the best acceptable product was found to be Cake (on 9 point hedonic scale), which was selected for the

consumer acceptability evaluation. A total of 100 subjects were selected randomly for the consumer acceptability evaluation which was conducted at the Department of Foods & Nutrition, Post Graduate & Research Centre, PJTSAU, Hyderabad and Indian Institute of Research (Formerly Directorate of Rice Research), Rajendranagar, Hyderabad by using a consumer acceptability questionnaire consisting of two sections describing the demographic profile and the hedonic scoring for cake. For consumer acceptability study, cake samples were freshly prepared and served to the consumers. They were given written instructions and asked to evaluate the products for acceptability based on its color, texture, sponginess, flavour, taste and overall acceptability by using five-point hedonic scale (1= Disliked very much to 5= Like very much). The consumer acceptability questionnaire is given in Appendix B.

3.8. Statistical analysis

Mean and standard deviation for four parallel replicates were calculated. Least significant difference (LSD) method was used to make pair wise comparisons between quantitative variables. The difference within each level under each variable was tested at a 95% confidence interval. General linear model (GLM) procedure in Statistical Analysis System Software (SAS version 9.1, Statistical Analysis System Institute, Inc. Cary, NC) was used for all the analysis. ANOVA was calculated for all the attributes of sensory evaluation to test the significance (Fisher and Yates, 1963).

Chapter IV

RESULTS AND DISCUSSION

The present investigation was conducted to study the "Effect of Rice Bran Oil Spread (RBOS) on the physical and sensory quality of baked products." The data was subjected to statistical analysis and the results obtained are presented in this chapter under the following headings

4.1 Physical quality characteristics of baked products
- 4.1.1 Cakes
- 4.1.2 Cookies
- 4.1.3 Piecrust
- 4.1.4 Muffins
- 4.1.5 Doughnuts

4.2 Sensory evaluation of baked products
- 4.2.1 Cakes
- 4.2.2 Cookies
- 4.2.3 Piecrust
- 4.2.4 Muffins
- 4.2.5 Doughnuts

4.3 Consumer acceptability

4.4 Scanning Electron Microscope (SEM) imaging of cake

4.5 Fatty acids composition of cake
- 4.5.1 Total fat content of cake
- 4.5.2 Saturated fatty acids content of cake
- 4.5.3 Unsaturated fatty acids content of cake
- 4.5.4 Trans fatty acids content of cake

4.1 Physical quality characteristics of baked products

The results of physical quality analysis data of cakes prepared with Hydrogenated Fat (HF), Rice Bran Oil (RBO), Rice Bran Oil Spread1 (RBOS1) and Rice Bran Oil Spread2 (RBOS2) are given in Table 4.1.

4.1.1 Cakes

Cake moisture: Results indicated that the moisture content was high in HF cake (16.035±0.36), when compared with RBO cake (14.152±0.09), RBOS1 cake (8.924±0.43) and RBOS2 cake (11.305±0.37). It was found that there was no significant difference between the moisture content of cake made with RBO, RBOS1 and RBOS2. However, there was a significant ($p<0.01$) difference among the moisture content of control and experimental RBOS (1 and 2) cakes.

Cake density: Cake density was higher in RBO (0.397±0.01) cake followed by RBOS2 (0.390±0.01) cake. As per the results, there was no significant difference between hydrogenated fat and RBOS1 cakes; RBO and RBOS2 cakes. However, there was a significant difference ($p<0.01$) between the density of control cake sample and experimental samples (RBO and RBOS2).

Table 4.1 Physical parameters of cake

Parameters	HF	RBO	RBOS1	RBOS2	SE value	F value
Cake moisture (%)	16.035± 0.36 [a]	14.152± 0.09[b]	8.924± 0.43 [b]	11.305± 0.37 [b]	0.19643	21.34**
Cake density (g/cm^3)	0.330± 0.01 [b]	0.397± 0.01 [a]	0.280± 0.01 [b]	0.390± 0.01 [a]	0.00687	64.06**

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, HF – Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread1; RBOS2 – Rice Bran Oil Spread2.

Studies conducted by NorAini *et al.* (1992); Stauffer (1998) and Zhou *et al.* (2011) suggests that the density of the final cake depends not only on the initial air incorporation, but also on the stability of the air bubbles in batter during baking. As the oven is heated up, bubbles tend to migrate and expand, while lipid crystals (adsorbed to the air bubbles during mixing) melt and form a uniform layer over the inside surface of the bubble, allowing bubble expansion without rupture (Brooker, 1993; Zhou *et al.* 2011) resulting in a stable bubble. Rutkowska and Zbikowska (2010) reported that liquid margarines could be recommended for cakes in large-scale manufacture and for reducing the intake of TFA in processed foods, it is justified to replace hydrogenated vegetable fat by liquid fats for manufacture of bakery products.

4.1.2 Cookies

The results of physical analysis of cookies prepared with Hydrogenated Fat (HF), Rice Bran Oil (RBO), Rice Bran Oil Spread 1 (RBOS1) and Rice Bran Oil Spread 2 (RBOS2) are given in Table 4.2.

Table 4.2 Physical parameters of cookie

Parameters	HF	RBO	RBOS1	RBOS2	SE value	F value
Cookie moisture (%)	1.255± 0.01 [a]	1.280± 0.01 [a]	1.115± 0.03 [b]	1.071± 0.05 [b]	0.02224	21.34**
Cookie height (cm)	1.107± 0.00 [a]	1.057± 0.01 [b]	1.033± 0.01 [c]	1.100± 0.01 [a]	0.00569	38.03**
Cookie dough density(g/cm^3)	1.090± 0.07 [bc]	1.163± 0.02 [ab]	1.233± 0.02 [a]	1.063± 0.04 [c]	0.02141	12.81**
Cookie spread (cm)	0.347± 0.00 [a]	0.350± 0.00 [a]	0.207± 0.00 [b]	0.340± 0.02 [a]	0.00660	95.66**

Note: Values are expressed as Mean ± SD, *significant at 5% level, ** significant at 1% level, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

Cookie moisture: As per the results obtained, it was found that the moisture content was high in RBO cookies (1.280±0.01) followed by control cookies (1.255±0.01). The moisture content in RBOS1 cookies was 1.115±0.03 and RBOS2 cookies was 1.071±0.05. It was also found that, there was no significant difference between the moisture content of cookies made with HF and RBO; RBOS1 and RBOS2. However, there was a significant difference ($p<0.01$) between the moisture content of control and experimental RBOS (1 & 2) cookies.

Cookie height: The cookie height was significantly ($p<0.01$) higher in HF cookies (1.107±0.00) and RBOS2 cookies (1.100±0.01) as compared to RBO cookies (1.057±0.01). The cookie made with RBOS1 (1.033±0.01) had significantly lower ($p<0.01$) cookie height as compared to all other cookies.

Cookie dough density: The results of cookie dough density indicate that the density was significantly ($p<0.01$) higher in RBOS1 cookies (1.233±0.02) followed by RBO cookies (1.163±0.02) followed by HF cookies (1.090±0.07) followed by RBOS2 cookies (1.063±0.04). Hard consistency of RBOS1 fat than the other fats used in this study could have led to higher cookie dough density in RBOS1 cookies.

Cookie dough containing a chemical leavening agent, starts to expand around 30°C due to the formation and expansion of leavening gasses (Doescher *et al.,* 1987). As with cakes, lipid with the right melting temperature is necessary for expansion of bubbles without rupture (NorAini and Miskandar., 2007). Therefore, lipids that begin melting too early in the baking process (before 30°C) are not able to adsorb to the bubbles, which allows air to escape before dough expansion can begin, resulting in bubble rupture and lower cookie height.

Cookie spread: As per the results obtained, it was seen that the cookie spread was similar in HF cookies (0.347±0.00), RBO (0.350±0.00) and RBOS2 (0.340±0.02) cookies, but significantly ($p<0.01$) lower in RBOS1 cookie (0.0207±0.00). The results of present study are in concurrence with the findings of Jacob and Leelavathi (2007), who reported that cookies containing rice bran oil had relatively higher spread value, while the cookies containing the non-emulsified hydrogenated fat (Dalda) had least spread value.

Pareyt and Delcour (2008), reported that the degree of spread resulting in the final cookie diameter is controlled by the spread rate and set time of the dough. Shortening functions as a lubricant in cookie dough, thus affecting spread rate: more shortening/more liquid oil portion gives a larger spread.

Generally, expansion due to leavening and the flow rate of the cookie dough, affect cookie spread (Hoseney and Rogers, 1994). Cookie spread in turn affects cookie height. After cookie dough is put in the oven, it starts to expand until it reaches its setting time (the point at which the dough stops spreading due to protein coagulation and starch gelatinization). Previous research suggests that lipid type is not an essential variable for cookie spread (Rogers, 2004; Vetter *et al.,* 1984). However, it does affect cookie spread by providing a lubricant effect, which in turn affects flow rate (Pareyt *et al.,* 2010). Basically, the earlier the lipid melts during the baking process, the larger the cookie spread and the flatter cookie will be. Therefore, cookies made from liquid oils, such as RBO used in this study, tend to have larger spreads and lower heights.

4.1.3 Piecrust

The results of physical analysis of piecrusts prepared with Hydrogenated Fat (HF), Rice Bran Oil (RBO), Rice Bran Oil Spread 1 (RBOS1) and Rice Bran Oil Spread 2 (RBOS2) are given in Table 4.3.

In piecrust production, the role of lipid is to prevent excessive gluten formation and create pockets in the dough (Stauffer, 1998). Gluten in dough is formed by the interaction of flour and water (Patient, 1994). Lipid with a more fluid character tends to limit gluten development, since flour particles are covered by lipid and have less chance to interact with water (Pyler and Merriam, 1988). However, solid lipids are more likely to be coated by flour particles than vice versa and form pockets of lipids within the dough. Therefore, flour proteins are able to interact with water, leading to increased gluten development. The majority of gluten is formed when water is added in the final stages of mixing, after the lipid and flour have been combined. Additionally, lipids can create and maintain pockets in dough during rolling and baking. During the cooking of pie crust, as temperature in the oven increases the pockets of lipid dispersed throughout the dough melt and leak out of the baking matrix leaving a gap, allowing air and steam to accumulate and expand. The size and number of these pockets directly influence the final flakiness and height of the baked pie crust. Breaking strength, an indicator of pie crust toughness, is directly influenced by gluten development, with more gluten development resulting in a tougher crust (Hirahara and Simpson, 1961).

Table 4.3 Physical parameters of Piecrust

Parameters	HF	RBO	RBOS1	RBOS2	SE value	F value
Pie crust moisture content (%)	5.247± 0.19 ^{a}	5.160± 0.24 ^{ab}	4.723± 0.485$^{a\ b}$	4.650± 0.35 ^{b}	0.15566	3.76*
Height point (cm)	7.407± 0.14 ^{a}	6.523± 0.28 ^{c}	6.953± 0.14 ^{bc}	7.263± 0.17 ^{ab}	0.12733	9.43*
Oiliness (%)	0.117± 0.03 ^{a}	0.120± 0.02 ^{a}	0.073± 0.02 ^{b}	0.063± 0.02 ^{b}	0.01266	5.32*
Crust height (cm)	3.133± 0.05 ^{a}	2.867± 0.05 ^{b}	3.133± 0.05 ^{a}	3.033± 0.05 ^{a}	0.03727	11.40**
% Shrinkage	9.200± 0.20 ^{d}	11.333± 0.15 ^{b}	14.267± 0.20 ^{a}	10.300± 0.20 ^{c}	0.11980	330.03**

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

Piecrust moisture: In the present study the results indicated that the moisture content of piecrust was significantly ($p<0.05$) highest in HF piecrust (5.247±0.19) followed by RBO piecrust (5.160±0.24), RBOS1 piecrust (4.723±0.485) and RBOS2 piecrust (4.60±0.35) respectively. It was also observed that there was no significant difference in

the moisture content of piecrusts made with HF, RBO and RBOS1; RBOS1 and RBOS2. However, there was a significant ($p<0.05$) difference between the moisture content of piecrust made with HF and RBOS2.

Height point: The height point was significantly ($p<0.05$) higher in HF piecrust (7.407±0.14) and RBOS2 piecrust (7.263±0.17) as compared to RBOS1 piecrust (6.953±0.14). The height point was significantly ($p<0.05$) lower in RBO piecrust (6.523±0.28) as compared to all other piecrusts. As RBO was a liquid fat, no air pockets were formed during kneading and so expansion could not occur during baking. Therefore, pie crust made with RBO could have had the lowest height point.

Oiliness: As per the results obtained it was observed that there was no significant difference between the oiliness of piecrust made with HF (0.117±0.03) and RBO (0.120±0.02); RBOS1 (0.073±0.02) and RBOS2 (0.063±0.02). However, the oiliness in control was significantly ($p<0.05$) higher than the experimental samples RBOS (1 and 2), indicating decreased oil absorption in the RBOS1 and RBOS2 fats.

Crust height: The results of crust height was similar in HF pie crust (3.133±0.05), RBOS1 pie crust (3.133±0.05) and RBOS2 pie crust (3.033±0.05) but significantly ($p<0.01$) lower in RBO pie crust (2.867±0.05). As RBO was a liquid fat, no expansion could occur during baking, resulting in lower crust height and height point.

Percentage shrinkage: As per the results obtained, it was found there was a significant ($p<0.01$) difference in percentage shrinkage of piecrusts made with HF (9.200±0.20), RBO (11.333±0.15), RBOS1 (14.267±0.20) and RBOS2 (10.300±0.20).

4.1.4 Muffins

The results of physical analysis of muffins prepared with Hydrogenated Fat (HF), Rice Bran Oil (RBO), Rice Bran Oil Spread 1 (RBOS1) and Rice Bran Oil Spread 2 (RBOS2) are given in Table 4.4.

Muffins moisture: As per the results obtained, it was seen that the moisture content of muffin was similar in HF muffin (15.484±1.09) and RBO muffin (16.037±0.18), but significantly ($p<0.01$) lower in RBOS2 muffin (13.874±0.70) and it was also observed that, there was no significant difference between the moisture content of muffins made with Hydrogenated fat and RBO. The moisture content in muffins made with RBOS1

(17.546±0.18) was significantly ($p<0.01$) highest as compared with the other three muffins.

Muffins bulk density: The muffin bulk density was significantly ($p<0.01$) higher in RBOS2 muffin (0.423±0.01) as compared to HF muffins (0.347±0.03), RBO muffins (0.253±0.01) and RBOS1 (0.257±0.01) muffins. The muffin density was lower in RBO muffin and RBOS1 muffin as compared to other muffins. Sharif *et al.* (2005) studied the development of improved quality of baked products by replacing bakery shortening with rice bran oil at 25, 50, 75 and 100% levels and reported similar results. Kaur *et al.* (2014) studied replacement of bakery shortening with refined rice bran oil in the preparation of muffins. Rice bran oil was found to have a higher content of the essential fatty acid linoleic acid (34.98%) as compared to that of bakery shortening (5.14%).

Table 4.4 Physical parameters of muffin

Parameters	HF	RBO	RBOS1	RBOS2	SE value	F value
Muffins moisture content (%)	15.484± 1.09 ^{b}	16.037± 0.18 ^{b}	17.546± 0.18 ^{a}	13.874± 0.70 ^{c}	0.35529	18.22**
Muffins Bulk density (g/cm^3)	0.347± 0.03 ^{b}	0.253± 0.01 ^{c}	0.257± 0.01 ^{c}	0.423± 0.01 ^{a}	0.00714	129.89**

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

4.1.5 Doughnuts

The results of physical analysis of doughnuts prepared with Hydrogenated Fat (HF), Rice Bran Oil (RBO), Rice Bran Oil Spread 1 (RBOS1) and Rice Bran Oil Spread 2 (RBOS2) are given in Table 4.5.

Doughnut moisture content: Results indicated that a significant ($p<0.05$) difference existed in the moisture content between RBO doughnut (13.393±0.97) and HF doughnut (12.007±0.71), RBOS1 doughnut (11.243±0.42) and RBOS2 doughnut (11.197±0.10). There was no significant difference between HF doughnuts, RBOS1 doughnuts and RBOS2 doughnuts.

Doughnut volume: The doughnut volume was significantly ($p<0.05$) lower in RBOS1 doughnut (41.867±0.75) as compared to HF doughnut (44.167±0.90), RBO doughnut

(43.100±0.10) and RBOS2 doughnut (44.333±0.41). It was found that there was no significant difference in the volume of doughnuts made with hydrogenated fat, RBO and RBOS2 fats. However, RBOS2 fat was found to give highest volume to the doughnut indicating that, RBOS2 can be used in place of other fats to get good volume and thereby texture to the doughnuts.

Doughnut specific Volume: As per the results obtained, it was observed that there was no significance difference between the specific volume of doughnuts made with HF (7.800±0.43), RBO (7.427±0.12) and RBOS2 (7.510±0.09). However, there was a significance ($p<0.05$) increase in the specific volume of doughnuts made with RBOS1 (8.400±0.10).

Table 4.5 Physical parameters of doughnut

Parameters	HF	RBO	RBOS1	RBOS2	SE Value	F Value
Doughnut moisture content (%)	12.007± 0.71 [b]	13.393± 0.97 [a]	11.243± 0.42 [b]	11.197± 0.10 [b]	0.34660	8.75*
Doughnut volume(cm^3)	44.167± 0.90 [a]	43.100± 0.10 [a]	41.867± 0.75 [b]	44.333± 0.41 [a]	0.41051	7.71*
Doughnut Sp. Vol (cm^3)	7.800± 0.43 [b]	7.427± 0.12 [b]	8.400± 0.10 [a]	7.510±0. 09 [b]	0.14204	9.62*

Note: Values are expressed as Mean ± SD, *significant at 5% level, ** significant at 1% level, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

During frying, moisture is lost from the doughnut due to evaporation, while oil is absorbed into the crust (Bouchon *et al.*, 2003; Shih *et al.*, 2005). As frying time increases, oil uptake and moisture evaporation increase (Tan and Mittal, 2006). Doughnuts that are fried for a shorter period of time have shown to have lower oil uptake and moisture loss, than doughnuts fried for a longer period of time at the same temperature (Velez-Ruiz and Sosa-Morales, 2003). Because mass change is dependent on oil uptake and moisture loss, lower moisture loss and oil uptake results in lower mass change. Since the RBOS2 doughnuts were fried for shorter period of time compared to the control, which was fried for longer period of time, they had lower oil uptake and higher moisture content than the control.

4.2 Sensory Evaluation

4.2.1 Cake

The results of sensory evaluation of cake developed with HF, RBO, RBOS 1 and RBOS 2 are given in Table 4.6.

The results of colour of cake made with HF (7.800±0.775), RBO (7.867±0.743), RBOS1 (7.667±0.816) and RBOS2 (8.000±0.655) had no significant difference. Similarly, there was no significant difference in flavor of the cakes made with HF (7.400±0.737), RBO (7.533±1.125), RBOS1 (7.600±0.910) and RBOS2 (7.667±0.617). Similarly, the cakes made with HF (7.600±1.121), RBO (7.933±0.961), RBOS1 (7.533±0.743) and RBOS2 (8.000±0.655) did not vary significantly in taste.

Table 4.6 Sensory evaluation of cake

Sensory attributes	HF	RBO	RBOS1	RBOS2	F Value	SE Value	CD value
Colour	7.800± 0.775	7.867± 0.743	7.667± 0.816	8.000± 0.655	0.96^{NS}	0.141	0.404
Texture	7.000± 0.775^{b}	7.800± 1.014^{a}	7.200± 0.775^{b}	7.733± 0.594^{a}	6.53^{**}	0.154	0.440
Sponginess	7.467± 1.060^{ab}	7.667± 0.900^{a}	7.200± 0.941^{b}	7.800± 0.775^{a}	2.15^{*}	0.177	0.507
Taste	7.600± 1.121	7.933± 0.961	7.533± 0.743	8.000± 0.655	1.64^{NS}	0.182	0.521
Flavor	7.400± 0.737	7.533± 1.125	7.600± 0.910	7.667± 0.617	0.51^{NS}	0.160	0.457
Overall Acceptability	7.733± 0.799^{ab}	7.933± 0.961^{a}	7.467± 0.640^{b}	8.133± 0.516^{a}	3.90^{*}	0.144	0.411

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, NS- non significant, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread.

The texture of cake made with HF (7.000±0.775) and RBOS1 (7.200±0.775) cake; RBO (7.800±1.014) and RBOS2 (7.733±0.594) cake did not vary significantly. However, there was a significant ($p<0.05$) difference observed in the texture of cake made with HF (7.000±0.775) and RBOS2 (7.733±0.594).

The results indicated that there was no significant difference in sponginess of cake made with HF (7.467±1.060), RBO (7.667±0.900) and RBOS2 (7.800±0.775). The

cake made with RBOS2 was rated high on 9 point hedonic scale for sponginess, as compared to RBOS1 cake which was rated as lowest. Overall acceptability of the cake made with RBOS2 was rated high (8.133±0.516), as compared to other cakes indicating that the RBOS2 can be utilized for cakes preparation over other fat sources used in the study. Among all the cakes, RBOS2 cake was rated significantly higher on a 9 point hedonic score card indicating that though the colour, flavor, taste did not differ much, but the texture, sponginess and over all acceptability was definitely superior when made with RBOS2.

4.2.2 Cookies

The results of sensory evaluation of cookies developed with HF, RBO, RBOS1 and RBOS2 are given in Table 4.7.

Table 4.7 Sensory evaluation of cookies

Sensory attributes	HF	RBO	RBOS1	RBOS2	F value	SE Value	CD value
Colour	7.200± 0.941^{a}	6.733± 1.033^{b}	7.000± 0.756^{ab}	7.600± 0.737^{a}	2.95*	0.212	0.607
Texture	6.867± 0.640	6.600± 0.910	6.733± 1.100	7.067± 0.458	0.88^{NS}	0.212	0.606
Taste	7.133± 0.743^{ab}	6.267± 0.799^{c}	6.733± 0.799^{bc}	7.333± 0.617^{a}	7.00**	0.178	0.508
Flavor	6.733± 0.884^{ab}	6.333± 0.816^{b}	6.600± 0.632^{b}	7.133± 0.640^{a}	3.68*	0.173	0.495
Overall Acceptability	7.133± 0.640^{ab}	6.400± 0.632^{c}	6.933± 0.594^{b}	7.400± 0.507^{a}	8.41**	0.146	0.416

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, NS- non significant, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

As per the results obtained, it was seen that there was a significant difference ($p<0.05$) between the colour of the cookies made with HF, RBO, RBOS1 and RBOS2. The colour of cookies made with RBOS2 was rated high with a score of 7.600±0.737 on a 9 hedonic scale. The cookies made with HF was given a score of 7.200±0.941; RBOS1 was given a score of 7.000±0.756 and the least scored was for cookies made with RBO (6.733±1.033). It was found that there was no significant difference between the colour of cookies made with RBOS2, RBOS1 and HF. There was a significant decrease ($p<0.05$) in the colour score of RBO cookies compared to all other three

cookies. There was no significant difference between the texture of cookies made with HF, RBO, RBOS1 and RBOS2. RBOS2 cookies were given highest score (7.067±0.458) for texture compared to RBOS1 (6.733±1.100), RBO (6.600±0.910) and HF (6.867±0.640) cookies on 9 point hedonic scale.

The taste of cookies made with HF (7.133±0.743), RBO (6.267±0.799), RBOS1 (6.733±0.799) and RBOS2 (7.333±0.617) had significant differences ($p<0.01$) in the hedonic score. Similarly, the flavor of cookies made with HF, RBO, RBOS1 and RBOS2 had significant differences ($p<0.05$) in the hedonic score. However, the taste and flavor received significantly ($p<0.01$) higher score for cookies made with RBOS2 and least score for cookies made with RBO as compared to other cookies. There was no significant difference in the overall acceptability of cookies made with RBOS2 (7.400±0.507) and HF (7.133±0.640) as compared to cookies made with RBO (6.400±0.632) and RBOS1 (6.933±0.594). Overall acceptability of the cookies made with RBOS2 were rated high (7.400±0.507) as compared to other cookies indicating that the RBOS2 can be utilized for cookies preparation over other fat sources used in the study.

Rice bran, rice bran oil and honey were investigated as ingredients of biscuits. Effects on quality of biscuits were studied and an orthogonal experiment which was designed to obtain the optimal formula. Amount of rice bran was the main factor affecting biscuit quality. Use of 5% rice bran, 5% rice bran oil and 10% honey produced good biscuits with even yellow colour, a fragrant flavor and fluffy texture as reported by Chen *et al.*, 2003**.** In batters and doughs, mono- and diglycerides are added to shortening for forming better emulsions. Commercial hydrogenation generates the majority of the *trans* fats found in diet today. However, the negative effects of *trans* fatty acids in the diet have brought about changes to produce healthy spreads with reduced amounts of *trans* fatty acids (Mc. Williams, 2008).

Trans Fatty Acids are reported to have many negative effects on human health such as to increase the risk of coronary heart disease, which is marked by an increase in the ratio of total cholesterol to high density lipoprotein, and a raise in plasma triglyceride concentration (Tarrago-Trani *et al*., 2006; Dhaka *et al.*, 2011). Other health issues include increasing the level of lipoprotein, increasing the risk of breast and colon cancer, diabetes and obesity, and interfering with n-3 and n-6 fatty acid metabolism (Dhaka *et al*., 2011).

4.2.3 Piecrust

The results of sensory evaluation of piecrust developed with HF, RBO, RBOS1 and RBOS2 are given in Table 4.8.

The results of piecrust made from RBOS2 (8.000±0.655) had significantly highest rating in colour, texture, tenderness, taste, flavor and overall acceptability on a 9 point hedonic scale. It was also found that there was a significance difference ($p<0.05$) between the color, texture, tenderness and flavor made with HF, RBO, RBOS1 and RBOS2. Pie crusts made with RBO (7.133±0.516) received the lowest rating in overall acceptability, flavor, colour, taste and texture.

Table 4.8 Sensory evaluation of piecrust

Sensory attributes	HF	RBO	RBOS1	RBOS2	F Value	SE Value	CD Value
Colour	7.800± 0.561^{a}	7.200± 0.941^{b}	7.533± 0.516^{ab}	8.000± 0.535^{a}	4.18*	0.169	0.483
Texture	7.733± 0.458^{a}	6.933± 0.594^{b}	7.400± 0.986^{ab}	7.667± 0.724^{a}	3.85*	0.185	0.528
Tenderness	7.333± 0.617^{ab}	7.200± 0.775^{b}	7.133± 0.516^{b}	7.800± 0.676^{a}	2.72*	0.182	0.520
Taste	7.000± 0.535^{bc}	6.733± 0.884^{c}	7.267± 0.458^{ab}	7.733± 0.961^{a}	6.34**	0.169	0.483
Flavor	7.400± 0.507^{ab}	7.133± 0.640^{b}	7.467± 0.640^{a}	7.667± 0.617^{a}	1.90*	0.159	0.455
Overall Acceptability	7.533± 0.516^{bc}	7.133± 0.516^{c}	7.667± 0.488^{ab}	8.000± 0.655^{a}	6.24**	0.143	0.409

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, NS- non significant, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

In piecrust production, shortening and flour are first pinched together. Then water is added to form the crust dough. During this process, the role of lipid is to prevent excessive gluten formation and create pockets in the dough (Stauffer, 1998). Gluten in dough is formed by the interaction of flour and water (Patient, 1994). The majority of gluten is formed when water is added in the second stage of mixing, after the shortening and flour have been combined. Lipids with a more fluid character tend to limit gluten development, since flour particles are covered by lipid and have less chance to interact with water (Pyler and Merriam, 1988). However, solid lipids are more likely

to be coated by flour particles than vice versa, and form pockets of lipid within the dough. Therefore, flour proteins are able to interact with water, leading to increased gluten development and tougher pie crusts. Additionally, the solid portion in lipids can create and maintain pockets in dough during rolling and baking.

The lowest ratings of piecrust made with RBO might be due to its crumbly texture, which comes from its underdeveloped gluten as RBO is a lipid with more fluid character. The high ratings of pie crust made with RBOS2 might due to its tenderness. (Ghotra *et al*., 2002) reported that in pie crusts, lipids with a proper solid character and melting point help contribute to desirable flakiness and tenderness.

4.2.4 Muffins

The results of sensory evaluation of muffins developed with HF, RBO, RBOS1 and RBOS2 are given in Table 4.9.

Table 4.9. Sensory evaluation of muffin

Sensory attributes	HF	RBO	RBOS1	RBOS2	F value	SE value	CD value
Colour	8.000± 0.764^{a}	7.667± 0.617^{a}	7.200± 0.676^{b}	7.867± 0.743^{a}	5.84**	0.144	0.413
Texture	7.933± 0.858^{a}	7.600± 0.737^{ab}	7.267± 0.594^{b}	7.133± 0.742^{b}	4.77**	0.164	0.468
Sponginess	7.533± 0.516	7.600± 0.828	7.400± 0.632	7.667± 6.816	0.40^{Ns}	0.181	0.516
Taste	7.333± 0.488^{ab}	7.733± 0.884^{a}	7.067± 0.799^{b}	7.800± 0.862^{a}	3.89*	0.175	0.500
Flavor	7.533± 0.516^{a}	7.400± 0.910^{a}	6.733± 0.594^{b}	7.533± 0.743^{a}	4.13*	0.188	0.538
Overall Acceptability	7.800± 0.414^{a}	7.733± 0.458^{a}	7.267± 0.594^{b}	7.867± 0.640^{a}	3.84*	0.138	0.396

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, NS- non significant, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

Muffins prepared with HF were awarded higher scores for color and texture in comparison to RBO, RBOS1 and RBOS2 muffins. It was found that there was a significant ($p<0.01$) difference between color and texture made with HF, RBO, RBOS1 and RBOS2 fats. Maximum scores for sponginess (7.667±6.816) and taste were awarded to RBOS2 (7.800±0.862) muffin, whereas minimum score was awarded to muffins prepared with RBOS1 on a 9 point hedonic scale. It was also observed that

there was no significant difference in sponginess of muffins made with HF, RBO, RBOS1 and RBOS2. Overall acceptability of RBOS2 muffins (7.867±0.64) was scored best on 9 point hedonic scale in comparison to other muffins.

4.2.5 Doughnuts

The results of sensory evaluation of doughnuts developed with HF, RBO, RBOS1 and RBOS2 are given in Table 4.10. As per the results obtained, it was observed that, there was no significant difference between colour and texture of doughnuts made with HF, RBO, RBOS1 and RBOS2. The colour score of doughnuts made with HF (7.467±1.125) and RBOS2 doughnut (7.467±0.834) were similar and higher as compared to other fats used in the study. It was also observed that there was no significant difference in taste of doughnut made with RBOS1 (7.067±0.961), HF (6.733±1.163) and RBOS2 (7.200±0.775); HF (6.733±1.163) and RBO (6.400±0.910). However, there was a significant ($p<0.05$) difference between RBO and RBOS1; RBO and RBOS2. The flavor of doughnut made with RBOS2 (7.607±0.961) was rated higher than other fats used in this study, while RBO (6.067±1.710) doughnut was rated least. The overall acceptability of doughnuts made with RBOS2 (7.533±0.743) was scored high and was considered as best among all the fats. The results show that the taste, flavor and overall acceptability of RBOS2 doughnuts was significantly higher ($p<0.05$) as compared to other doughnuts indicating that, RBOS 2 can be used as a potential fat source for preparation of doughnuts.

Table 4.10. Sensory evaluation of doughnut

Sensory attributes	HF	RBO	RBOS1	RBOS2	F value	SE value	CD value
Colour	7.467± 1.125	7.133± 0.915	7.000± 0.756	7.467± 0.834	1.37^{NS}	0.203	0.579
Texture	7.600± 0.828	7.067± 1.100	7.400± 0.828	7.467± 0.915	1.20^{NS}	0.206	0.590
Taste	6.733± 1.163^{ab}	6.400± 0.910^{b}	7.067± 0.961^{a}	7.200± 0.775^{a}	2.42^{*}	0.230	0.657
Flavor	6.667± 1.397^{ab}	6.067± 1.710^{b}	6.933± 1.000^{a}	7.607± 0.799^{a}	2.53^{*}	0.278	0.795
Overall Acceptability	7.200± 0.941^{a}	6.800± 0.862^{b}	7.000± 0.926^{ab}	7.533± 0.743^{a}	2.36^{*}	0.203	0.580

Note: Values are expressed as Mean ± SD, *significant at 5% level, **significant at 1% level, NS- non significant, HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

Baked foods serve as one of the main staple food sources for consumers and lipids play an important role in the majority of baked products (Chung and Pomeranz, 1983). When choosing a lipid source for baking, the nutritional characteristics of lipid and the ability of the lipid to impart a desired physical quality to the finished product need to be considered. One of the key dietary recommendations is to consume less than 10 percent of calories from saturated fatty acids by replacing them with monounsaturated and polyunsaturated fatty acids, and to reduce TFA intake as much as possible. Therefore, it is important to consider the nutritional characteristics of the lipid sources included in food production. Usually, animal fats, such as lard, are more saturated than plant oils such as soybean oil, and their polyunsaturated fatty acid (PUFA) vs. SFA ratio is much lower (Chung and Pomeranz, 1983). In addition to the nutritional profiles, lipids should be selected according to their specific performance in the finished product (Wainwright, 1999). In general, lipids in baking contribute to products' quality characteristics such as tenderness, moist mouthfeel, lubricity, flavor, structure and shelf life. During processing, lipids affect air incorporation, lubrication and heat exchange in the baked products (Baldwin *et al.*, 1971; Stauffer, 1998; Wainwright, 1999; Ghotra *et al.*, 2002; Rogers, 2004). As per the results of sensory evaluation of cookies, cakes, piecrusts, muffins and doughnuts, it was observed that all the baked products made with RBOS2 were given significantly higher scores compared to products made with other fat sources like hydrogenated fat, Rice bran oil and RBOS1. The results indicate that RBOS2 can be used as a potential fat for preparation and production of baked products like cakes, cookies, muffins, pie crusts and doughnuts with desired quality characteristics such as tenderness, moistness, sponginess, flavor, texture and overall acceptability. Apart from physical quality characteristics, RBOS2 is plant source oil derived from a mix of Rice bran oil and hydrocolloids, which is healthier compared to hydrogenated fats or other animal fats.

4.3 Consumer acceptability

As per the results of sensory evaluation (Table 4.6), the overall acceptability of cake was found to be significantly higher compared to other products like cookies, muffins, doughnuts and piecrust. Hence cake was selected for conducting the consumer acceptability study. Two cakes prepared from hydrogenated fat and RBOS2 were used for carrying out the consumer evaluation study. As hydrogenated fat is generally used to prepare most of the baked products, it was chosen as a control in the consumer evaluation study against the RBOS2 cake.

A total of 100 subjects, including 23 males and 77 females were randomly selected for the consumer acceptability study from the age group of 18-60 years based on their interest in participating in the study. The study was done by using a consumer acceptability questionnaire consisting of two sections describing the demographic profile and the hedonic scoring for the cake. The demographic profile and information on RBOS2 cake from the respondents is illustrated in Table 4.11, and the hedonic scoring by respondents for consumer acceptability is illustrated in Table 4.12.

It was found that 80% of the subjects were in the age group of 18-29 years, 9% of the subjects were in age group of 30-39 years, 8% of the subjects were in age group of 40- 49 years and 3% of the subjects were in age group of 50-60 years (Fig. 4.1a).Among the 100 respondents, 23% were male and 77% were female respondents (Fig. 4.1b). Among the respondent's education status, it was observed that 51% of subjects have completed their higher education, 7% of subjects completed their under-graduation, 32% of subjects completed their post-graduation, 7% of the subjects completed their doctorate and only 3% were illiterate (Fig 4.1e).

The results of consumer evaluation regarding frequency of consumption of rice bran oil, it was found that 40% of the respondents consumed rice bran oil daily (Fig 4.1c). The average amount of rice bran oil consumption per month was found to be 1-2 ltrs. There were 15% respondents who consumed rice bran oil rarely and 45% of respondents never consumed rice bran oil.

Based on the responses on awareness of rice bran oil spread, it was found that 2% of the respondents were aware of rice bran oil spread, whereas 98% were unaware of rice bran oil spread (Fig 4.1 D).

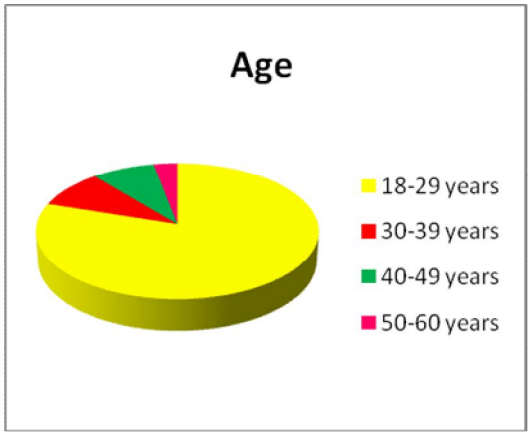
Among the respondents, all 100% of respondents had never tasted rice bran oil spread at all. Based on the responses about awareness of health benefits rice bran oil spread, it was found that 40% of the respondents were aware about the beneficial effects of consumption of rice bran oil spread, whereas 60% of respondents were unaware about health benefits of rice bran oil spread (Fig 4.1G).

According to the willingness to buy rice bran oil spread products, it was found that 94% of the respondents were willing to buy rice bran oil spread products, whereas 6% were not willing to buy (Fig 4.1H). The reason for 94% of respondent's willingness to buy the rice bran oil spread was due to the health benefits of rice bran oil spread. There are studies which suggest that acceptability for products is related to consumer

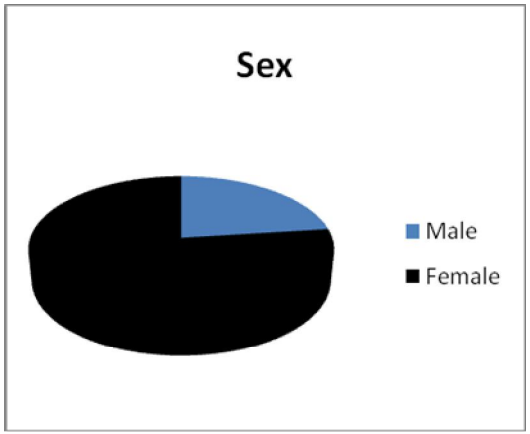
attitude and knowledge about the health claims of the product (Drake and Gerard, 2003; Ramcharitar *et al.*, 2005).

Table 4.11 Demographic profile of respondents participated in consumer evaluation

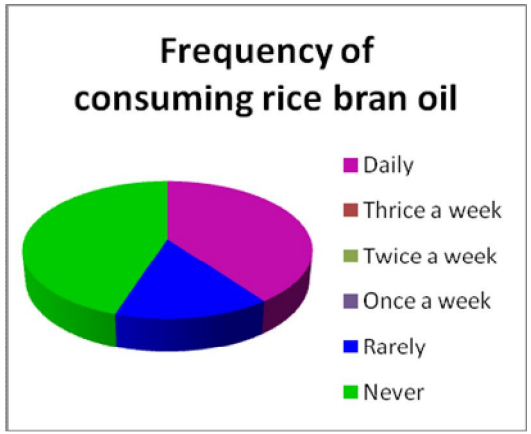
1.Age (n=100)	
i. 18-29 years	80%
ii. 30-39 years	9%
iii. 40-49 years	8%
iv. 50-60 years	3%
2.Sex (n=100)	
i. Male	23%
ii. Female	77%
3. Education (n=100)	
i. Primary	0
ii. Secondary	0
iii. Higher Secondary	51 %
iv. UG	7%
v. PG	32%
vi. Doctorate	7%
vii. Other	3%
4.Frequency of consuming rice bran oil (n=100)	
i. Daily	40%
ii. Thrice a week	0
iii. Twice a week	0
iv. Once a week	0
v. Rarely	15%
vi. Never	45%
5.Amount of rice bran oil consumption per month (No. of litres) (n=100)	
i. 1-2 ltrs	100%
ii. 3-5 ltrs	0
iii. 5-10 ltrs	0
6. Awareness of rice bran oil spread (RBOS) (n=100)	
i. Yes	2%
ii. No	98%
7. Tasted product developed with RBOS any time before	
i. Yes	0
ii. No	100%
8. Awareness of health benefits of RBOS (n=100)	
i. Yes	40%
ii. No	60%
9. Willingness to buy (n=100)	
i. Yes	94%
ii. No	6%
10. Price willing to pay (n=100)	
i. Rs.80 or less	72.41%
ii. Rs.100	27.59%
iii. Rs.150	0



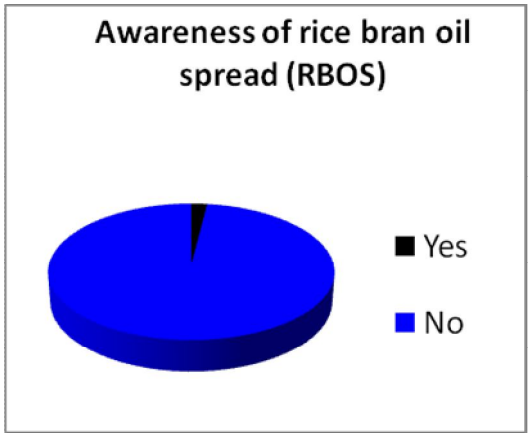
(a) Age

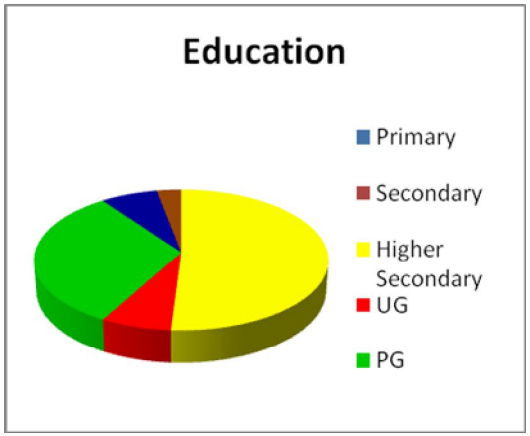


(b) Sex



(c) Frequency of consuming rice bran oil



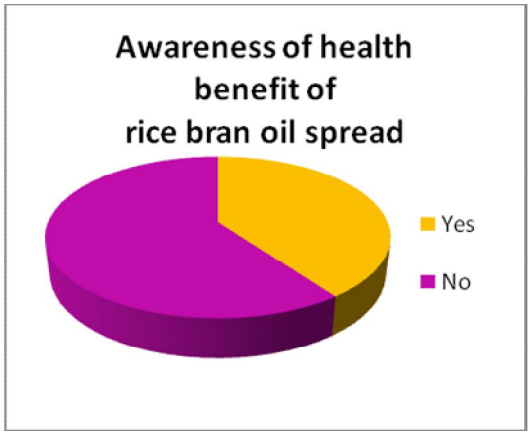


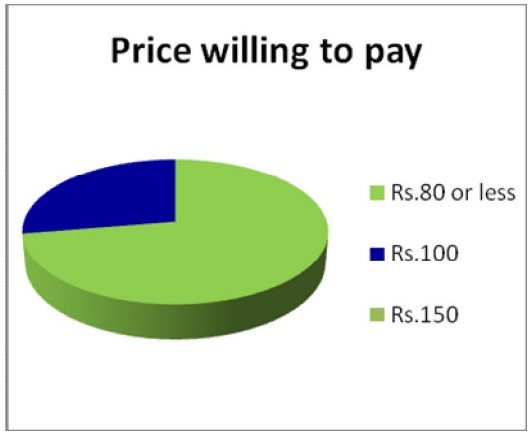
(d) Awareness of rice bran oil spread (RBOS)

(e) Education

(f) Responses of whether rice bran oil spread products were tasted before

Figure 4.1 Demographic profile of respondents and the responses about RBOS2 Cake from the consumer evaluation study participants





(g)Awareness of health benefit of rice bran oil spread

(h) Willingness to buy

(i) Price willing to pay

Table 4.12 Hedonic scoring by respondents for consumer acceptability

Samples	HF Cake (n=100)	RBOS2 Cake (n=100)	F Value	SE Value
Colour	4.370±0.52	4.480±0.59	1.77NS	0.058
Texture	4.340±1.06	4.170±0.60	1.81NS	0.089
Sponginess	4.580±0.51	4.490±0.84	4.19NS	0.067
Taste	4.230±0.75	4.100±0.81	1.53NS	0.074
Flavor	4.140±0.98	4.070±0.76	0.27NS	0.094
Overall Acceptability	4.420±0.60	4.280±0.52	9.04NS	0.062

Note: Values are expressed in Mean ± SD. NS indicates Non Significant difference.
HF –Hydrogenated Fat; RBOS2 – Rice Bran Oil Spread 2

Drake and Gerard (2003) studied consumer attitude and acceptability of soy fortified yoghurts among 547 consumers with the objective to determine consumer knowledge of soy and dairy foods and consumer acceptance of yoghurts fortified with 2.5% soy protein concentrate (SPC). Consumers filled out a questionnaire requesting demographics information as well as knowledge and attitudes on dairy and soy foods. The results showed that the acceptability scores for yoghurts significantly ($p<0.05$) increased with frequency of dairy yoghurt consumption and knowledge of the health claim significantly ($P<0.05$) increased the acceptability scores for yogurts.

Ramcharitar *et al.* (2005) evaluated consumer acceptability of muffins with flaxseed in a focus group of 18-25 years by using hedonic scoring and food action rating scale (FACT). The results showed that the majority of consumers (82.6%, $p<0.01$) rated the control muffin (0% flaxseed) higher than the flax muffin for appearance, color, flavor, texture, overall acceptability and food acceptance. It was concluded by the observation that, many persons aged 18 to 25 years did not think they needed to consume “health foods”. This “indifference” and in some cases “negative” attitude towards the flax muffins, which they considered a “health food”, may have influenced the ratings the flax muffins received.

In terms of price willing to be paid by the consumers for the rice bran oil spread, it was found that 72.41% of respondents were willing to pay Rs.80 or less per litre of rice bran oil spread and 27.59% of respondents were willing to pay Rs.100 per litre (Fig

4.1i). As per the market information on the price for premium quality of rice bran oil, it ranges between Rs.60-80 or above per litre of oil.

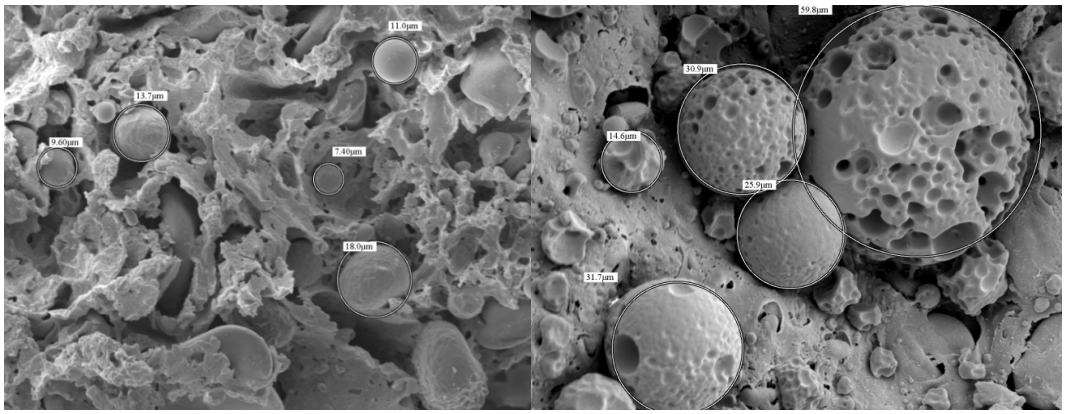
The consumer evaluation study questionnaire was of two sections – one was regarding the demographic profile and the second part was regarding hedonic scoring of the control and experimental cake. After completion of the demographic profile questionnaire, all the respondents were given two cakes (HF cake and RBOS2 cake) to be tasted and were asked to fill in the 5 point hedonic score card. After tasting the cakes, the respondents filled in the score card for attributes like colour, flavour, texture, sponginess taste and overall acceptability on a 5-point hedonic scale (5 = Like very much to 1 = Dislike very much). The two cakes prepared with Hydrogenated Fat (HF) and Rice Bran Oil Spread 2 (RBOS2) were coded with 3 digit random numbers and presented simultaneously, but in random order, to each consumer on white paper plates.

Mean and Standard deviation was calculated for each attribute of the products with the scores obtained by hedonic rating, the attributes like general appearance and color are considered to directly influence consumer decision to purchase a product and, consequently, to consume it (Ferreira, 2000). The results of hedonic scores of consumer acceptability study are summarized in Table 4.12.

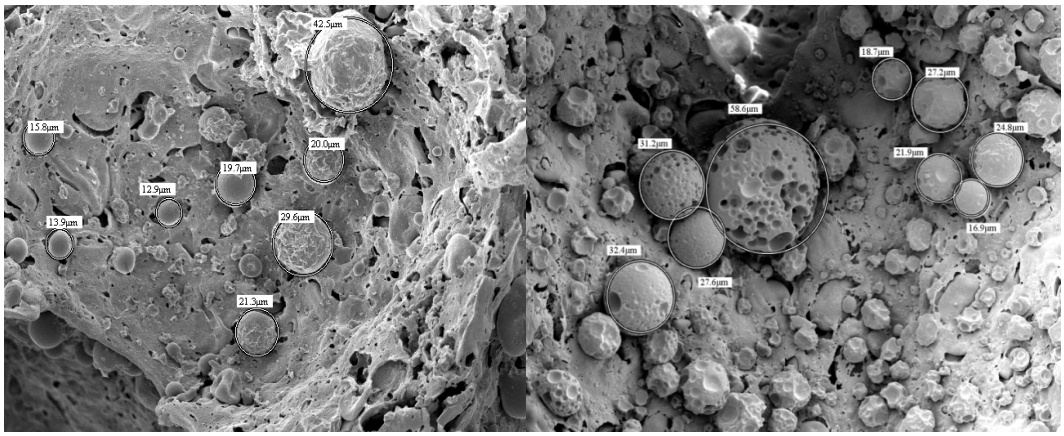
From the results of the study (Fig 4.2), it was found that the mean score for colour of RBOS2 cake was 4.480 ±0.59 on a 5-point hedonic scale, which was higher than the colour score of HF cake (4.370±0.52) indicating that cake prepared with rice bran oil spread 2 was better liked in terms of colour than the cake prepared with hydrogenated fat. The hedonic score for taste of HF cake was 4.230±0.75, which was higher than the score of RBOS2 cake (4.100±0.81). The hedonic score for overall acceptability of HF cake was 4.420±0.60, which was higher than RBOS2 cake (4.280±0.52), whereas the mean score for flavor parameter of HF cake was found to be higher (4.140±0.98), as compared to the RBOS2 cake (4.070±0.76). However, it was observed that there was no significant difference in the colour, flavour, texture, taste and overall acceptability parameters between the HF cake and RBOS2 cake, which suggests that there was no identifiable difference between the products prepared with Hydrogenated fat and Rice bran oil spread 2.

4.4 Scanning Electron Microscope (SEM) imaging of Cake

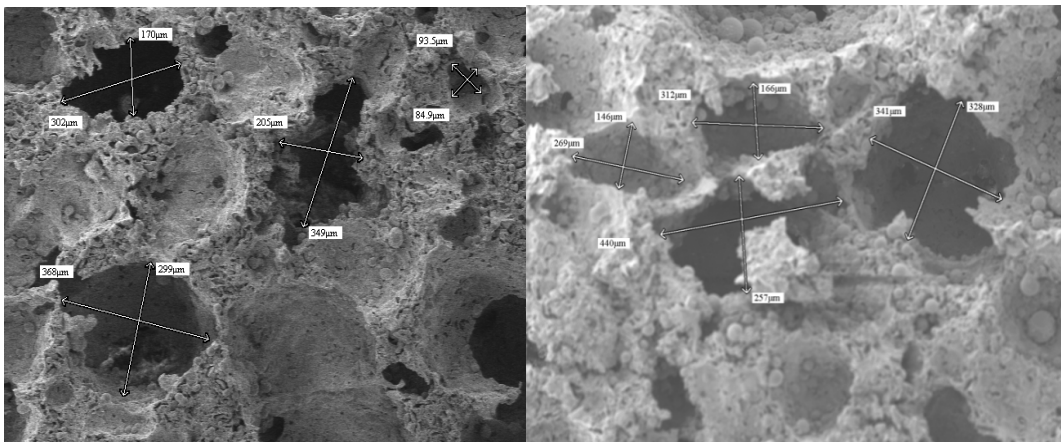
Figure 4.3 presents the micro structure of control cake prepared with hydrogenated fat and cake prepared with RBOS2 which was most acceptable in the



A.Control (HF) cake at 1000x magnification B. RBOS2 cake at 1000x magnification



C. Control (HF) cake at 500x magnification D. RBOS2 cake at 500x magnification



E. Control (HF) cake at 100x magnification F. RBOS2 cake at 100x magnification

Figure 4.3 SEM images of control (HF) cake and RBOS2 cake. **A)** SEM image of control (HF) cake measured at 1000X. **B)** SEM image of RBOS2 cake measured at 1000X. **C)** SEM image of control (H) cake measured at 500X. **D)** SEM image of RBOS2cake measured at 500X. **E)** SEM image of control (HF) cake measured at 100X. **F)** SEM image of RBOS2 cake measured at 100X.

sensory evaluation, under Scanning Electron Microscopy with 1000x, 500x and 100x magnifications.

The pores within the core varied in diameter between 7.40µm -18.0µm and 14.6µm - 59.8µm at 1000x magnification in control and RBOS2 cakes respectively. The internal structure and pore structure of RBOS2 cake was finer and smoother than the control cake. The pores within the core varied in diameter between 13.9µm - 29.6µm in control cake and between 16.9µm - 58.6µm in RBOS2 cake at 500x magnification. Larger amounts of interior heating in baking results in increased moisture vapor generation inside the food which creates high pressure gradient. Higher pressure gradient occurring inside the cakes during microwave heating as compared to conventional heating can cause looser and more porous structures in cakes (Elif *et al.*, 2010). Ozkoc *et al.* (2009) obtained similar results for breads baked in different ovens in their study.

It was stated by Mc Clements (2007) that, pore diameter range of 100µm–100mm represents macro-scale pore structure, which is related to appearance of the samples. In our study, the pore diameter range at 100x magnification was between 84.9µm - 349µm in the control cake, where as it was between 146µm - 440µm in RBOS2 cake indicating presence of macro pore size in both the cakes. However, the macro pore size of the RBOS2 cake was much better than the control cake as indicated by the results of sensory evaluation as well as SEM imaging, indicating that RBOS2 as an ingredient instead of hydrogenated fat, can contribute to better texture and loaf volume in the cake. The micro structure of RBOS2 presents distinct distinguishable open pore structures as a continuous matrix, which is built up by starch, protein and fat. Similar microstructure observation in the gluten-free rice and low-protein starch breads has been reported by Hung *et al.* (2007).

4.5 Fatty acids Composition of cake

4.5.1Total fat content of cake

As per the results of sensory evaluation, cake was rated better among the other products like cookies, muffins, pie crusts and doughnuts. Hence cake was selected for the fatty acid and trans fatty profiling. The total fat (g/100 g food) content of the cake made with HF, RBO, RBOS1 and RBOS2 are given in Table 4.13. It is evident from the results of analysis that the fat contents of cake with HF, RBO, RBOS1 and RBOS2 varied. Total fat content in the cake samples analyzed was in the order: RBOS1 (17.15

g/100g) > HF (17.09 g/100g) > RBO (15.08 g/100g) > RBOS2 (14.95 g/100g). The type of fatty acids present in any food product will attain significance with respect to health implications, only if the total fat content is higher like in RBOS1 which is almost 17.15% fat in cake, as compared to RBO and RBOS2 cake. The higher the fat content, intakes will be more, and simultaneously, the presence of unhealthy fatty acids like saturated fatty acids and trans-fatty acids will attain significance with respect to pre-disposition to heart diseases. Foods that are high in trans or saturated fatty acids are associated with an increased risk of cardiovascular disease and diabetes (Mozaffarian *et al.*, 2006). TFA are also associated with markers of systemic inflammation, which may be involved in the pathogenesis of coronary artery disease (Mozaffarian *et al*., 2005). A daily intake of 5g of primarily industrial TFA is associated with a 29% increased risk of coronary heart disease (Stender *et al.*, 2008). The chromatograms of HF cake, RBO cake, RBOS1 and RBOS2 cakes are present in figure 4.4a,b,c and d.

Table 4.13 Total Fat and Trans Fatty Acid Content of Cake

Sl. No.	Sample Name	Total Fat (g/ 100g)	Elaidic Acid	Linolelidic acid	Vaccenic Acid	Total TFA (%)
1	HF cake	17.09	6.64	8.48	0.34	15.46
2	RBO cake	15.08	2.62	0.94	-	3.56
3	RBOS 1 cake	17.15	3.59	0.95	-	4.54
4	RBOS 2 cake	14.95	2.87	0.91	-	3.78

HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

Trans Fatty Acid content of cake: In cake samples, the amount of total TFA was 15.46% in HF cake, 3.56% in RBO cake, 4.54% in RBOS1 cake, 3.78% in RBOS2 cake (Table 4.13). Total *trans* fat content was higher in HF cake (15.46%) as compared to experimental samples RBO, RBOS1 and RBOS2. The TFA content assumes significance in terms of their ill effects on the health of consumers, only if fat content is also high. Hence, consumption of the HF products might prove to be harmful, if consumed in large amounts and at higher frequencies.

The major trans fatty acid observed in all samples was elaidic acid (C18:1 trans-9) and Linolelidic acid (C18:2, trans-6). Elaidic acid was highest in HF cake (6.64%) and least in RBO cake (2.62%). Linolelidic acid was highest in HF cake (8.48%) and least in RBOS2 cake (0.91%). Vaccenic Acid was detected only in HF cake (0.34%). Presence of vaccenic acid in HF cake could be due to presence of ingredients such as

milk, which is of animal origin. Vaccenic acid is a TFA which is known to be produced in the rumen of epigastric animals naturally during the partial biohydrogenation of linoleic acid (Adlof *et al.* 2000; Lock and Bauman, 2004), and it acts as a precursor for the endogenous synthesis of cis9, trans11-conjugated linoleic acid (CLA) via the action of the n9 desaturase enzyme in both humans and animals (Adlof *et al.* 2000). As per the results obtained, it is understood that hydrogenated fat if used in baking cakes, leads to formation of hazardous trans fats whereas, RBO or RBOS1 or 2 used in place of hydrogenated fats could lead to significant reduction in formation of trans fats. Hence, RBOS can be promoted as healthy fat for production of baked products.

4.5.2 Saturated fatty acid content of cake

Table 4.14 shows saturated fatty acid content of cake. The SFA content of the cake ranged from 24.08% in RBO cake to 44.55% in HF cake. However, as discussed in the previous section, the SFA content of foods assumes significance if the total fat content is also high. In terms of high fat as well as high SFA content, the food products might be more harmful to health if consumed in higher amounts and more frequently, the predominant fatty acids present in these items which had high fat content as well as high SFA was palmitic acid, which ranged from 22.07% in RBO cake to 41.64% in HF cake. Other SFA like butyric acid (C4:0), caprylic acid (C6:0), caproyic acid (C10:0) and behenic acid (C22:0) were absent in all cake samples. Lauric acid (C12:0), myristic acid (C14:0), stearic acid (C18:0) and arachidic acid (C20:0) were present in lower concentration.

Hegsted *et al*. (1965) showed that myristic acid (14:0) was four times more potent than Palmitic acid (16:0) in raising total plasma cholesterol (TC), whereas Stearic acid (18:0) had no effect. Since palmitic acid is not as harmful as other short chain fatty acids, higher SFA content, which actually reflects higher palmitic acid content, might not be as harmful as consumption of foods with higher amounts of short chain fatty acids, unless they are also rich in TFA.

4.5.3 Unsaturated fatty acids content of cake

Table 4.15 includes the monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA) of cake made with HF, RBO, RBOS1 and RBOS2. The total amount of unsaturated fat in cake sample ranged from 39.99% in HF cake to 72.36% in RBO cake followed by RBOS2 cake (72.35%) and RBOS1 cake (70.64%). Oleic acid (C18:1) was

Table 4.14 Saturated Fatty Acid Content of Cake

Sl. No	Sample Name	Butyric Acid (C4:0)	Caproic Acid (C6: 0)	Caprylic Acid (C8: 0)	Capric Acid C(10: 0)	Lauric Acid (C12:0)	Myristic Acid (C14:0)	Palmitic Acid (C16:0)	Stearic Acid (C18:0)	Arachidic Acid C(20:0)	Tricosanoic Acid (C23:2)	Behenic Acid (C22:0)	Total SFA
1.	HF cake	-	-	-	-	0.71	1.19	41.64	1.01	-	-	-	44.55
2.	RBO cake	-	-	-	-	-	0.39	22.07	0.89	0.73	-	-	24.08
3.	RBOS1 cake	-	-	-	-	-	0.43	22.89	0.72	0.78	-	-	24.82
4.	RBOS2 cake	-	-	-	-	-	0.43	22.57	0.44	0.72	-	-	24.16
5.	RBO cake	-	-	-	-	-	0.42	23.61	1.02	0.62	-	-	25.67

HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

Table 4.15 Unsaturated Fatty Acid Content of Cake

Sl. No.	Sample Name	Myristoleic Acid (C14:1)	Oleic Acid (C18:1)	Linoleic Acid (C18:2) (PUFA)	Palmitoleic Acid (C16:0)	α-Linolenic Acid (C18:3)	Cis 11, 14-Eicosadinoic Acid (C20:2)	Cis 13, 16-Docosadienoic Acid (C22:2)	Total UFA
1.	HF cake	-	39.99	-	-	-	-	-	39.99
2.	RBO cake	-	42.60	29.29	-	0.47	-	-	72.36
3.	RBOS1cake	-	41.01	29.18	-	0.45	-	-	70.64
4.	RBOS2 cake	-	41.66	29.97	-	0.72	-	-	72.35

HF –Hydrogenated Fat; RBO – Rice Bran Oil; RBOS1 – Rice Bran Oil Spread 1; RBOS2 – Rice Bran Oil Spread 2.

present in range of 39.99 (HF cake) to 42.60% (RBO cake). Linoleic acid (C18:2) was not present in HF cake but was present in RBO (29.29%), RBOS1 (29.18%) and RBOS2 (29.97%) cakes. α linolenic acid (C18:3) was not present in HF cake, but was present in RBO(0.47%), RBOS1(0.45%) and RBOS2 (0.72%) cakes. Myristoleic acid (C14:1), palmitoleic acid (C16:0), eicosadinoic acid (C20:2) and docosadienoic acid (C22:2) were absent in all the cake samples. However, as described earlier, the types of fatty acids present in this cake sample attains significance only if the total fat content in the products is present in higher amounts, like in HF cake. Binkoski *et al*. (2005) reported that, PUFAs are an important component of a cholesterol-lowering diet. Berry *et al.* (1991) also indicated that, in humans, dietary regimens rich in monounsaturated fatty acids enhance the oxidative stability of LDL. Fats in the order of being less harmful to health in cake were RBO < RBOS2 < RBOS1 < HF.

4.5.4 Trans fatty acids content of cake

Hydrogenated fat which is commonly used for production of various baked products could be harmful due to presence of higher amount of saturated fats as well as trans fats. Recently, consumers have become more concerned about the health implications of TFA consumption. Ascherio and Willet (1997) suggested that TFA intake can be reduced by lowering total fat intake. However, there is concern that this will lead to an insufficient intake of essential fatty acids (n-3 and n-6) (Tarrago-Trani *et al.* 2006). Lipids such as RBOS with reduced TFA and saturated fatty acids can be used as TFA replacements. Apart from reduced formation of trans fats, RBOS2 also proved to be having good functional properties that are important to ensure baked food quality. Some of the physicochemical properties including texture, taste, flakiness and sponginess were found to be good when RBOS2 was incorporated as fat instead of other fat sources. Therefore, new technologies for production of RBOS2 in bulk can be promoted that will improve the nutritional profile of lipids while still maintaining the functional properties required by the food producer for production of baked products.

Chapter V

SUMMARY AND CONCLUSIONS

The “Effect of Rice bran Oil Spread (RBOS) on the physical and sensory quality of b aked products” was conducted in the Department of Foods and Nutrition, Post Graduate and Research Centre, Professor Jayashankar Telangana State Agricultural University and Indian Institute of Rice Research, Rajendranagar, Hyderabad during the year 2014 – 16.

Baked foods serve as one of the main staple food sources for consumers and lipids play an important role in the majority of baked products. When choosing a lipid source for baking, the nutritional characteristics of lipid and the ability of the lipid to impart a desired physical quality to the finished product need to be considered. Generally hydrogenated fats are used for preparation of baked products as they contribute to desired characteristics such as tenderness, moistness, mouth feel, lubricity, flavor, structure and shelf life in the baked products. Recently consumers have become more concerned about the health implications of trans fats which are produced in the baked products due to usage of hydrogenated fat at high baking temperatures. Hence alternate sources of hydrogenated fats in baking are catching up the attention of researchers.

Rice bran oil is generally considered to be one of the highest quality vegetable oil in terms of its cooking quality, shelf life and fatty acid composition. Rice bran oil is obtained from the outer bran layer of rice. Studies have shown that Rice bran oil semisolid fraction (RBOF) can also be incorporated into baked food formulations with improvement in oxidative stability in baked foods. Therefore, a study was planned to develop and standardize baked products with Rice bran oil and 2 varieties of Rice bran oil spread to evaluate the physico chemical and sensory properties of the developed products, as well as to determine the consumer acceptability of the best acceptable product. The best accepted product was subjected to scanning electron microscopic study and fatty acid estimation with special reference to trans fatty acids..

Five different baked products namely cakes, cookies, piecrusts, muffins and doughnuts were developed and standardized by using Hydrogenated fat (HF) , Rice bran oil (RBO) and two varieties of Rice bran oil spread (RBOS 1 and RBOS2) in the Foods

Lab of the Department of Foods & Nutrition, Post Graduate & Research Centre, PJTSAU, Hyderabad. The five standardized products with HF (control) & RBO, RBOS1 and RBOS2 (experimental) were subjected to physico chemical analysis and sensory evaluation by 15 semi-trained panel members using a 9-point hedonic scale.

The results of physical analysis (moisture and density) of cake had significant differences ($P<0.01$) between the type of fat used in preparation of cake. Cake moisture was highest in HF cake (16.035±0.36) and least in RBOS1 cake (8.924±0.43). The cake density was statistically similar in HF cake and RBOS1 cake; RBO cake and RBOS2 cake.

The physical parameters in cookie (moisture, cookie height, cookie dough density and cookie spread) were significantly different in HF cookies, RBO, RBOS1 and RBOS2 cookies. The cookie moisture (1.280±0.01) and cookie spread (0.350±0.00) were highest in RBO cookies, whereas cookie dough density (1.233±0.02) was highest in RBOS1 cookie.

The physical parameters in piecrust viz., moisture, height point, oiliness were significantly different at ($P<0.05$), where as crust height and percentage shrinkage were significantly different at ($P<0.01$) among the various types of fats used. The pie crust moisture content, height point and crust height were highest in HF pie crust compared to other pie crusts. The oiliness was least in RBOS2 pie crust (0.063±0.02) indicating the healthiness of piecrust made with RBOS2. Lesser the oiliness in a product, lesser will be the risk of high fat intake related health disorders.

The physical parameters in muffins (moisture content and bulk density) were significantly different ($P<0.01$) in HF, RBO, RBOS1 and RBOS 2 muffins. The moisture content was highest in RBOS1 muffin (17.546±0.18) and least in RBOS 2 muffin (13.874), where as the bulk density was highest in RBOS2 muffin (0.423±0.01) and least in RBOS1 muffin (0.257±0.01) indicating an inverse association between the two parameters.

The physical parameters in doughnut (moisture content, volume and specific volume) were significantly different ($P<0.05$) in the doughnuts prepared HF, RBO, RBOS1 and RBOS2. The doughnut moisture was least in RBOS2 doughnut (11.197±0.10), whereas the volume was highest (44.333±0.41) in the same. The specific

volume of doughnut was highest in RBOS1 (8.400±0.10) and least in RBO (7.427±0.12) doughnut.

The physical parameters results showed lower moisture content in all the baked products (cookie, cake, pie crust, muffin and doughnut) made with RBOS2. Hence, it can be assumed that due to lower moisture content in all the products made with RBOS2, the shelf life of the product made with RBOS2 could be better as compared with all other sources of fat. Similarly density of the products was high in all the products made with RBOS2. Oiliness was less in RBOS2 products. The results indicate that RBOS2 can be used as a very good source of fat which can impart desired characteristics in the baked products.

Sensory evaluation of control and experimental samples at lab level showed that there is a significance difference among the five products made with HF, RBO, RBOS1 and RBOS2 in the sensory attributes like colour, texture, taste and overall acceptability. According to the results of overall acceptability, it was observed that the cake made with RBOS2 was highly acceptable compared to other products and hence the same product was used to carry out consumer acceptability study, scanning electron microscopic study and fatty acids estimation with special reference to trans fatty acids.

A study on consumer acceptance of RBOS2 cake was conducted on a total of 100 subjects (n=100), including 23 males and 77 females who were randomly selected from the age group of 18-60 years based on their interest in participating in the study. The study was conducted at the Department of Foods & Nutrition, Post Graduate & Research Centre, PJTSAU, Hyderabad and Indian Institute of Rice Research (Formerly Directorate of Rice Research), Rajendranagar, Hyderabad by using a consumer acceptability questionnaire consisting of two sections describing the demographic profile and the hedonic scoring (5-point hedonic scale) of attributes like appearance, color, texture, flavour, taste and overall acceptability of the product. HF cake (control) and cake prepared with RBOS2, were served to the consumers. The results indicated that no significant difference was found between the hedonic ratings of the sensory attributes of HF cake and RBOS2 cake. The results of consumer acceptance study showed that there was no significant difference in the sensory attributes of the product prepared with HF and RBOS2. The RBOS2 cake was well accepted by the respondents.

The scanning electron microscope results indicated that the pore size of HF cake ranged from 84.9µm-349µm while it ranged 146µm-440µm in RBOS2 cake at 100x

magnification indicating the presence of macro size pores in both the cakes. However, the macro size pores of the RBOS2 cake was much better than the control cake as indicated by the results of sensory evaluation as well as SEM imaging, indicating that RBOS2 as an ingredient instead of hydrogenated fat can contribute to better texture and loaf volume in cake.

Total fat content in the cake samples analyzed was in the order: RBOS1 (17.15 g/100g) > HF (17.09 g/100g) > RBO (15.08 g/100g) > RBOS2 (14.95 g/100g). The SFA content of the cakes ranged from 24.08% in RBO cake to 44.55% in HF cake. . The total amount of unsaturated fat in cake sample ranged from 39.99% in HF cake to 72.36% in RBO cake followed by RBOS 2 cake (72.35%) and RBOS 1 cake (70.64%). The amount of total TFA was 15.46% in HF cake, 3.56% in RBO cake, 4.54% in RBOS1 cake and 3.78% in RBOS2 cake. Total *trans* fat content was higher in HF cake (15.46%) as compared to experimental samples RBO, RBOS1 and RBOS2. The TFA content assumes significance in terms of their ill effects on the health of consumers, only if fat content is also high. Hence, consumption of the HF products might prove to be harmful if consumed in large amounts and at higher frequencies. The major *trans* fatty acid observed in all samples was elaidic acid (C18:1 trans-9) and Linolelidic acid (C18:2, trans-6). Elaidic acid was highest in HF cake (6.64%) and least in RBO cake (2.62%). Linolelidic acid was highest in HF cake (8.48%) and least in RBOS2 cake (0.91%). Vaccenic Acid was detected only in HF cake (0.34%). Hydrogenated fat which is commonly used for production of various baked products could be harmful due to presence of higher amount of saturated fats as well as trans fats.

The results indicate that RBOS2 can be used as a potential fat for preparation and production of baked products like cakes, cookies, muffins, pie crusts and doughnuts with desired quality characteristics such as tenderness, moistness, sponginess, flavor, texture and overall acceptability. Apart from physical quality characteristics, RBOS2 is a plant source oil derived from a mix of Rice bran oil and hydrocolloids, which is healthier compared to hydrogenated fats or other fats as seen from the results of the study. The trans fats were very less in RBOS2 product as compared to other sources of fats used in the study. Hence, RBOS2 can be promoted as healthy fat for production of baked products

RECOMMENDATIONS FOR FURTHER STUDIES

The study demonstrates potential application of Rice Bran Oil Spread for the development of healthier baked products by replacing more traditional forms of fats especially hydrogenated fats. There are few limitations of the study which can be overcome by doing further research in the following areas:

- In addition to expanding the scope of sensory testing, instrumental tests can be added as well and results from both can be correlated for better understanding of sensory and organoleptic characteristics of the products.
- In terms of different preparation technique, more research can be done with different types of baking (microwave baking, steam baking, oven baking etc) and their affect on trans fatty acids production.
- The consumer acceptability test can be conducted in target audiences who are most familiar with baked products and those who regularly consume baked products.

LITERATURE CITED

AACC, Approved Methods of the American Association Cereal Chemist, AACC, 1983, St. Pauls, Minnesota, USA.

Adlof, R.O., Duval, S and Emken, E.A. 2000. Biosynthesis of conjugated linoleic acid in humans. *Lipids*. 35:131–135.

Amarasinghe, B.M.W.P.K and Gangodavilage, N.C. 2004. Rice bran oil extraction in Sri Lanka: Data for process equipment design. *Food Soup Journal*. 82: 54-59.

Amarjeet, K., Vishaldeep, J., Thind, S.S and Poonam, A. 2012. Rice bran oil an alternate bakery shortening. *Journal of Food Science and Technology*. 49(1): 110–114.

Anonymous. 2002. *National workshop on rice research and extension. Feeding the extra millions by 2025* (p. 1). Gazipur: Bangladesh Rice Research Institute.

Anonymous. 2007. Rice production and consumption by country and Geographical Region, 1961-2008. United States Department of Agriculture. Retrieved from http://www.USDA.Gov.in/1961-2007.xi.1of12.

Anonymous. Sea Handbook-2009. 9th edition. The solvent extractor's association of India: India, 885-891.

AOAC. 2001.996.06. Official methods of Analysis. Association of Official Analytical Chemists.

AOAC. 2003.06. Official Methods of Analysis. 19th edition. Association of Official Analytical Chemists.

AOAC (2005) Official Methods of analysis of the AOAC. Association of Official Analytical Chemists. 18th Edition. Arlington VA 2209, USA. AOAC 929.03, chap 32, pp02.

Ascherio, A and Willett, W.C. 1997. Health effects of Trans fatty acids. *The American Journal Clinical Nutrition*. 66: 1006S-1010S.

Azam, M.M.,Waris, A and Babu, V.R. 2015. *Rice Bran Oil: Nutritional and Health Benefits*. Technical Bulletin No.89/2015. ICAR-Indian Institute of Rice Research.

Baik, O.D and Mittal, G.S. 2005. Heat and moisture transfer and shrinkage simulation of deep-fat tofu frying. *Food Research International*. 38: 183-191.

Bakota, E.L., Mooser, W.J.K and Liu, S.X. 2014. Evaluation of a rice bran oil-derived spread as a functional ingredient. *European Journal of Lipid Science and Technology*. 116: 521-531.

Bakota, E.L., Mooser, W.J.K., Hwang, H.S and Bowman, M.J. 2013. Solvent fractionation of rice bran oil to produce a spreadable rice bran product. *European Journal of Lipid Science and Technology*. 115: 847–857.

Baldwin R.R, Baldry R.P and Johansen R.G. 1971. Fat systems for bakery products. *Journal of American Oil Chemist Society* 49: 473-477.

Berger, A., Rein, D., Schafer, A., Monnarad, I., Gremaud, G., Lambelet, P and Bertoli, C. 2005. Similar cholesterol lowering properties of rice bran oil, with varied gamma oryzanol, in midly hypercholesterolaemic men. *European journal of clinical Nutrition*. 44:163-173.

Berglund, P.T and Hertsgaard, D.M. 1986. Use of vegetable oils at reduced levels in cake, piecrust, cookies and muffins. *Journal of Food Science*.51: 640-644.

Berry, E., Eisenberg, S., Haratz, D., Friedlander, Y., Norman, Y., Kaufmann, N and Stein, Y. 1991. Effects of diets rich in monounsaturated fatty acids on plasma lipoproteins: The Jerusalem Nutrition-High MUFAs vs high PUFAs. *American Journal of Clinical Nutrition*. 3: 899-907

Binkoski, A.E., Etherton, K.P.M., Wilson, T.A., Mountain, M.L and Nicolosi, R.J. 2005. Balance of unsaturated fatty acids is important to a cholesterol-lowering diet: comparison of mid-oleic sunflower oil and olive oil on cardiovascular disease risk factors. *Journal of American Dietetic Association*. 105(7): 1080-1086.

Bouchon, P., Aguilera, J.M and Pyle D.L. 2003. Structure Oil-Absorption relationships duringdeep-fat frying. *Journal of Food Science.* 68(9): 2711-2716.

Bozzola, J.J and Russell, L.D. 1999. Electron microscopy principles and techniques for biologists, 2nd edition. *Jones and Bartlett, Boston*, pp. 19 -24, 54 - 55 and 63 – 67.

Bramley, P.M., Elmadfa, I., Kafatos, A., Kelly, F.J., Manios, Y., Roxborough, H.E., Suhuch, W., Sheehy, P.J.A and Wagner, K.H. 2000. Vitamin E. *Journal of Science Food and Agriculture*. 80: 913-938.

Brooker, B.E. 1993. The stabilization of air in cake batters-the role of fat. *Food Structure*12: 285-296.

Calder, P.C. 2003. N-3 polyunsaturated acids and inflammation: from molecular biology to the clinic. *Lipids*. 38: 343-352.

Cantral, R.P and Reeves, T.J. 2002. The cereal of the world's poor take center stage. *Science, 296*, 53.doi:10.1126/science.1070721.

Champagne, E.T., Wood, D.F., Juliano, B.O and Bechtel, D.B. 2004. The rice grain and its gross composition. In: Champagne ET(edition) Rice. AACC St. Paul, USA. 77-107.

Chen, M.H and Bergman, C.J. 2005. A rapid procedure for analyzing rice bran tocopherol, tocotrienol and g-oryzanol contents. Journal of Food Composition and Analysis. 18: 319-31.

Cheong, L.Z., Tan, C.P., Long, K and Idris, N.A. 2011. Baking performance of palm diacylglycerol bakery fats and sensory evaluation of baked products. *European Journal of Lipid Science and Technology.* 113: 253–261.

Chotimarkorn, C and Silalai, N. 2008. Addition of rice bran oil to soybean oil during frying increases the oxidative stability of the fried dough from rice flour during storage. *Food Research International* 41(3): 308-317.

Chung, O.K and Pomeranz, Y. 1983. Recent trends in usage of fats and oils as functional ingredients in the bakery industry. *Journal of American Oil Chemist Society* 60: 1848-1851.

Cicero, A.F.G and Gaddi, A. 2001. Rice bran oil and gamma Oryzanol in the treatment of hyperlipoproteinaemias and other conditions. *Phytotherapy Research*. 15: 227-289.

Crevel, R.W.R., kerlhoff, M.A.T and koning, M.M.G. 2000. Allergihicity of refined vegetable oils, *Food and chemistry Toxicology*. 38: 385-393.

Danielskia, L., Zetzla, C., Henseb, H and Brunnera, G. 2005. A process line for the production of raffinated rice oil from rice bran. *The Journal of Supercritical Fluids*. 34: 133–141

Darweesh, L.L., Toma, R.B and Lee, H.C. 1991. Chemical and sensory evaluation of lipid blends used in pie crusts. *Food Chemistry*. 39: 87-98.

Deckere, E.A.M and Korver, O. 1996. Minor constituents of rice bran oil as functional foods. *Nutrition Reviews*.11: 120-133.

Debnathb, S., Navin, K., Rastogi, A., Gopala Krishnab, A.G and Lokeshb, B.R. 2012. Effect of frying cycles on physical, chemical and heat transfer quality of rice bran oil during deep-fat frying of poori: An Indian traditional fried food. *Food and Bioproducts Processing*. 90: 249–256.

Dhaka, V., Gulia, N., Ahlawat, K.S and Khatkar, B.S. 2011. Trans fats-sources, health risks and alternative approach - a review. *Journal of Food Science and Technology*. 48: 534-541.

Dietary guidelines for Indians-A Manual. 2010 National Institute of Nutrition, Hyderabad.

Doescher, L.C., Hoseney, R.C and Milliken, G.A. 1987. A Mechanism for cookie dough setting. *Cereal Chemistry*. 64: 158-163.

Dogan, I.S., Javidipour, I and Akan, T. 2007. Effet of interesterified palm and cottonseed oil blends on cake quality. *International Journal of Food Science and Technology*. 42:157-164.

Drake, M.A and Gerard, P.D. 2003. Consumer attitudes and acceptability of soy-fortified yogurts. *Journal of Food Science*. 63 (3): 1118-1122.

Dreher, M.L., Berglund, P.T and Dreher, C.J. 1983. Use of vegetable oils in pie crust. *Journal of Food Science*. 48: 1566-1567.

Eady, S., Wallace, A., Willis, J., Scott, R and Frampton, C. 2011. Consumption of a plant sterol based spread derived from rice bran oil is effective at reducing plasma lipid levels in mildly hypercholesterolaemic individuals. *The British Journal of Nutrition*. 105: 1808–1818.

Elif, Y., Gulum, S and Serpil, S. 2010. Quantitative analysis of macro and micro-structure of gluten-free rice cakes containing different types of gums baked in different ovens. *Food Hydrocolloids* 24(8): 755-762.

Farhoosh, R and Kenari, R. E. 2009. Anti-Rancidity Effects of Sesame and Rice Bran Oils on Canola Oil during Deep Frying. Journal of the American Oil Chemists Society. 86(6): 539-544.

Farkas, B.E., Singh, R.P and Rumsey, T.R. 1996. Modeling heat and mass transfer in immersion frying. I, Model development. *Journal of Food Engineering*. 29: 211-226.

Fat and Oils in Human Nutrition, Report of a Joint Expert Consultation; Food and Agriculture Organization of the United Nations and the World Health Organizations: 1993.

Ferreira,V.L.P,2000.AnáliseSensorial:Testesdiscriminativoseafetivos.*Manual:SérieQualidade*.Campinas:SBCTA.127.

Fischer, R.A and Yates, F. 1963. Statistical table for biological, agricultural and medical research. 6th edition. Edinberg: Oliver and Boyd.

Frenk,.D.F. 2012. Fats and Cholesterol: Out with the Bad, In with the Good. Havard School of Public Health. The Nutrition Source > What Should I Eat? http://www.hsph.harvard.edu/nutritionsource/what-should-you eat/fats fullstory/index.html.

Frenk, D.J. 2012. Shinning the Spotlight on Trans Fats. Harvard School of Public Health. The Nutrition Source > Nutrition in the News.

Funk, K., Zabik, M.E and Elgidaily, D.A. 1969. Objective measurements for baked goods. *Journal of Home Economics*. 61: 119-123.

Galli, F., Stabile, A.M., Betti, M., Conte, C., Pistilli, A., Rende, M., Floridi, A and Azzi, A. 2004. The effect of α- and γ-tocopherol and their carboxyethyl hydroxychroman metabolites on prostate cancer cells. *Archives of Biochemistry and Biophysics*. 423: 97-102.

Gerhardt, A.L and Gallo, N.B. 1998. Full fat rice bran and oat bran similarly reduce hypercholesterolaemia in humans. *Journal of Nutrition*. 128: 865-869.

Ghotra, B.S., Dyal, S.D and Narine, S.S. 2002. Lipid shortenings: a review. *Food Research International*. 35: 1015-1048.

Godber, J.S., Xu, Z., Hegsted, M and Walker, T. 2002. Rice bran and rice bran oil in functional foods development. *Louisiana Agriculture*. 45(4): 9-10.

Gopala, K.A.G. 2002. Nutritional Components of rice bran oil in relation to processing. *Lipid Technology*. 14: 80–84.

Hartnett, D.I and Thalheimer, W.G. 1979. Use of oil in baked products-Part II: sweet goods and cakes. *Journal of American Oil Chemists Society* 56: 948-952.

Hernandez, N., Alegría, R.M.E., Gonzalez, F and Lopez-Munguia, A. 2000. Enzymatic treatment of rice bran to improve processing. *Journal of the American Oil Chemists Society*.77: 177-180.

Hirahara, S and Simpson, J.I. 1961. Microscopic appearance of gluten in pastry dough and itsrelation to the tenderness of baked pastry. *Journal of Home Economics* 53:681-686.

Hoed, V.V., Depaemelaere, G., Ayala, J.V and Grevt, W.D .2006. Influence of chemical refining on the major and minor components of Rice bran oil. *Journal of the American Oil Chemists' Society*. 83(4): 315-321.

Hoseney, R.C and Rogers, D.E. 1994. Chapter 5. In: The science of cookie and crackerproduction. 1st edition. New York: Chapman & Hall. 203-225.

Hossain, S.M., Singh, K.A and Zaman, F. 2009. Cooking and eating characteristics of some newly identified inter sub-specific (indica/japonica) rice hybrids. *Science Asia.* 35: 320–325.

Hung, P.V., Maeda, T and Morita, N. 2007. Dough and bread qualities of flours with whole waxy wheat flour substitution. *Food Research International* 40: 273-279.

Iqbal, J., Minhajuddin, M and Beg, Z.H. 2004. Suppression of diethylnitrosamine and 2-acetylaminofluorene-induced hepatocarcinogenesis in rats by tocotrienol-rich fraction isolated from rice bran oil. *European Journal of Cancer Prevention.* 13(6): 515-520.

Iqbal, S., Bhanger, M.I and Anwar, F. 2005, Antioxidant properties and components of some commercially available varieties of rice bran in Pakistan. *Food Chemistry*. 93: 265-272.

Itani, T.T., Tamaki, M and Arai, E. 2002. Distribution of amylose, nitrogen, and minerals in rice kernels with various characters. *Journal of Agricultural and Food Chemistry.* 50: 5326–5332.

Jacob, J and Leelavathi, K. 2007. Effect of fat-type on cookie dough and cookie quality. *Journal of Food Engineering* 79: 299-305.

Jeyarani, T., Khan, I.M and Khatoon, S. 2009. Trans-free plastic shortenings from coconuts stearin and palm stearin blends. *Food Chemistry* 114: 270-275.

Jiang, Y and Wang, T. 2005. Phytosterols in cereal by-products. *Journal of the American Oil Chemists Society*. 82: 439-444.

Joo, S.K., Godber, J.S., King, J.M and Prinyawiwatkul, W. 2001. Inhibition of cholesterol autoxidation by the nonsaponifiable fraction in rice bran in an aqueous model system. *Journal of the American Oil Chemists Society*. 78 (7): 685-689.

Juliano, C., Cossu, M., Alamanni, M.C and Piu, L. 2005. Antioxidant activity of gamma-oryzanol: Mechanism of action and its effect on oxidative stability of pharmaceutical oils. *International Journal of Pharmaceutics*. 299: 146–154.

Kaur, A., Jassal, V and Bhise, S.R. 2014. Replacement of bakery shortening with rice bran oil in the preparation muffins. *African journal of Biochemistry Research*.8(7): 141-146.

Kilne, K., Yu, Y and Sanders, B.G. 2004. Vitamin E and breast cancer. *Journal of Nutrition*. 134: 345-346.

Kong, L. 2012.What a fat debacle: saturated and Trans fatty acids effect on health and the future of food manufacturing processes.*M.Sc. Thesis* Baylor University, Waco, Texas.

Latha, R.B and Nasirullah, D.R. 2014.Physico-chemical changes in rice bran oil during heating at frying temperature. *Food Science and Technology*. 51(2): 335–340.

Lee, J.W., Lee, S.W., Kim, M.K and Rhee, C. 2005. Beneficial effect of the unsaponifiable matter from rice bran on oxidative stress in vitro compared with a α-tocopherol. *Journal of the Science of Food and Agriculture*. 85: 493–498.

Lock, A.L and Bauman, D.E. 2004. Modifying milk fat composition of dairy cows to enhance fatty acids beneficial to human health. *Lipids*. 39: 1197–206.

Lichtenstein, A.H. 1997. Tran's fatty acids, plasma lipid levels, and risk of developingcardiovascular disease. *Circulation*. 95: 2588-2590.

Luh, B.S., Barber, S and Barber, C. 1991. Rice bran: chemistry and technology. In: Luh BS (ed) Rice production and utilization.Van Nostrand Reinhold, New York, 313.

Marangoni, A.G. and Garti, N. 2011. *Edible Oleogels Structure and Health Implications.*

Martini, S., Suzuki, A.H and Hartel, R.W. 2008. Effect of high intensity ultrasound on crystallization behavior of anhydrous milk fat. *Journal of American Oil Chemist Society*. 85: 621-628.

Matthews, R.H and Dawson, E.H. 1963. Performance of fats and oils in pastry and biscuits. *Cereal Chemistry*. 40: 291-302.

McClements, D.J. and Decker, E.A. 2010. *Lipídeos*. 4th edition. Porto Alegre: Artmed

McClements, D.J. 2007. Critical review of techniques and methodologies for characterization of emulsion stability. *Critical Reviews in Food Science and Nutrition* 47(7): 611-649.

McWilliams, M. 2008. *Experimental Perspectives*, 6e. Upper Saddle River, New Jersey: Pearson Prentice Hall.

Meilgaard, M., Civille, G.V and Carr, B.T. 1999. Sensory evaluation techniques (3rd edition). CRC Press.Boca Raton, Florida. ISBN 0-8493-0276-5.

Mellema, M. 2003. Mechanism and reduction of fat uptake in deep-fat fried foods. *Trends in Food Science & Technology.* 14: 364-373.

Miller, A and Engel, K.H. 2006. Content of g-oryzanol and composition of steryl ferulates in brown rice (*Oryza sativa L.*) of European origin. *Journal of Agricultural and Food Chemistry.* 54: 8127– 8133.

Most, M., Tulley, R., Morales, S and LeFevre, M. 2005. Rice bran oil, not fiber, lowers cholesterol in humans. *Americans Journal of Clinical Nutrition.* 81: 64-68.

Mozaffarian, D., Ascherio, A., Hu, F.B., Stampfer, M.J., Willett, W.C., Siscovick, D.S and Rimm, E.B. 2005 Interplay between different polyunsaturated fatty acids and risk of coronary heart disease in men. *Circulation* 111(2): 157-64

Mozaffarian, D., Katan, M.B., Ascherio, A., Stampfer, M.J and Willett, W.C. 2006. Tran's fatty acids and cardiovascular disease. *The New England Journal of Medicine*. 354: 1601–1613.

Musavi, A., Tekin, A. and Erinc, H. 2011. Formulation of trans-free margarines using hydrogenated and interesterified palm olein. *Journal of Oil Palm Research*. 23: 1153-1158.

Nicolosi, R.J., Ausman, L.M and Hegsted, D.M. 1991. Rice bran oil lowers serum total and low density lipoprotein cholesterol and apo B levels in nonhuman primates. *Atherosclerosis*. 88: 133–142.

NorAini, I., Embong, M.S., Abdullah, A and Oh, F.C.H. 1992. Characteristics and performance of some commercial shortenings. *Journal of the American Oil Chemists' Society*. 69: 912-916.

NorAini, I and Miskandar, M.S. 2007. Utilization of palm oil and palm products inshortenings and margarines. *European Journal of Lipid Science and Technology*. 109: 422-432.

Normen, E., Rovedo, C.O and Singh, R.P. 1998. Mechanical properties of an immersion fried potato starch-gluten gel during post frying period. *Journal of Texture Studies*. 29: 681-697.

Norton, D., Angerman, S., Istfan, N., Lopes, S.M., Babayyan, V.K., Putz, M.C., Steen, S.N and Blackburn, G.L. 2004. Comparative Study of Coconut Oil, Soybean Oil, and Hydrogenated Soybean Oil. *PJCS, 29 (1 and 2)*.

Oko, A.O and Ugwu, S.I. 2011. The proximate and mineral compositions of five major rice varieties in Abakaliki, south-eastern Nigeria. *International Journal of Plant Physiology and Biochemistry*. 3: 25–27.

Orthoefer, F.T. 1996. Rice bran oil: healthy lipid source, *Food technology*. 50: 62-64.

Ou, S and Kwok, K.C. 2004. Ferulic acid: pharmaceutical functions, preparation and application in foods. *Journal of the Science of Food and Agriculture*. 84: 1261-1269.

Ozkoc, S.O., Sumnu, G and Sahin, S. 2009. The effects of gums on macro and micro-structure of breads baked in different ovens. *Food Hydrocolloids* 23: 2182-2189.

Pandey, M.K., Mishra, K.K., Khanna, S.K and Das, M. 2004. Detection of polycyclic aromatic hydrocarbons in commonly consumed edible oils and their likely intake in the Indian population. *Journal of the American Oil Chemists' Society* 81(12): 1131–1136.

Pareyt, B and Delcour, J.A. 2008. The role of wheat flour constituents, sugar, and fat in lowmoisture cereal based products: a review on sugar-snap cookies. *Critical Reviews in Food Science and Nutrition*.48: 824-839.

Pareyt, B., Brijs, K and Delcour, A.J. 2010. Impact of fat on dough and cookies properties of sugar-snap cookies. *Cereal Chemistry* 87: 226-230.

Patel, M and Naik, S.N. 2004. Gamma–oryzanol from rice bran oil-A Review. *Journal of Scientific and Industrial Research*. 63: 569-578.

Patient, D. 1994. The chemistry of pastry products. *Nutrition and Food Science*. 94: 33-35.

Pyler, E.J and Merriam, K.S. 1988. Sosland Publishing Company. *Baking science and technology*. 2(3): 984-987.

Qureshi, A.A., Sami, S.A., Salser, W.A and Khan, F.A. 2002. Dose dependent suppression of serum cholesterol by tocotrienol-rich fraction (TRF250 of rice bran in hypercholesterolaemic humans. *Atherosclerosis* 161: 199-207.

Raghuram, T.C and Rukmani, C. 1995. Nutritional significance of rice bran oil. *Indian Journal of medical research* 10: 593–601.

Rahman, M.S. 1995. *Food Properties Handbook*. CRC Press, Boca Raton. FL. USA.

Ramachandran, P. 1989. Nutrition in Pregnancy. In C. Gopalan and S. Kaur (eds.) - Women and Nutrition in India, Nutrition Foundation of India, New Delhi. Special Publication No. 5. 155.

Ramcharitar, A., Badrie, N., Mattfeldt-Beman, M., Matsuo, H and Ridley, C. 2005. Consumer acceptability of muffins with flaxseed (*Linumusitatissimum*). *Journal of Food Science.* 70 (7): s504-s507

Rangaswamy, B.L and Nasirullah, D.R. 2014. Physico-chemical changes in rice bran oil during heating at frying temperature. *Journal of Food Science Technology*. 51(2): 335–340.

Remig, V., Franklin, B., Margolis, S., Kostas, G., Nece, T and Street, J. 2010. Trans Fat in America: A Review of Their Use, Consumption, Health Implications and Regulation. *Journal of the American Dietetic Association.* 110 (4): 585-592.

Rimac-Brncic, S., Lelas, V., Rade, D and Simundic, B. 2004. Decreasing of oil absorption in potato strips during deep fat frying. *Journal of Food Engineering.* 64: 237–241.

Rios, R.V., Pessanha, M.D.F., Almeida, P.F.d., Viana, C.L and Lannes, S.C.D.S.2014Application of fats in some food products. *Food Science and Technology* (Campinas). 34: 3-15.

Rogers, D. 2004. Functions of fats and oils in bakery products. *American Oil Chemists Society.*15: 572-574.

Rukmini, R. 1988. Chemical, nutritional and toxicological studies of rice bran oil. *Food Chemistry.* 30: 257-268.

Rukmini, C and Raghuram, T.C. 1991. Nutritional and biochemical aspects of the hypolipidemic action of rice bran oil: a review. *Journal of the American College of Nutrition.*

Rutkowska, J and Zbikowska, A. 2010. Effects of fatty acid composition of liquid margarines on sensory quality of cakes. *Acta Alimentaria*. 39 (2): 125–137.

Sahri, M.M and Dian, N.L.H.M. 2011. Formulation of trans-free and low saturated margarine. *Journal of Oil Palm Research*. 23: 958-967.

Sakin, M., Kaymak-Ertekin, F and Ilıcalı, C. 2004. A numerical model for cupcake baking. *9th International Congress on Engineering and Food*, 7-11 Montpellier, France. 479-484.

Sakin, M., Kaymak-Ertekin, F.and Ilıcalı, C. 2007. Drying curves of white layer cake during baking in a convective oven. 5th International Congress on Food Technology. *Consumer Protection Through Food Process Improvement & Innovation In The Real World*, 9-11 March 2007, Thessaloniki, Greece, 1:146-153.

Semma, M. 2002.Trans Fatty Acids: Properties, *Benefits and Risks Journal of Health Science*. 48(1) 7–13.

Sen, C.K., Khanna, S and Roy, S. 2006. Tocotrienols: Vitamin E beyond tocopherols. *Life Science*. 78: 2088–2098.

Sen, C.K., Khanna, S and Roy, S. 2007. Tocotrienols in health and disease: The other half of the natural vitamin E family. *Molecular Aspects of Medicine*. 28: 692–728.

Sevimli, M., Sumnu, G and Sahin, S. 2005. Optimization of halogen lamp-microwave combination baking of cakes: a response surface methodology study.*European Food Research Technology*. 221: 61-68.

Sharma, A. R. 2002. Edible rice bran oil - consumer awareness programme. Rice bran oil promotion committee. Solvent Extractors Association of India, Mumbai.

Sharma, H.K., Kaur, B., Sarkar, B.C and Singh, C. 2006. Thermal behavior of pure rice bran oil, sunflower oil and their model blends during deep fat frying. *Grasas y aceites*. 57 (4): 376-381.

Sharma, R.D and Rukmini C. 1987. Hypocholesterolemic activity of unsaponifiable matter of rice bran oil. *Indian Journal of Medical Research*. 85: 278–281.

Sharif, K., Masood, S.B., Faqir, M.A., and Muhammed, N. 2005. Improved Quality of Baked Products By Rice Brain Oil. *M.Sc Thesis, Institute of Food Science & Technology, University of Agriculture Faisalabad, Pakistan*.

Sharif, K., Masood, S.B., Faqir, M.A., Nasir, Rashid, M and Qayyum, M.N. 2003. Extension of Cookies Shelf Life by Using Rice Bran Oil. *International Journal of Agriculture & biology*. 455–457.

Shiela, P.M., Sreerama, Y.N and Gopala-Krishna A.G. 2004. Storage Stability Evaluation of some Packed Vegetable Oil Blends. *Journal of the American Oil Chemists Society*. 81(12): 1125-1129

Shih, F.F., Daigle, K.W and Clawson, E.L. 2001. Development of low oil-uptake donuts. *Journal of Food Science*. 66(1): 141-144.

Shih, F.F., Bett-Garber, K.L., Daigle, K.W and Ingram, D. 2005. Effects of Rice Batter on Oil Uptake and Sensory Quality of Coated Fried Okra. *Journal of Food Science*. 70(1): S18-21.

Shin, D.H and Chung, J.K. 1998. Changes during storage of rice germ oil and its fatty acid composition, Korean. *Journal of Food Science and Technology*. 30: 77–81.

Sierra, S., Villoslada, L.F., Olivares, M., Jimenez, J., Julio, B and Xaus, J. 2005. Increased immune response in mice consuming rice bran oil. *European Journal of Nutrition*. DOI 10.1007/s00394-005-005-0554-y.

Singh, A., Sharma, H.K., Sarkar, B.C., Singh, C and Shitandi, A.A. 2007. Thermal behaviour of pure rice bran oil, safflower oil and their model blends during deep fat frying. *Journal of Food Science and Technology*. 44: 52–55.

Singh, N., Kaur, L., Singh Sodhi, N and Singh Sekhon, K. 2005. Physicochemical, cooking and textural properties of milled rice from different Indian rice cultivars. *Food Chemistry*. 89: 253–259.

Stauffer, C.E. 1998. Fats and oils in bakery products. *Cereal Food World*. 43: 120-126.

Stender, S., Astrup, A and Dyerberg, D. 2008. Ruminant and industrially produced trans fatty acids: health aspects. *Food and Nutrition Research*. 52: 12.

Sugano, M and Tsuji, E. 1997. Rice bran oil and cholesterol metabolism. *Journal of Nutrition*. 127: 521-524.

Sumnu, G., Ndife, M. K and Bayindirli, L. 2000. Optimization of microwave baking of model layer cakes. *European Food Research and Technology*. 211:169–174.

Sumnu, G., Sahin, S and Sevimli, M. 2005. Microwave, infrared and infrared-microwave combination baking of cakes. *Journal of Food Engineering*. 71 (2), 150-155.

Sundararaj, P., Siddhu, A and Valsalan, A. 2002. Storage and frying stability of rice bran oil. *Saarc Oils and Fats Today*. 9-14.

Tahira, R., Rehman, A and Butt, M.A. 2007. Characterization of rice bran oil. *Journal of agricultural research*. 45(3).

Tan, K.J and Mittal, G.S. 2006. Physiochemical properties changes of donuts during vacuum frying. *International Journal of Food Properties*. 9: 85-98.

Tarrago-Trani, M.T., Phillips, K.M., Lemar, L.E and Holden, J.M. 2006. New and existing oils and fats used in products with reduced trans-fatty acid content. *Journal of the American Dietetic Association*. 106: 867- 880.

Taylor, J.B., Richar, T.M., Wilhelm C.L., Chrysam, M.M., Otterburn, M and Leveille, G.A. 1996. Rice bran oil antioxidant. *U.S. Patent No*. 5,552,167.

Trans fat; *American Heart Association*: 2014.

Turabi, E., Sumnu, G and Sahin, S. 2008. Optimization of baking of rice cakes in infrared-microwave combination oven by response surface methodology. *Food Bioprocess Technology*.1: 64-73.

Turpeinen, A.M., Mutanen, M., Aro, A., Salminen, I., Basu, S., Palmquist, D.L and Griinari, J.M. 2002. Bio-conversion of vaccenic acid to conjugated linoleic acid in humans. *Americal Journal Clinical Nutrition*. 76: 504–510.

U.S. Department of Agriculture and U.S. Department of Health and Human Services. Dietary Guidelines for Americans, 2010. 7th edition. Washington, DC: U.S. Government Printing Office, December 2010.

Valantina, R., Sahayaraj, P.A and Prema, A.A. 2010. Antioxidant stability in palm and rice bran oil using simple parameters. Rasayan J Chem. 3(1): 44–50.

Velez-Ruiz, J.F and Sosa-Morales, M.E. 2003. Evaluation of physical properties of dough of donuts during deep-fat frying at different temperatures. *International Journal of Food Properties*. 6(2): 341–353.

Vetter, J.L., Bright H., Utt, M and McMaster G. 1984. Sugar, mixing affect specific gravity, spread. *Bakers Dig* 40: 40-45.

Vissers, M., Zock, P.L., Meijer, G.Wand Katan, M.B. 2000. Effect of plant sterols from rice bran oil and triterpene alcohols from sheanut oil on serum lipoprotein concentrations in humans. *American journal of clinical Nutrition*. 72: 1510-1515.

Vitrac, O., Dufour, D., Trystram, G and Raoult-Wack, AL. 2002. Characterization of heat and mass transfer during deep-fat frying and its effect on cassava chip quality. *Journal of Food Engineering*. 53: 161-176.

Vliet, T., Visser, J.E and Luyten, H. 2007. On the mechanism by which oil uptake decreases crispy/crunchy behavior of fried products. *Food Research International*. 40: 1122–1128.

Wada, S., Satomi, Y., Murakoshi, M., Noguchi, N and Yoshikawa, H. 2005. Tumor suppressive effects of tocotrienol in vivo and in vitro. *Cancer Letters* 229: 181-191.

Wainwright, B. 1999. Oil and fats for the baking industry. *Cereal Food World*. 44: 16-19.

Warner, K., Miller, J and Demurin, Y. 2008. Oxidative stability of crude mid oleic sunflower oils from seeds with high g and d-tocopherol levels. *Journal of American Oil Chemists Society*. 85: 529–533.

Wassell, P and Young, N.W.G. 2007. Food applications of Trans fatty acid substitutes. *International Journal of Food Science and Technology*. 42: 503-517.

Wheeler, F.G and Stingley, D.V. 1963. Effect of moisture on fat absorption in cake doughnuts. *Cereal Science Today*. 8: 102–108.

Wilson, T.A., Nicolosi, R.J., Woolfrey, B and kritchevsky, D. 2007. Rice bran oil and Oryzanol reduce plasma lipid and lipoprotein cholesterol concentrations and aortic cholesterol ester accumulation to a greater extent than ferulic acid in hypercholesterolaemic hamsters. *Journal of Nutritional Biochemistry*. 18: 105-112.

Yokoyama, W.H. 2004. Plasma LDL cholesterol lowering by plant phytosterols in a hamster model. *Trends in Food Science and Technology*. 15: 528–531.

Yilmazer, S., Isleroglu, H., Kemerli, T., Ozdestan, O., Guven, G., Kaymak-Ertekin, F., Uren, A and Ozyurt, B. 2011. Quality characteristics and drying behavior of muffins baked in steam assisted and convectional ovens. In proceedings of the 11th International Congress on Engineering and Food, Athens, Greece.

Zhou, J., Jon, M.F and Chuck, E.W. 2011. Evaluation of different types of fats for use in high ratio layer cakes. *Food Science and Technology* 44: 1802-1808.

Zhou, Z., Robards, K., Helliwell, S and Blanchard, C. 2002. Composition and functional properties of rice. *International Journal of Food Science Technology*. 37: 849–868.

Zigoneanu, I.G., Williams, L., Xu, Z and Sabliov, C.M. 2008. Determination of antioxidant components in rice bran oil extracted by microwave-assisted method. *Bioresource technology*. 99: 4910-4918.

APPENDICES

Appendix A

Score card for sensory evaluation

Post Graduate & Research Centre
Prof. Jayashankar Telangana State Agricultural University

Score card for sensory analysis of baked products

Name: Date: Time:
Age: Sex: Occupation:

Instructions: You are provided with baked product samples. Please evaluate the samples using 9 point hedonic scale. Write the preferred score in the column as per evaluation. Rinse your mouth between evaluations of each sample.

Hedonic scale:

9 Like extremely
8 Like very much
7 Like moderately
6 Like slightly
5 Neither Like nor Dislike
4 Dislike slightly
3 Dislike moderately
2 Dislike very much
1 Dislike extremely

Sample code No.	Appearance	Colour	Flavour	Texture	Taste	Overall acceptability

Comment:

Signature of the judge

Appendix B

Consumer acceptability questionnaire

POST GRADUATE & RESEARCH CENTRE
PROF. JAYASHANKAR TELANGANA STATE AGRICULTURAL UNIVERSITY

CONSUMER ACCEPTABILITY QUESTIONNAIRE

SECTION- A: DEMOGRAPHIC PROFILE

1. Name:
2. Age:
   i. 18-30 ☐
   ii. 30-40 ☐
   iii. 40-50 ☐
   iv. 50-60 ☐

3. Education:
   i. Primary ☐
   ii. Secondary ☐
   iii. Higher Secondary ☐
   iv. UG ☐
   v. PG ☐
   vi. Doctorate ☐
   vii. Other ☐

4. How frequently do you consume Rice bran oil?
   i. Daily ☐
   ii. Weekly ☐
   iii. Monthly ☐
   iv. Rarely ☐
   v. Never ☐

5. How much of rice bran oil do you consume per month (in number of liters)?
   i. 1-2 ltrs ☐
   ii. 3-5 ltrs ☐
   iii. 5-10 ltrs ☐

6. Are you aware of Rice Bran Oil spread (RBOS)?
   i. Yes ☐ ii. No ☐

7. Have you consumed any product developed with RBOS?
   ii. Yes ☐ ii. No ☐

8. Do you have any knowledge about the health benefits of RBOS?
   i. Yes ☐ ii. No ☐

9. Are you willing to buy RBOS products?
   i. Yes ☐ ii. No ☐

10. How much price are you willing to pay per liter of RBOS?
    i. Rs. 80 or less ☐
    ii. Rs. 100 ☐
    iii. Rs. 150 ☐
    iv. Rs. 150 or more ☐

SECTION- B: HEDONIC SCORING

Instructions: You are provided with two cake samples. Please evaluate the samples using 5 point hedonic scale. Write the preferred score in the column as per evaluation.

Hedonic rating system for the level of acceptance for color, odor, taste & overall acceptability:

Hedonic rating for level of acceptance	Corresponding descriptor
5	Like Very Much
4	Like Moderately
3	Neither Like nor Dislike
2	Dislike Moderately
1	Dislike Very Much

Location	**Hedonic rating**					
	Colour	Texture	Sponginess	Taste	Flavour	Overall acceptability
Sample 643						
Sample 752						

*****Thank you*****