DETERMINATION OF BIOACTIVE COMPONENTS, FIBER CONTENT FROM ALMOND SKINS AND FORMULATION OF SUPPLEMEND ALMOND SKIN POWDER CAKE

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Roll No.: 0119/26

Registration No.: 684

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A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Applied Human Nutrition and Dietetics

Department of Applied Food Science and Nutrition

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This is to certify that we have examined the above Master's Thesis and have found that it is complete and satisfactory in all respects, and that all revisions required by the thesis examinations committee have been made.

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DEDICATED TO MY RESPECTED AND BELOVED FAMILY AND TEACHERS

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Abbreviation

AOAC	: Association of Official Analytical Chemists
ANOVA	: Analysis of Variance
B.P	: Boiling point
℃	: Degree celcius
DPPH	: 2,2-diphenyl–1–picryl hydrazyl
et al	: Et alli/et aliae/et alia
MUFA	: Mono Unsaturated Fatty Acid
PUFA	: Poly Unsaturated Fatty Acid
SD	: Standard Deviation
SPSS	: Statistical Package for Social Science
%	: Percentage

Abstract

Almond skins are by-products which contains a rich amounts of protein, fiber, vitamins other bioactive compounds. The study aimed to properly utilize almond skin as a supplemental nutritious food i.e. supplemented almond fiber. Bioactive Compounds like antioxidant capacity, total Phenolic content, flavonoids contents were also determined and found the values (0.04± 0.01) , (1.39± 0.003) , (0.27± 0.05) mg/100gm. respectively. The fiber content of almond skin powder was recorded (14.6± 0.04) %. The proximate compositions of cake formulas were determined. The cake formulas were prepared with three different substitution levels of almond skin powder (sample B=20 %, sample C=25 % and sample D=30%) in all purpose flour . All of these formulas were compared with control sample A which contained 0% almond skin powder. The highest moisture content was recorded in sample D (25.9±0.04) % which was substituted with 30 % almond skin powder. The lowest value founded in sample B (25.2±0.04) %. Sample D showed the highest protein value (7.7±0.04)% and sample C is the lowest (7.4±0.04) %. Sample A obtained the highest fat value (19.5±0.04) % and the lowest value observed in sample D (18.2±0.03) %. Sample D also showed the highest fiber and ash content (6.3±0.03) % and (0.7±0.04) %. Sample A scored the lowest as it contain 0% almond skin powder. The highest CHO value was (44.8±0.04) % scored by sample A and the lowest value by sample D (40.9±0.04)%. Sample D supplemented with 30% incorporation obtained the best acceptance (5.8±0.86) by panelists where sample A the lowest (4.7±1.003). Energy content of sample A was calculated in the highest amount (390.4 kcal/100 gm.) and the lowest in sample D (358.2 kcal/100gm.). Healthy cake formulas can be successfully prepared from almond skin powder.

Keywords: Almond, cakes, proximate, sensory, antioxidant capacity, malnutrition.

Chapter 1: Introduction

Almonds (*Amygdalus communis L*.) belongs to the family of rosaceae are valuable source of nutrients and phytochemicals like vitamin E, monounsaturated fatty acids, polyunsaturated fatty acids, arginine, potassium, these mutrients are helpful to reduce heart and obesity related diseases (Nuts, Almonds, 2007). Other health promoting compounds found in almonds are polyphenols, which have been shown to be capable of protecting cancer and cardiovascular diseases (Liu,2004 ;Yang et al.,2009). Besides, almond skins are enriched with fiber, which is considered as a useful ingredient for the control of oxidative processes in food products and as a functional food ingredient (Monagas et al., 2007). Dietary fiber consists of intrinsic plant cell-wall polysaccharides which are not broken down by endogenous enzymes in the upper gastrointestinal tract. However, through the fermentation process in the large bowel, cell-wall polysaccharides exert a major control on colonic function, including bowel habit, transit times, metabolism and balance of the commensal flora and large bowel epithelial health(Cummings and Stephen, 2007). Almonds are rich with 12.2% fiber and recognized as one of the highest fiber contents of all the edible nuts (Holland et al., 1991). Almond skins can be considered functional food ingredients because they contain several bioactive phenolic compounds, namely flavonoids, phenolic acids, and tannins, the latter both hydrolysable and condensed. The phenolic content of fresh almond skins comprises between 11.1 and 17.7 mg/g, depending on the extraction protocol, whereas 0.25–0.85 mg/g dm. (dry matter) quantified in dried almond skins, with the lowest amount in sun-dried skins and the highest in skins oven-dried at a temperature of 45–60. It is recommended regularly as an addition to a healthy and balanced diet. According to the peer-reviewed Nutrients journal, epidemiological studies and clinical trials have shown positive effects of nuts consumption against a significant number of pathologies such as obesity, hypertension, diabetes mellitus and metabolic syndrome. In addition, people who consume nuts regularly present lower waist circumference and improved metabolic profiles. The demand of nutritionally healthy and sustainable viable foods has been increased considerably. Special attention has been provided to the utilization of by-products. The proper use of by-products can contribute to new food products and minimize waste. Almond skin is inexpensive and widely distributed. Almond skin can be diversified into a wide range of processed products that can increase peel consumption. Snacks are popular food

among all ages of people. Snack food plays a vital role in providing supplemental nutrition and therefore, snack should contain complete nutrient content, including carbohydrates, protein, fat, vitamins and minerals (Hapsari, 2013).

The demand of nutritionally rich and healthy food items has been increased gradually. The proper utilization of by-products can contribute to minimization of waste as well as development of new products. Snacks can be beneficial in a diet like increasing nutrient intake, sustaining energy level and giving individuals with plenty of healthy options. Certain snacks can be beneficial to improve the quality of diet. Sometimes people restrict themselves to eating cakes because of increasing body weight and concerning about health issues. But they can hardly realize that in moderation and good quality cake is actually beneficial to our health. It offers enough energy to our body as it consists of flour, sugar, both of them contains carbohydrates, which are primary source of energy of our body. Some cake contains fruits or other fiber rich content , helps to have better digestive systems & minimize the risk of heart diseases. According to the research, by the institute of optimum nutrition, cakes help to improve mood level by releasing endorphins chemicals into our body that helps to remove stress & anxiety and bring the feeling of happiness . In addition, the almond skin powder cake formulation can be modified easily to meet the nutritional demands of target customers.

That’s why this study aims to formulate cakes that are rich in fiber and can be taken as a healthier option than other cakes. Almonds are consumed by some people of Bangladesh. In previous there was no work done and so this study is designed to utilize almond skin properly by developing cakes supplemented with almond skin powder for nutritional improvement and evaluating the nutritional contents and organoleptic attributes of supplemented cake.

Aims and objectives

- To determine the proximate compositions, fiber contents, bioactive components (polyphenols, flavonoids, antioxidant capacity) of almond skin powder.
- To formulate the cakes supplemented with almond skin powder.

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Chapter 2: Review of literature

Almonds are super essential element for human body as they offer a lot of nutritional benefits. The skin of almonds are rich in fiber due to the presence of polyphenols and helps in digestion and nourishes skin. Fortification of bakery products refers to the addition of certain functional ingredients to enhance its nutritional value. This fortification performs a specific therapeutic function in human body after consumption. So the formulation of cake from almond skin can be a healthier choice to meet the nutritional demand due to this fortification.

2.1 Overview of almonds

Nuts such as almonds are intrinsic to dietary guidelines in several countries owing to increasing beneficial health effects. Epidemiological studies suggests that people who consume nuts frequently at least five times in a week, tend to have lower body mass indices (Garcia-Lorda et al.,2009). Due to high fiber and protein content, almonds have a lower glycemic index, which helps to increase satiety and suppress appetite (Holt et al.,1995). Human studies involving mixed nuts have been directed in six countries like Australia, Canada, Israel, India, New Zealand and United states. The result found significant reductions in plasma total cholesterol (7-25%) and plasma LDL cholesterol (10-33%) which reviewed by (Ternus et al.,2009). People call almonds a nut, but they are seeds, rather than a true nut.

2.2 Scientific classification of almonds

Family: Rosaceae

Kingdom: Plantae

Genus: Prunus

Subgenus: *Prunus* subg. *Amygdalus*

Species: *P. amygdalus*

Figure 2.1 Raw almonds and skin

Source: (https://www.istockphoto.com/photos/almond)

Almonds belongs to the family rosaceae .Usually people consider almond skin as waste which are mostly used as cattle feed or compost .However, it can be considered as functional food ingredients because they contain several bioactive and phenolic compounds like flavonoids, antioxidants (chen et al., 2005). Furthermore, almond skins are also a vital source of fiber and therefore have prebiotic effects. Almond skin can be used to functionalize foods to represent an example of up cycling, responding to the need to increase sustainability in the food industry within the framework of the principles of a circular economy (Rondeau et al., 2020). Functional ingredients like almond skins could be easily added to cereal based products as to fulfill the consumer expectations for healthy and pleasant foods. Cakes are popular baked goods, eaten easily and characterized by a long shelf life. These features make this product a good recipient for the addition of functional ingredient (pasqualone et al.,2018).

Table 2.1 : Nutrient value per 100 gm. of blanched almond

Principle	**Nutrient Value**	**% Daily values ***
Carbohydrates	19.94 g	7%
Protein	21.94 g	9%
Total fat	50.62 g	65%
Cholesterol	0 mg	0%
Dietary fiber	10.4 g	37%
Vitamin C	0 mg	0%
Vitamin A	0 mg	0%
Vitamin E	23.75 mg	2%
Sodium	28 mg	1%
Potassium	687 mg	15%
Iron	216 mg	17%
Calcium	3.72 mg	21%

Source : United States Dept. of Agriculture National Data base (2008)

2.3 Constituents of almond skin

Recently the reuse of by-products has become very popular or particularly important research topic to develop systems capable of minimizing environmental impact and waste resources. Almond skin powder is particularly rich in fiber (52.69/100 g) as shown by the characteristics of its nutritional compositions. This byproduct of almond processing also indicated a relevant presence of lipid (21.3 gm./ 100 gm..). The lipid fraction of almond skins being composed mainly Mono Unsaturated Fatty Acid (MUFA) and Poly Unsaturated Fatty Acids (PUFA) associated with higher amounts of vitamin E (Mandalari et al., 2010). Protein content of almond skin accounted for about (119/100g) and low amounts of carbohydrate were found. The lipid composition of almond skin parallels the lipid content of whole seed (Summo et al., 2018).

2.4 Fiber content of almond skin

According to the New European Regulations, whole natural almond contain about 12 gm. dietary fiber per 100 gm. which can be considered as sufficient as to claim 'naturally high in fiber'. Plant cell wall are supra molecular networks cellulose, hemi-cellulose, pectic substances & phenolic compounds which is a major source of dietary fiber (Ellias et al., 2004). Furthermore, there is a general concept based on epidemiological and human intervention studies that dietary fiber from plant cell wall like cereal, fruits, vegetables, nuts are associated with a wide range of health benefits including reducing the risk of CHD, diabetes and positive effects on digestive systems. A typical serving of almonds (28-30 gm.) contains about 14% of the daily fiber requirements.

2.5 Antioxidant capacity of almond skin

Antioxidants are compounds which can delay or inhibit the oxidation of lipid by inhibiting the initiation of oxidizing chain reactions (Velioglu, Mazza, Gao & Oomah, 1998). Food plants like fruits, vegetables, herbs, spices, nuts are primary source o0f naturally occurring anti-oxidants for humans (Loliger, 1991; Sang, Chen, et al.,2002). Antioxidants present in food commodities helps to decrease oxygen

concentrations, prevent first chain initiation by scavenging initial radicals and decompose primary products of oxidation to non-radical compounds (Shahidi, 1997). Nuts are one of the most valuable source of antioxidants that are very known source to possess health promoting contents (Andreasen, Landbo, Christensen, Hansen & Meyer, 2001).

2.6 Use and applications of almond skin by products

Almond skin are agricultural by products which is a source of phenolic compounds and can be produced by processing of almonds. Besides, almond skin, resulting from hot water blanching process can be used as animal feed or burned as fuel in processing plants (Harrioson & Were, 2007). The skin consists of about 4% of the almond fruit & it is a potential source of phenolics (Chen et al.2005). It inhibits oxidation by scavenging free radicals, activating antioxidant enzymes & inhibiting enzymes that cause oxidation reactions (Heim, Tag liaferro & Bobliya, 2002). Almond skin powder can be used to fortify or produce various bakery items like cake, bread , biscuits and can be consumed as snacks. Almond skin powder is regarded as functional, healthy & sustainable food ingredients due to its fiber, bio-active components and energy content. Selecting healthy snack like containing almond skin powder can increase energy level & balance hunger by providing regular fuel to the body.

2.7 Health benefits of almond skin

Almond skin is a rich source of vitamin E which is an antioxidants that protect cells from getting toxic. Antioxidant protest against stress that leads to molecule damage and thus results in inflammation, cancer and ageing. However, consumption of 84 grams of almonds per day is proven to increase the levels of antioxidants in body that fight against ageing. Almond skin contains flavonoids this component nourishes skin and is an anti –ageing property for skin. Almond skins are considered to be the best remedy for skin that can prevent acne, blackheads and stretch marks by nourishes the skin. Lack of magnesium in body leads to loss of hair. Almond skin contains magnesium helps to grow hair fast as well as curing dandruff and other types of hair problems. Almond skin have high amounts of fiber that tends to help

detoxifying the body. Besides, the presence of this amount fiber reducing the risk of colon cancer. Almond skin contains a certain amount of magnesium that benefits the nerves system as well as developing a healthy metabolic rate.

2.8 History of snack food

Now the industries are focusing on producing healthier foods for snacking like low oil, sodium and low calorie foods. Peanuts and popcorn are considered Americans first commercial snack food which were tasty, cheap and easily portable. Between 1950 and 2000, the United States became a snack lover. The people started consuming snack everywhere- at home, work, school, while in the car or walking down the sidewalk. Thousand varieties of snacks like chips, cookies, cake or anything that our heart desire are available (Mc carthy, 2001)

2.9 Cake and health benefits

Flour, sugar, milk, extra flavorings can all be found in cake's ingredient list. All of the elements listed here are required by our bodies daily activities. Flour and sugar both contains carbohydrate which is the body's primary source of energy. Besides, milk and egg added to it, are naturally rich in protein and calcium which is beneficial for healthy bone and teeth. The cake fortified with fruit or other functional ingredients contains fiber and minerals which helps to increase digestibility. Not only that, but the fruits in the cake also helps to lower the risk of heart disease.As a part of healthy balanced diet, there are some health benefits of eating cake. It can be a wonderful addition to diet.

2.9 Evaluating the baking quality of cake

The acceptability of cake depends on texture, appearance, taste, volume etc. Sensory panels are needed to examine for these attributes. In theses panels, people taste the given samples as well as describe their opinion regarding the overall acceptability of the product. The observed values showed that the consumer acceptability was higher for those formulas which are supplemented with fiber rich almond powder. This

factor indicates a better quality and is connected to an increase in consumer acceptability (kluklin et al., 2018)

Conclusion

Although almond skins are not widely used, these are beneficial due to their chemical composition, therapeutic and industrial properties. The popular nut like almonds are used to produce oil, in the bakery and can be consumed as a snack. Almond skin powder is regarded as a functional, healthy and sustainable food ingredient due to its fiber, protein, fat and energy content. Selecting a healthy snack in a good portion can increase energy level by balancing hunger and provides regular fuel to the body.

Chapter -3: Materials and method

3.1 Study area

The research was conducted in the laboratory of Department of Applied Food Science and Nutrition, Department of Food Processing and Engineering, Department of Animal Sciences and Nutrition, Department of Fishing and Post harvest Technology in the Chattogram Veterinary And Animal Sciences University located at Khulshi, Chattogram.

3.2 Collection of raw materials

Almonds were purchased from super shop Basket located at Khulshi, Chittagong . Other ingredients of cake like flour, baking powder, oil, egg, icing sugar, vanilla flavorings were purchased from local market of khulshi.

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3.3 Experimental design:

After the collection of raw materials, it was used for preparation of almond skin powder .Then the powder was used for formulation of cake and determination of crude fiber, bioactive components (Polyphenols, flavonoids, antioxidant). Proximate analysis was done for the cake samples Prepared for almond skin powder.

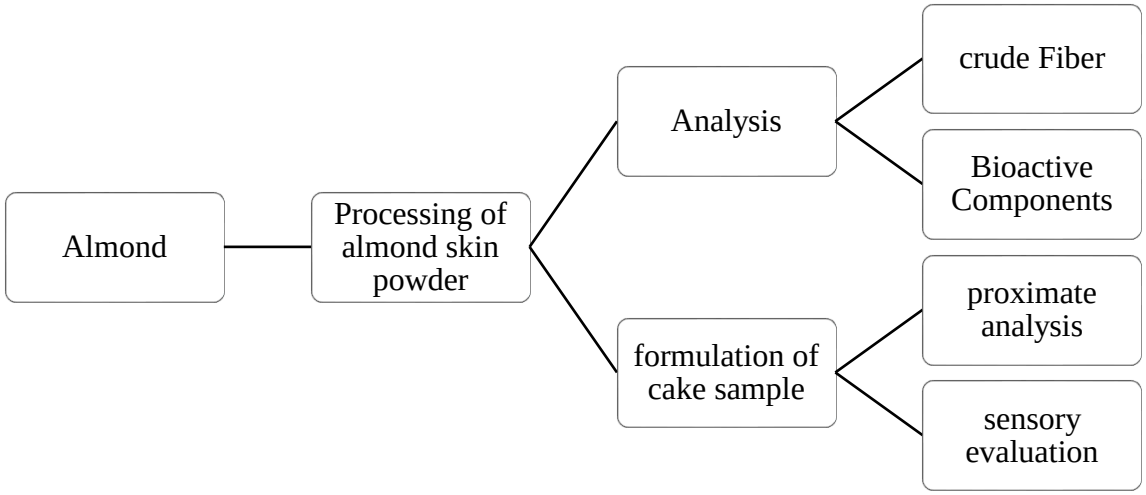


Figure 3.1 Experimental design

3.4 Almond skin powder preparation

Almonds were soaked in boiling water 100 degree Celsius temperature for five minutes. After cooling, it was pressed gently to separate the skin from flesh between two fingers. Then the removed skin was kept in the cabinet dryer for 24 hours at 60 degree Celsius temperature the cabinet dried sample was ground and weighted. The yield was considered as almond skin powder. Accurate quantity was weighted as required for different analysis.

3.5 Determination of bioactive compounds of almond skin powder

3.5.1 Determination of Total Phenolic Content (TPC)

At first, stock solution was prepared which was done by dissolving 10 mg of Gallic acid into the 10 ml of distilled water. It makes the concentration of solution 1mg/ml. Then for the preparation of the different concentration solution (2ppm, 4 ppm,8 ppm, 16ppm, 32ppm), Serial dilution was performed. The total phenol content of extracts was determined by the Folin-ciocalteu method. About 1 ml of sample extracts for standard different concentrations were mixed with 2ml of Folin-ciocalteu reagent which is made by 10 times dilution. Then it was incubated at room temperature for 3 minutes. The next step was, the addition of 10 ml of 20% sodium carbonate to the mixture and left for incubation at room temperature for an hour. The absorbance was measured against a blank solution at765 nm with a Shimadzu UV-vis-2600 Spectrophotometer. The blank solution content all the reagent mixture without the extract or standard sample. For the assessment of total phenolic contents, Gallic acid standard curve was used and the result were expressed as mg of gallic acid equivalent per gram of dried weight.

3.5.2 Determination of Total Flavonoid Content (TFC) assay

Total Flavonoid Content (TFC) was determined using the aluminum chloride colorimetric method. For this, Stock solution (1mg/ml) was prepared standard solutions (0.025, 0.050, 0.075 and 0.100 mg/ml) was made by dissolving Quercetin in 80% ethanol to plot a standard curve. Aliquots of 0.5 ml of diluted extract or standard solution was mixed with 1.5 ml of 95% ethanol, 0.1 ml of 10% aluminum chloride, 0.1 ml of 1mol/L Potassium acetate and 2.8 mL distilled water in the cuvette. The mixer was kept for 30 minutes at room temperature . Then absorbance

was measured at the wavelength of 415nm. For the measurement of blank, 10% aluminum chloride was substituted with distilled water of same amount. Thus TFC was calculated and it was expressed as milligrams querectin equivalents (QE) per gram of extract (mg QE/g) .

3.5.3 Determination of antioxidant capacity:

Antioxidant capacity was determined by following DPPH with slight modification stock solution (1mg/ml) of extract diluted in the concentration of 0.10, 0.20, 0.30,0.40 low 0.60 and 0.80 mg/ ml in menthol. Methanolic DPPH solution was prepared by adding 6mg of DPPH in 100ml Methanol. 2ml DPPH Solution was added to 1 ml of each extract solution of different concentrations and there it was kit for 30 minutes at the observance was measured at the wavelength of 517 nm. For The preparation off control, 1ml of methanol was mixed 2 ml of the solution. Antioxidant capacity based on the DPPH the free radical scavenging ability was determined by using the following calculation

$$\%\ \text{inhibition} = \frac{1 - \text{absorbance of Sample}}{\text{Absorbance of control}} \times 100\%$$

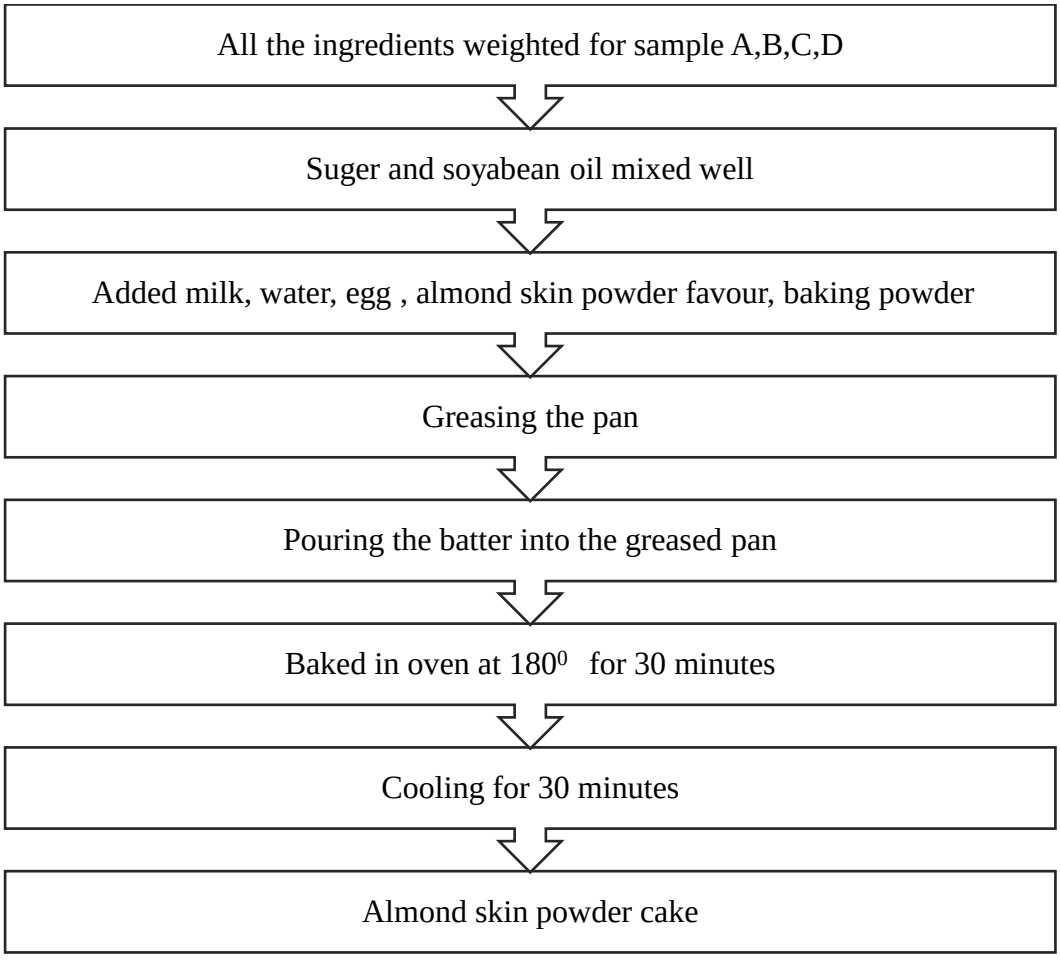
3.6 Preparation of almond skin powder cake

Sugar and soybean oil were taken in a bowl and mixed for four minutes. Whole egg was added to the bowl and mixed for two minutes. Then flour, baking powder, vanilla, water, and dry milk powder, almond skin powder gradually added into a bowl and mixed for four minutes. Water was poured into a bowl and then mixed for one minute. Then the cake batter was poured into a cake pan and baked at 180^0C temperature for 30 minutes. The cake was allowed to cool for 30 minutes. And then removed from the pans. The cake samples prepared with 0% (control), 20%, 25%, and 30% replacement of flour with almond skin powder.

Table 3.1 : Formulation of supplemented almond skin powder cake

Ingredients (g)	Sample A	Sample B	Sample C	Sample D
Almond skin powder	0	20	25	30
All -purpose flour	100	80	75	70
Sugar	72	72	72	72
Oil	57	57	57	57
Milk powder	2	2	2	2
Baking powder	2	2	2	2
Water	30	30	30	30
Vanilla	0.5	0.5	0.5	0.5

Figure 3.2: Shows the development of supplemented almond skin powder cake



3.7 Proximate analysis of supplemented cake

3.7.1 Determination of moisture content

Moisture determination is widely used method in processing and testing of food. Moisture content has great economic importance to the processor and consumer. Moisture content was determined by using the standard procedure of the Association of Official Analytical Chemists (AOAC, 2005). The amount of dry matter in food is inversely related to the amount of moisture it contains. At first, the empty crucible was taken and weighted accurately. Then about 5 gram of sample was weighted in crucible. The crucible was placed in electric oven and the temperature at $(105 \pm 1)^0\ c$ for 5 hours. The crucible was removed from the oven and cooled in desiccators. After cooling the crucible was weighted.

Moisture content can be calculated as follows:

$\%\ Moisture = \frac{W_1 - W_2}{W_1 - W} \times 100$

Where W= mass (weight) in gram of empty crucible

W_1 = mass (weight) in gram of the crucible with sample before drying .

W_2 = mass (weight) in gram of the crucible with sample after drying .

3.7.2 Determination of protein content

The kjeldahl method is used to determine the nitrogen content in organic and inorganic samples. Kjeldahl method of nitrogen analysis is the worldwide standard for calculating the protein content in a wide variety of materials like human and animal food, fertilizer, waste water and fossil fuels. The digestion step is done to break down the intricate Structure and chemical bond feed substance to simple ionic structure. As a result protein and other forms of Nitrogen are broken down and converted to ammonia. For that, firstly 1 gram of sample was weighted accurately. then 5 gram of digestion mixture and 20 ml of con. H_2SO_4 was added. The digestion flask was placed on kjeldahl digestion set. Then the heat was increased gradually and digested up to clear residue. The flask was removed and cooled it. Distillation is the step which involves the separation of ammonia-nitrogen from the

digestate. This is done by raising the PH with NaOH which changes the ammonia ion (NH_4^+) to ammonia (NH_3) . Nitrogen is separated by distilling the ammonia and collecting the distillate in a suitable trapping medium. Ammonia is collected by absorption into a solution of 2% of boric acid and it is bound to the boric acid in the form of ammonia borate. Determination of the amount of Nitrogen on the condensate flask can be measured by several methods. Among them, titration of ammonia with standard solution of N/10 HCl in the presence of mixed indicator is the most common method.

Protein content can be calculated as follows:

$$\%\ crude\ protin = \frac{A \times B \times 0.014}{W} \times 6.25 \times 100$$

A= volume of standard N/10 HCl solution

B= Normality of standard HCl solution

W= weight of the sample

6.25= Protein factor

3.7.3 Determination of fat

Fat is determined by dissolving food samples into organic samples such as (chloroform : methanol) separating the filtrate by filtration. The filtrate was placed into the separating funnels and then the separated mixture was dried to measure the extract. (AOAC, 2005) method using a soxhlet apparatus were used to determine the fat content of the samples. The dried sample was taken and it was transferred to a thimble. The top of the thimble was plugged with a wood of fat free cotton. The thimble was dropped into the fat extraction tube which was attached to a soxhlet apparatus. Approximately , 75ml or more of anhydrous petroleum ether was added through the sample in the tube into the flask . the top of the fat extraction tube was connected to the condenser. The sample was extracted about 16 hours or longer period on a water bath at 70-80^{0}C temperature. At the end of the extraction period, the thimble was removed and distilled of the petroleum ether. The ether was poured off when the tube was about to fill up and when it was reached a small volume . It was poured into a small, dry, previously weighted beaker through a small funnel containing plug cotton. The flower was rinsed and filtered properly by using ether. the Ether was evaporated on a steam bath at low heat, then dried at 100^{0}C for

1 hour , cooled and weighted. The difference in the weights indicates the ether soluble materials present in the sample.

Fat content can be calculated as follows:

The percent of crude fat was expressed as follows:

$$\%\ crude\ fat = \frac{weight\ of\ petrorum\ ether\ soluble\ material}{weight\ of\ sample\ taken} \times 100$$

3.7.4 Determination of crude fiber

Crude fiber is water soluble fraction of carbohydrate consists mainly of cellulose, hemicellulose and lignin.. The crude fiber was determined according to the AOAC method (2005). The weight of crucible was measured and 1 gram sample was taken into the crucible. Then the filter crucible was set with the fiber analyser and boiling of each sample was done by using 150 ml H_2SO_4 (1.25%) for 30 minutes. After waiting few minutes for cooling, each sample was washed by using 30 ml hot distilled water for three times. Again boiling was done for each sample by using 150 ml sodium hydroxide(1.25%) for 30 minute. Similarly, waiting few minutes for cooling each sample washed by using 30ml hot distilled water for 3 times. The sample was washed with 30ml normal distilled water for one time and then by 25ml acetone in condenser chamber for three times. The sample was kept at hot air oven for 1 hour at 105^0C. The crucible was cooled in desiccator and taken the sample weight. The sample was kept in muffle furnace at 550 ^{0}C for 3 hours. The crucible was kept at desiccator for 30 minutes and taken the final weight.

content can be calculated as follows:

$$\text{Fiber \%} = \frac{\text{wieght residue with crucible(g)} - \text{weight of ash with crucible}}{\text{weight of sample taken}} \times 100$$

3.7.5 Determination of Ash

The ash fraction of sample contains all the mineral elements. This method performs oxidization of all organic matter by incineration and determines the weight of remaining ash. The crucible was weighted properly. Then about 5 gram dried sample ignited in the crucible with the help of a suitable burner for about an hour. The ignition was completed by keeping in a muffle furnace at 550-600 C for about three

hours until gray color ash obtained. Then the crucible was removed through the furnace and cooled in a desiccator. . After cooling the crucible was weighted.

Ash content can be calculated as follows:

$$\% \text{ Total Ash } = \frac{W_1 - W}{W_2 - W} \times 100$$

Where, W= mass (weight) in gram of empty crucible

W_1 = mass (weight) in gram of the crucible with ash .

W_2 = mass (weight) in gram of the crucible with sample.

3.8 Sensory evaluation

Prepared cake formulas were subjected to sensory evaluation by 10 trained panelists in the CVASU premises, where the panelists were both teacher band students of CVASU. They assessed four cake samples for the attribute of color, odor, softness, crumb, texture and overall acceptability. The samples were encoded with sample A, sample B, sample C, sampleD. Water was given to each panelists in between samples. Sensory evaluation was done by hedonic scale. Each panelists scored samples independently and recorded the scores in the sheet which was arranged such that that : Dislike very much=1, Dislike moderately= 2, Dislike slightly= 3, Neither like or dislike= 4, Like slightly= 5, like moderately= 6, Like very much=7

(Ostermann-Porcel et al., 2020; Akosua et al., 2015).

3.9 Statistical analysis

Data (proximate composition, bioactive compounds and sensory evaluation) were determined and the recorded data stored in the MS Excel 2013 and exported to Statistical Package for Social Sciences (SPSS version 20.2). All samples were done in three replicates. Descriptive statistics (mean, standard deviation) were done for proximate composition, bioactive compounds and sensory evaluation for all cake samples. Proximate composition and sensory evaluation data almond skin powder cake sample were analyzed by using one – way ANOVA procedures to find a significant level of variation at 95 % confidence level. Level of confidence were shown at $P < 0.05$

Chapter 4: Results

4.1 Bioactive compounds of almond skin powder

4.1.1Total Phenolic Content (TPC)

The finding for Total Phenolic Content (TPC) of almond skin powder at different wave lengths was presented in the table 4.3. From the table the mean value of Total Phenolic Content (TPC) was found to be (1.39± 0.003) mg/100gm. (This value showed ME± SD of data. ME= Mean, SD=Standard deviation)

Table 4.3 Total Phenolic Content (TPC)

SL No	Con (mg/100)	WL 415.0
1	9.172	1.392
2	9.182	1.393
3	9.180	1.393

4.1.2 Total Flavonoid Content (TFC)

The finding for Total Flavonoid Content (TFC) of almond skin powder at different wave lengths was presented in the table 4.3. From the table the mean value of Total Flavonoid Content (TFC) was found to be (0.27± 0.05) mg/100gm.. (This value showed ME± SD of data. ME= Mean, SD=Standard deviation)

Table 4.2 Total Flavonoid Content (TFC)

SL No	Con (mg/100)	WL 415.0
1	70.954	0.271
2	70.831	0.270
3	70.846	0.270

4.1.3 Antioxidant capacity

The antioxidant capacity of almond skin powder at different wavelengths is shown in table 4.2. From the table the mean value of antioxidant capacity was found to be (0.04 ± 0.01) mg/100gm. (This value showed ME $\pm$ SD of data. ME= Mean, SD=Standard deviation)

Table 4.1 Antioxidant capacity

SL No	Con (mg/100)	WL 517.0
1	2.921	0.053
2	2.925	0.053
3	2.925	0.053

4.2 Fiber content of almond skin powder

Data represented in table showed the fiber content of almond skin powder. From the table the mean value of fiber content of almond skin powder was found to be (14.6± 0.04) %.(This value showed ME± SD of data. ME= Mean, SD=Standard deviation)

Table 4.4 Fiber content of almond skin powder

Sample name	Fiber content
Almond skin powder	14.6 ± 0.04

4.3 Proximate analysis of cake

Data represented in table 4.1 showed the nutritional value of almond skin powder cake samples. Sample A had the highest moisture content $(27.6 \pm 0.05)\%$ whereas sample B had the lowest $(25.2 \pm 0.04)\%$. The highest value of crude fiber content was found in sample D $(6.3 \pm 0.03)\%$ and the lowest sample A (0.00%). Ash content was higher in sample C $(0.7 \pm 0.04)\%$ and the lowest found in Sample in sample A(0.4± o.04)%. Fat content was higher in sample A $(19.5 \pm 0.04)\%$ comparatively than other sample in B,C and D. Protein content was higher in sample D(7.7 ± 0.04) % than other formulations. Carbohydrate content was higher in sample A $(44.8 \pm 0.04)\%$ and other lower in sample D $(40.9 \pm 0.04)\%$.

Table 4.5 Proximate composition of cake

Parameters	Sample A	Sample B	Sample C	Sample D	P-value
Moisture (%)	27.6±0.05^{a}	25.2±0.04^{d}	25.7±0.48^{c}	25.9±0.04^{b}	0.00
Fiber (%)	0^{d}	3.3±0.03^{c}	4.6±0.03^{b}	6.3±0.03^{a}	0.00
Ash (%)	0.4±0.04^{b}	0.7±0.03^{a}	0.7±0.04^{a}	0.7±0.04^{a}	0.00
Fat (%)	19.5±0.04^{a}	18.9±0.04^{b}	18.6±0.02^{c}	18.2±0.03^{d}	0.00
Protein (%)	7.5±0.02^{c}	7.6±0.03^{b}	7.4±0.04^{d}	7.7±0.04^{a}	0.004
CHO(%)	44.8±0.04^{a} 4	4.1±0.04^{b}	42.2±0.03^{c}	40.9±0.04^{d}	0.00

Legends: All values showed ME ± SD of data. ME = Mean. SD = Standard Deviation. The presence of different superscript along a row indicates a significant difference and the same superscripts are not significantly different at $P < 0.05$.

A, B, C and D represented formulation 1 (0% almond skin powder), formulation 2 (20%almond skin powder), formula 3 (25% almond skin powder) and formula 4 (30% almond skin powder) respectively.

4.4 Energy content of almond skin powder cake

Figure 4.1 Shows the energy content of four formulations of cakes. Energy content of sample A was calculated in the highest amount (390.4 kcal/100 gm.) and lowest in sample D (358.2 kcal/100gm).

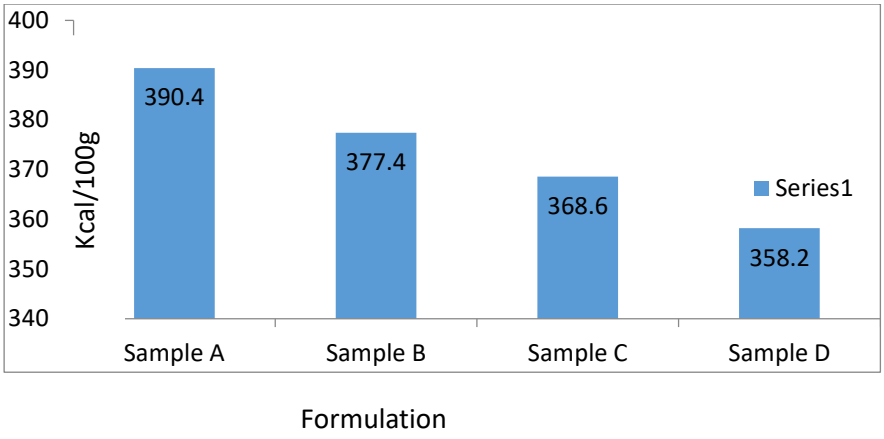


Figure 4.1: Comparison of energy content of cakes

4.5 Sensory evaluation

In Table 4.6 highest (ME ± SD) score for Softness , Sweetness , crumbness was recorded (5.8 ± 0.95),(5.7 ± 0.86) ,(5.9 ± 0.76) respectively in the case of Sample D. Sample C scored highest in odor (5.3 ± 1.73) and sample D scored highest in color (5.6 ± 0.99). On the other hand, lowest score for all attributes was recorded in Sample A. Sample D had the highest acceptance rate (5.8 ± 0.86). However, Sample A scored the least acceptance (4.7 ± 1.003) compared to other samples.

Table 4.6: Hedonic rating test for sensory evaluation of cake formulas

Parameters	Sample A	Sample B	Sample C	Sample D	P-value
Color	5.1 ± 1.13^{d}	5.4 ± 1.33^{c}	5.6 ± 0.93^{b}	5.9 ± 0.05^{a}	0.106
Odor	5.2 ± 1.78^{b}	5.3 ± 0.05^{a}	5.3 ± 1.73^{a}	5.2 ± 0.04^{b}	0.971
Softness	4.9 ± 1.27^{d}	5.6 ± 0.48^{c}	5.7 ± 1.12^{b}	5.8 ± 0.95^{a}	0.117
Sweetness	5.0 ± 1.21^{d}	5.3 ± 1.13^{c}	5.4 ± 1.29^{b}	5.7 ± 0.86^{a}	0.217
Crumb	4.4 ± 0.78^{c}	5.6 ± 1.23^{b}	5.6 ± 0.001^{b}	5.9 ± 0.76^{a}	0.001
Appearance	5.3 ± 1.05^{d}	5.6 ± 0.04^{c}	5.8 ± 0.06^{b}	6.2 ± 0.83^{a}	0.117
Overall acceptability	4.7 ± 1.003^{d}	5.2 ± 1.17^{c}	5.4 ± 0.93^{b}	5.8 ± 0.86^{a}	0.002

Legends: All values showed in ME ± SD of data. ME = Mean, SD = Standard Deviation. The presence of different superscript along a row indicates a significant difference and the same superscripts are not significantly different at $P<0.05$

Chapter 5: Discussion

5.1 Bioactive compounds of almond skin powder

DPPH was widely used substrate to determine the antioxidant capacity of biological and chemical substances. From the result, the antioxidant capacity was found (0.04 ± 0.01). According to the study (Lunec et al.,1986) the antioxidant capacity showed (4.21± 0.24)mg that are so higher than the present study. Free radicals that are generated in body due to its metabolic function may lead to oxidative stress; when it produced excessively or not adequately neutralized may turn cause tissue damage (Yoshikawa et al., 1997).

The methods of determination of almond polyphenols were mainly by High Performance Liquid Chromatography (HPLC) coupled to UV/VIS detection and mass spectrometry. The recorded data from the result was found ((1.39± 0.003) mg/100gm that is lower than the reported data range from (6.11 to 7.0) mg/100gm (Abe LT et al.,2010). Almonds have a diverse polyphenol profile including the contribution to both its food quality and health promoting actions (Mandalari et al., 2011).

Almond skin is rich sources of water soluble bioavailable flavonoids and other polyphenols. At least 25 different flavonoids have been identified in almonds, almond skin or blanched almond water (Harnly et al., 2006). The flavonoid content of the almond skin powder was recorded ((0.27± 0.05) mg/100gm which is almost similar to the study (Hugchey C et al., 2008). The high phenolic content in almond skin may contribute to the antioxidant and nutritive value associated with almond consumption (Milbury et al., 2006)

5.2 Nutritional composition of almond skin powder cake

Moisture content plays a significant role in maintaining proper conditions for the preservation, stability, packaging and shipping of food products (Maur et al., 2017). In this study, moisture content was found in cake is (27.6± 0.05)% which was quite higher compare to other study (20.1± 0.02)% (Summo et al.,2018). Highest moisture content was recorded in sample A (27.6± 0.05)%. Sample B (25.6± 0.04)%, sample C (25.7± 0.48) and D (25.9 ± 0.04) had lower moisture content than sample A. Sample A contain no almond skin powder. The variation in moisture content may be

due to different drying process in the laboratory, regional production or weather condition of production area. Moisture content is a vital factor the growth of microorganisms which may be harmful for the quality of products.

The protein content was quantified to be (7.7±0.04) % . The protein content was found in this research was almost similar to the previous study which was about (5.6±0.03) % (Rodrigues et al., 2016). Protein content became high with increasing substitution of almond skin powder. High protein content was observed in sample D (30% almond skin powder + 70 % flour) .Sample D contained (7.7±0.04) % protein while in control (7.5±0.02) %.).The reason of this variation may be due to the microhabitat variation in study area or different spices of almond. It is desirable because it increases the protein content in diet.

The fat content reported in the study from the range (10.3± 0.9 to 12.4±0.04) % which is lower from the value (caponio et al., 2005. The highest fat content was found in sample A (19.5±0.04) % than sample B (18.9±0.04) %, sample C (18.6±0.02) % and sample D (18.2±0.03) %. Different processing methods in laboratory and species may contribute to this variation. It indicates that fat values decreases with increase of supplementation level in cakes.). Fat is a concentrated source of energy which is used when body requires energy supply. Generally fat helps to protect the internal organs like kidney, lungs and subcutaneous tissues of skin (Mohamed et al., 2017).

Ash content found quite well in sample B (0.07± 0.03), sample C (0.07 ± 0.04), sample D (0.07± 0.04) .The ash content is slightly close to the other study (Ahmed et al., 2012). It indicates that the cake made with almond skin powder is a better source of minerals. Fiber content of cake samples increased with fortification level while the control sample A had the least value of fiber content (0%) due to no addition of peel powder in cake. Highest fiber content was recorded in sample D (6.3±0.03)% as this cake formula was fortified with highest content of almond skin powder. Fiber content found to the range which was closely to that obtained by some researchers (5.8±0.03)% (Rodrigues et al., 2016). It can be an indication of containing high fiber content of almond peel powder. Increasing fiber content is a desirable fact as it is a vital element to control cholesterol level, many digestive problems and decrease the risk of cancer (Kanawal et al., 2015).

The carbohydrate content ranged from (44.8 ± 0.04) % in sample A to (40.9± 0.04) % in sample D. There was no significant difference in sample B and sample C. The RDA (Recommended Daily Allowances) of carbohydrate for adults and children aged one year or older is 130g/ day (Slavin et al., 2018). The energy content was found (390.4 kcal/100 gm.) which can contribute to meet the nutritional requirements.

5.3 Sensory evaluation of cake samples

The utilization of almond skin powder with different proportions to produce cake formulas was sensory evaluated and then compared to control cake which contained 0% almond peel powder (sample A). Supplementation of cake with different ratios of almond skin powder significantly affected the sensory attributes of produced cakes.

In case of appearances, the control cake sample A had the lowest score (5.3 ±1.05) while the cake supplemented with 30% almond skin powder had the highest score (6.2 ±0.83) which is higher than the previous study (Rodrigues et al., 2016). Sensory evaluation findings indicated the Increase of supplementation ratio up to 30% of almond skin powder, prominently increase the acceptance of cake and the appearance became gradually better.

Odor of supplemented cake scored quite similar to control and no significant difference was found. Regarding the color, significant differences were observed among all supplemented cake formulas. The cake sample supplemented with 30% almond skin powder (sample D) recorded the highest score (5.9 ± 0.05) while the control sample A recorded the lowest score (5.1 ± 1.13). Other study related to this attribute scored the range 4-5% in average which is almost similar to this study (Summo et al.,2018).

In case of softness, sweetness and crumbness, sample D (5.8 ± 0.95), (5.7 ± 0.86), (5.9 ± 0.76) respectively supplemented with 30% almond skin powder recorded the highest score than other samples whereas the control recorded the lowest score (4.9 ± 1.27), (5.0 ± 1.21), (4.4 ± 0.78) respectively.

The highest score for overall acceptability was recorded by sample D (5.8 ± 0.86) which contained sample 30 % almond skin powder and the sample supplemented with

25 % and 30% were sample B (5.2 ± 1.17) and sample C (5.4 ± 1.03) respectively. The acceptability of previous study recorded (4.2 ± 1.17) which is lower than the present study (Rodrigues et al., 2016). In contrast, sample A was the lowest score (4.7 ± 1.03). Findings indicated that the an increase of almond skin powder in the formulation improved the overall acceptability of cakes for panelists.

Chapter 6 : Conclusion

This study concludes that the supplementation of cake formulas with almond skin powder in an exceptional value added product and it is also indicated to have acceptability with respect to all parameters like appearance, aroma, taste, texture. The nutrient profile of this product was also significant from the point of view. It is also important to note that this product is specially include fiber, antioxidants, protein, fat, energy value which make it an healthy choice for fulfill the nutritional demand of country. In terms of acceptability, panelists gave performance to the sample supplemented with almond skin powder which is more acceptable than the control.

Chapter 7: Recommendations and Future Perspectives

The global consumption of nuts and nut derived products has greatly increased during recent decades, due to a number of distinct factors. Nut constitutes a healthy part of human diet, mainly presence of bioactive compounds, which play an essential role in human health, also the presence of its fiber and mineral contents. Present study is conducted to investigate the formulations and quality (chemical, nutritional, bioactive and sensory) evaluation of almond skin powder cake. In country, almonds are widely available and its skin can be used to develop different types of healthy snack foods like cake, chips, biscuits and other bakery items for malnourished children and people by considering the nutritional factors. In this case, the almond skin can be collected from industrial by products. Then it will be more cost effective. Based on the present investigation, the following suggestions and prospects are made for further research work;

- Mineral parameters of cakes should be analyzed including magnesium, iron.
- Fat soluble vitamins like D, E, K should be analyzed.
- The physical characteristics test and the shelf life of cakes should be analyzed.
- The findings will be helpful from a therapeutic point of view as they have medicinal value.
- The formulas may be modified with a different flavor for better taste.
- Modern packaging and storage conditions would be developed to better almond skin powder products.
- Awareness should be created about the health benefits of almond nuts and its possibility of being scope for the food industry.

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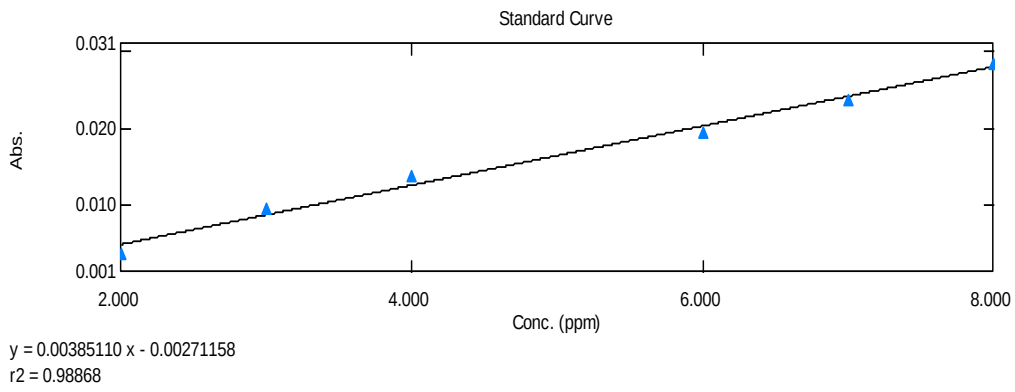
Appendices

Appendix A: Total Flavonoid Content (TFC)

Standard Table of Qwercetin:

	Sample ID	Type	Conc. (ppm)	WL 415.0	Wt factor
	Comments				
1	Std_1	Standard	2.000	0.004	1.000
	Dilution Factor				
2	Std_2	Standard	3.000	0.010	1.000
	Dilution Factor				
3	Std_3	Standard	4.000	0.014	1.000
	Dilution Factor				
4	Std_4	Standard	6.000	0.020	1.000
	Dilution Factor				
5	Std_5	Standard	7.000	0.024	1.000
	Dilution Factor				
6	Std_6	Standard	8.000	0 .029	1.000
	Dilution Factor				

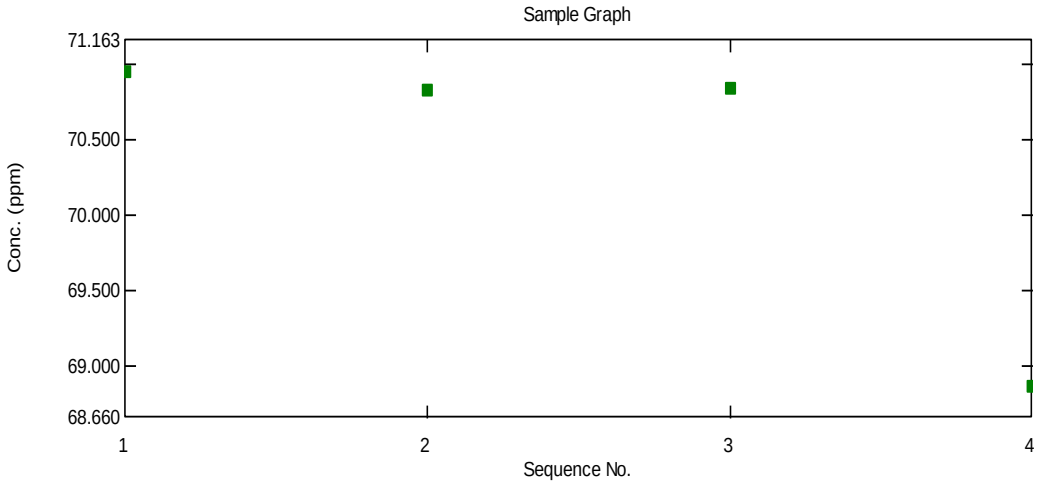
Standard Curve:



Sample Table

	Sample ID	Type	Conc(mg/100g)	WL415.0
	Comments			
1	Sample1.1	Unknown	70.954	0.271
2	Sample1.2	Unknown	70.831	0.270
3	Sample1.3	Unknown	70.846	0.270
4	Sample1.4	Unknown	68.868	0.263

Sample Graph

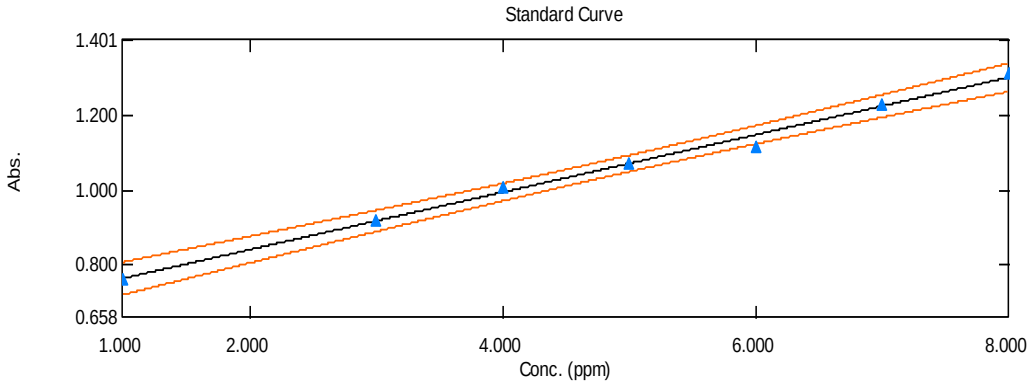


Appendix B: Total Phenolic Content (TPC)

Standard table of Gallic Acid:

	Sample ID	Type	Conc. (ppm)	WL760.0	Wt factor
1	STD1	Standard	1.000	0.763	1.000
2	STD2	Standard	2.000	0.780	1.000
3	STD3	Standard	3.000	0.920	1.000
4	STD4	Standard	4.000	1.007	1.000
5	STD5	Standard	5.000	1.074	1.000
6	STD6	Standard	6.000	1.115	1.000
7	STD7	Standard	7.000	.230	1.000
8	STD8	Standard	8.000	1.314	1.000

Standard Curve:

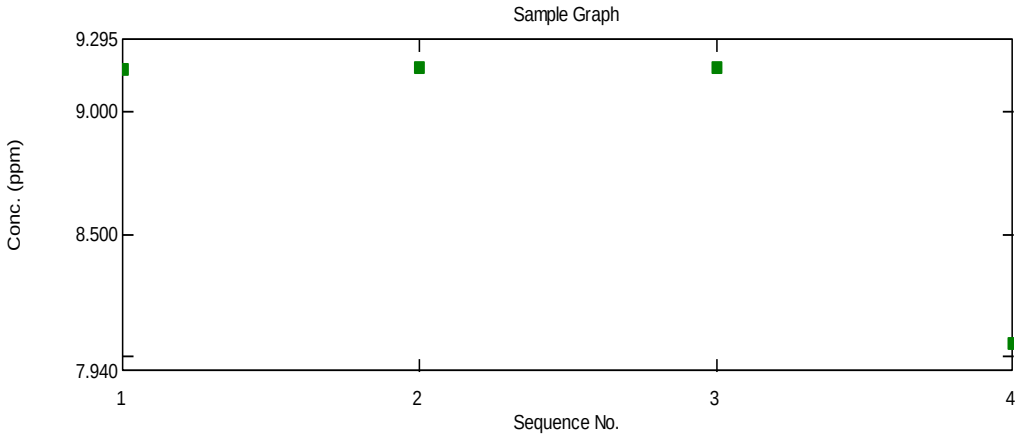


y = 0.0768527 x + 0.687090
r2 = 0.99301

Sample table

	Sample ID	Type	Conc(mg/100g)	WL760.0
	Comments			
1	Sample1.1	Unknown	9.172	1.392
2	Sample1.2	Unknown	9.182	1.393
3	Sample1.3	Unknown	9.180	1.393
4	Sample1.4	Unknown	8.053	1.306

Sample Graph

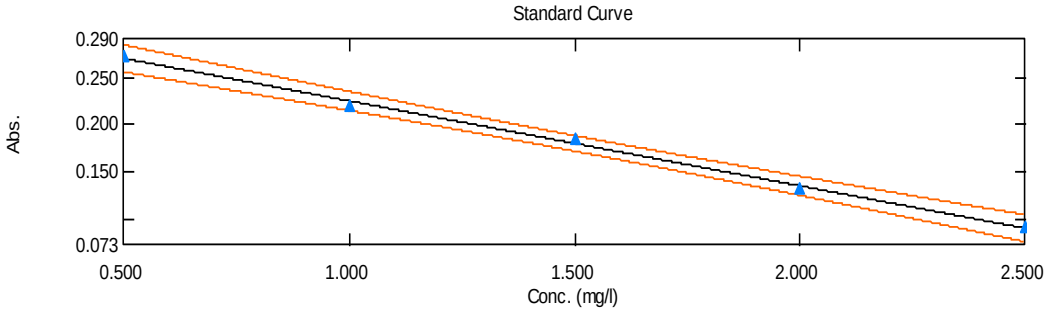


Appendix C : Antioxidant capacity

Standard Table of Trolox:

	Sample ID	Type	Conc(ppm)	WL517.0
1	std1	Standard	0.500	0.272
2	std2	Standard	1.000	0.221
3	std4	Standard	1.500	0.185
4	std5	Standard	2.000	0.133
5	std6	Standard	2.500	0.092

Standard Curve:

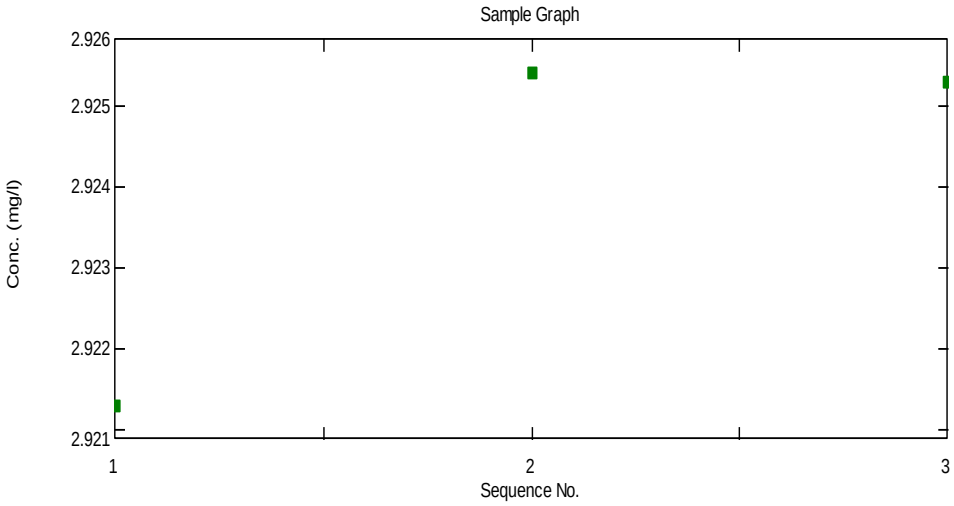


y = - 0.0894539 x + 0.314536
r2 = 0.99735

Sample Table:

	Sample ID	Type	Conc(mg/100g)	WL517.0
	Comments			
1	Sample1.1	Unknown	2.921	0.053
2	Sample1.2	Unknown	2.925	0.053
3	Sample1.3	Unknown	2.925	0.053

Sample Graph



Appendix D : Acceptance test

Acceptance test

Date: **Sample code**:

Gender: Male / female **Age**:

Instruction:

You are given four samples. Please start your evaluation from left to right. Evaluate each attribute by circling the appropriate scale which indicates your degree of liking. Rinse your mouth with plain water before tasting each sample. (1 – Dislike Extremely, 2 – Dislike moderately, 3 – Dislike slightly, 4 – Neither like nor dislike, 5- Like slightly, 6 – Like moderately, 7 – Like extremely)

Color

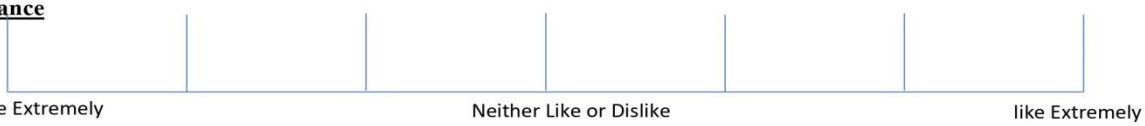
Odor

Softness

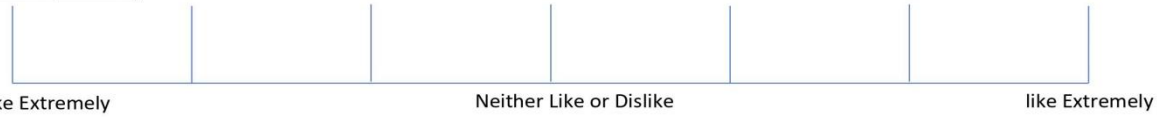
Sweetness

Crumbness

Appearance



Overall acceptability



Comments (If any)

Appendix E : Photo gallery

Appendix E1: Pictorial presentation of almond skin powder

Raw almonds

Soaked almond

Peeling the skin

Drying in cabinet dryer

Almond skin powder

Appendix E2: Pictorial presentation of almond skin powder

1. Weighted ingredients

2. Adding all ingredients

3. Mixing

4. Greasing the pan

5. Cake batter

6. Baking

7. Cooling the cake

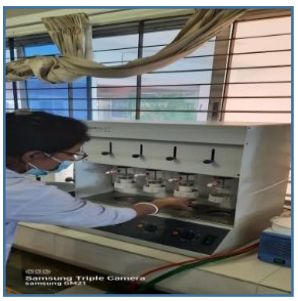
8. Storing

Sensory evaluation

Appendix E3 : Pictorial presentation of laboratory work

Drying in cabinet dryer

Moisture determination



Fat determination

Fiber determination

Protein determination

Sampling for spectrometric determination

Working in UV spectrophotometer

Brief Biography

Shimu Ghosh passed the Secondary School Certificate Examination in 2011 and then Higher Secondary Certificate Examination in 2013. She obtained her BSc (Honors) in Food Science and Technology from the Faculty of Food Science and Technology at Chattogram Veterinary and Animal Sciences University, Chattogram, Bangladesh. Now, she is a candidate for the degree of Master of Science in Applied Human Nutrition and Dietetics under the Department of Applied Food Science and Nutrition, Faculty of Food Science and Technology, Chattogram Veterinary and Animal Sciences University (CVASU). She has an immense interest in exploration on clinical nutrition and dietetics to improve the health of people through proper guidance and suggestions with a vision of developing the overall nutritional status of Bangladesh.