

Quality Characteristic Assessment of *Nelumbo nucifera* Developed Custard Powder

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Abstract

Lotus seeds are a vital component of ancient ayurvedic medicine and are used to treat a wide range of illnesses, and skin problems with abundant nutrients. The plant-based milk and milk products are preferred by people who are vegan, lactose intolerant, and allergic to cow's milk. Consumers use milk powders to replace fresh milk and as a component in a variety of processed food products. This study aims to standardize and spray dry lotus seed milk, formulate it into a custard powder mix, and subjected to evaluate the physical, proximate, functional, microbial, and sensory analysis. The lotus seeds (fresh and dry) were cleaned, ground to extract milk, selected seed was spray-dried, formulated into custard powder mix, and analyzed. The results showed that formulated powder has higher protein ($33.84\text{g}\pm 1.0$), with low fat ($1.402\text{g}\pm 0.1$) and carbohydrate ($19.91\text{g}\pm 1.01$), calcium ($394.7\text{g}\pm 1.0$), potassium ($70.5\text{mg}\pm 1.04$), magnesium ($94\text{mg}\pm 1.0$), exhibits antioxidant activity of 80.45%. The prepared custard exhibits the textural property of springiness (0.912 ± 0.3), cohesiveness (0.80 ± 0.1), and chewiness (60.4 ± 0.3). The sensory attributes resulted in overall good acceptability with 30 days of stable shelf life. Thus, the developed custard powder mix was considered to be nutritionally dense with good textural properties and is considered as a value-enhanced convenience food.

Key words: Lotus seeds, Plant-based milk, Antioxidant, Spray drying, Instant powder mix

The global market for functional and specialty beverages that use plant-based or non-dairy milk alternatives is expanding at a rapid rate. Cow milk allergy, lactose intolerance, calorie-rich foods and the incidence of hypercholesterolemia, as well as a growing demand for vegan diets, have all led consumers to choose cow milk substitutes in modern times (Sethi *et al.*, 2016). Plant-based milk substitutes and products are a great option despite the extra sugar and low total protein intake since they include phenolic compounds, unsaturated fatty acids, antioxidant activity, and bioactive components including phytosterols and isoflavones (Aydaret *et al.*, 2020). The market for plant-based milk alternatives has also been encouraged by active vegan activists, with food producers choosing alternate sources of plant-based ingredients. Certain legumes and oilseeds have been widely used in the creation of non-dairy, healthy, inexpensive, and nutritious plant-based milk replacements in this regard. However, in recent years, researchers have focused on investigating the utilization of indigenous and locally available use of cereals, oilseeds, and nuts for new culinary applications based on their functional qualities, which show the physical characteristics of food components and their interactions (Cruz, *et al.*, 2007).

Spray drying is a process that is frequently used in the food industry to produce a variety of foods and ingredients, including milk powder, whey/casein powder, cheese powder,

ice cream mix powder, egg powder, cereal powder, dried infant foods, instant coffee/tea powder, juice powder, instant soup powder, fish powder, as well as flavor and color ingredients. The milk powder is usually produced using spray drying technology, which removes water and creates particles (Shishir and Chen, 2017). The lotus (*Nelumbo nucifera*), an aquatic perennial that has been farmed for more than 2,000 years, is extensively dispersed in Asia, Australia, and North America. It is quite energizing that practically all components of the lotus have been used as food and vegetables, especially the seeds as they are rich in nutritional composition and functionally bioactive compounds (Zhu *et al.*, 2016). Lotus seeds are a vital component of ancient Ayurvedic medicine and are used to treat a wide range of illnesses, including tissue inflammation, cancer, diuretics, skin problems, and poison antidote (Dinkar and Mishra, 2020). Thus, the production of plant-based milk products is experiencing high demand and the need to improve food nutrient quality is increasing day by day.

One of the food categories that cannot be substituted in the food market is plant-based milk alternatives as they are necessary components of many vegan food items like plant-based ice cream, yogurt, cheese, kefir, tofu, ghee, cream, condensed milk, milk powder, custard, puddings, mousse, bars, and butter. Additionally, people who are lactose intolerant or allergic to cow's milk require plant-based milk replacements

(Aydar *et al.*, 2015). Value addition can enhance the food's nutritious content and can be utilized to develop products to boost the nutritional value of foods rich in protein, energy, minerals, vitamins, fiber, etc. Thus, the addition of this nutrient-rich seed in various other food products possesses many health benefits (Faiza *et al.* 2015). Custard powder is a fine-textured, dry food manufactured from maize starch that may or may not also contain egg yolk solids, vitamins, and minerals. Other ingredients, such as salt, flavoring, and coloring agents, may also be added. Custard powder is a carbohydrate-rich breakfast food that many people eat as a breakfast meal and also as a supplement for newborn feeding (Awoyale *et al.* 2016). Thus, the value addition of this particular food product is very much required to improve its nutritional quality. Therefore, the study was aimed to formulate the ready-to-cook custard powder from spray-dried lotus seed milk and the objective was to evaluate the physical, microbial, proximate, functional, and sensory analysis of Nelumbo nucifera custard powder mix.

MATERIALS AND METHODS

Extraction of lotus seed milk

The plant-based milk was formulated using fresh and dry lotus seeds that were procured from the local market of Chromepet. Fresh lotus seed skin was peeled, cut, and the green stalk was removed to avoid the bitter taste and color change, and the Dry lotus seeds were broken, and soaked for 12 hours, the skin of the seed was peeled and the green stalk was removed. The milk was extracted as mentioned by Ayder *et al.* 2020 with slight modifications. Both the Fresh Lotus Seed Milk (FLSM) and Dry Lotus Seed Milk (DLSM) were prepared by grinding the seeds and water with a proportion of 1:3 ratio and filtered with a 4-layered muslin cloth (porous size of 2mm) and pasteurized at 72°C for 15 seconds. Both the samples were subjected to physical and nutritional analysis and the best sample was selected and further processed.

Physical analysis of FLSM and DLSM

The moisture content, pH and total soluble solids were analyzed through methods of AOAC 2000.

Nutritional and antioxidant characterization of FLSM and DLSM

The proximate composition of the lotus seed milk was determined for ash content, carbohydrates, protein, fat was carried out by AOAC, 2000 method. The antioxidant activity was analysed by DPPH Assay by following (Blois 1958).

Spray drying dry lotus seed milk

The feed comprising of lotus seed milk was spray dried in lab-scale spray dryer (Chennai). The inlet temperature and outlet temperature were 80°C and an outlet temperature range of 160°C The Compressor pressure and feed rate were 1.2 bars and 200ml/hr, respectively (Shishir *et al.*, 2014).

Formulation of custard powder

The custard powder was prepared by blending the ingredients - spray-dried lotus seed milk powder, corn flour, sugar, and vanilla bean. The ingredients were blended at various proportions with sugar at a constant value and were sieved to avoid lumps and larger particles. The blended ingredients were stored and sealed in air-tight containers until analysis was carried out (Ali M *et al.*, 2019).

Physical analysis of DLSM custard powder mix

Bulk density

By weighing the mixture and calculating the associated volume, the bulk density of the custard powder was ascertained. 10ml graduated measuring cylinder was filled with approximately 5gm of material. The mass of the mixture was divided by the volume it took up in the cylinder to determine the bulk density (Goula *et al.*, 2004).

$$\text{Bulk density} = \frac{\text{weight of the powder}}{\text{volume of the powder}} \text{ g/ml}$$

Moisture content

The AOAC technique was used to conduct the moisture content analysis. One gram of sample was taken & dried in an oven at 70°C until constant weight & triplicated the analysis and calculated as (Shinde DD, 2017).

$$\text{Moisture content} = \frac{\text{weight of water loss}}{\text{weight of the powder sample}} \times 100$$

Water absorption capacity

Sample (1g) was weighted into conical graduate centrifuge tubes of known weights and mixed with 10ml of distilled water for one minute with a glass rod. The tubes were then centrifuged at 500 rpm for 30 min. the volume of free water was discarded and each tube together with its content was reweighed as water absorbed per gram of sample. The gain in mass was the volume of water absorbed by 1g of the test sample. Absorption capacity is expressed in grams of water absorbed per gram of sample (Onwuka, 2005).

Water absorption capacity

$$= \frac{\text{density of water} + \text{volume absorbed}}{\text{weight of the sample}}$$

Microbial quality

The total plate count (TPC) of the custard powder mix was estimated by the procedure laid down in the method of (Ahmed *et al.*, 2020) using nutrient agar (NA) media, Potato dextrose agar (PDA) was used to enumerate yeast and mould count as per the procedure laid down in the method of Awoyale *et al.* 2015.

Nutritional and antioxidant analysis of DLSM custard powder mix

The proximate composition of the lotus seed milk and the custard powder mix was determined for ash content, moisture content, carbohydrates, protein, fat, vitamin B6, vitamin C was carried out by (AOAC, 2000) method. Magnesium, and calcium by IS 5949 method. Phytochemical was analyzed by using the procedure of (Harborne, 1998). The antioxidant activity was analyzed by DPPH Assay (Blois, 1958).

Physical characterization of custard developed from DLSM custard powder mix

The formulated custard made from prepared custard powder was analyzed for physical properties such as texture property. The texture property such as hardness, cohesiveness, springiness, and chewiness of the custard was analyzed with the texture analyzer of TA XT plus model.

Sensory evaluation of custard developed from DLSM custard powder mix

The sensory characteristics of the custard prepared from the custard powder was determined in terms of colour, appearance, texture, flavour, and overall acceptance according to the sensory evaluation technique described by (Ranganna 1986).

shelf-life analysis of DLSM custard powder

According to (Krause *et al.*, 2008) with modifications, Storage studies were carried out for the prepared sample and quality parameters like microbial load and sensory evaluation were evaluated during storage on the interval of 0th, 1st, 15th and 30th day in air tight containers.

RESULTS AND DISCUSSION

Evaluation of physical properties of lotus seed milk (LSM) extracts

pH

The pH helps to improve shelf stability and influence the flavor of ready-to-serve beverage (Harihana *et al.*, 2016). At any medium pH is inversely proportional to the acidity of the beverage (Gaikwad *et al.*, 2012). The acidity and pH measurements in certain foods provide an indication of the degree of deterioration, confirmed by the acidity or alkalinity developed (Dantas *et al.*, 2014). It is an important parameter as

pH measures the degree of acidity or alkalinity of a product. The pH was determined using a pH meter. The pH of the DLSM was found to be 6.2±0.3 and FLSM shows 6.3±0.4. Kim *et al.*, 2012 also noted similar pH results of 6.75 in soy milk. Dhankar *et al.*, 2018 also exhibited a pH result of 6.92 in almond milk.

Total soluble solids

Solubility is the amount of solute dissolved in a solvent at a given temperature. Manufacturing of dairy ingredients can be enhanced by increasing the total solids (TS) content before spray drying (Bista *et al.*, 2020). The total solid content of DLSM was found to be 24%±0.1 and FLSM was found to be 20%±0.3 which is lower than DLSM (table1). According to Jyotika *et al.*, 2018 the total solid content of millet milk was found to be 9.167% and coconut milk was found to be 25% similar to the LSM results. The value of total solids for cow milk was recorded as 13.07% and a similar result was reported by Yoganandi *et al.*, 2015.

Table 1 Physical properties of the sample

| Sample | pH | Total solid content % | Water content % |
|------------------------------|---------|-----------------------|-----------------|
| Fresh lotus seed milk (FLSM) | 6.3±0.4 | 20%±0.3 | 85%±0.2 |
| Dry lotus seed milk (DLSM) | 6.2±0.3 | 24%±0.1 | 80%±0.1 |

Moisture content

Moisture content is the amount of water contained in food, expressed in units of a percent (%). Moisture content is one of the important characteristics that can affect the appearance, texture, and taste of a product (Blahovec, 2007). The moisture content of DLSM was found to be 80%±0.1 and FLSM to be 85%±0.2 as mentioned in (Table 1). Hutabarat and

Irene, 2021 reported that the moisture content of soy milk was 83.256%, almond milk was 76.29% and Almond-soy milk was 78.07%. Jyotika *et al.* 2018 found the moisture content of coconut milk was 75.10%.

Nutritional composition and antioxidant activity of FLSM and DLSM

Table 2 Nutritional composition of the sample

| Sample | Protein (g) | Fat (g) | Ash (g) | Carbohydrate (g) |
|------------------------------|-------------|-----------|-----------|------------------|
| Fresh lotus seed milk (FLSM) | 3.2±0.2 | 0.13±0.34 | 0.2±0.12 | 1.5±1 |
| Dry lotus seed milk (DLSM) | 11.09±0.95 | 0.2±0.1 | 0.79±0.01 | 0.82±1 |

Values represent Mean ± Standard deviation

A) Protein

Proteins are essential macronutrients that play a significant role in the body's mechanisms and functions. The protein content of the DLSM was found to be 11.09g±0.95 and the FLSM was found to be 3.2g±0.2. DLSM has higher protein activity than FLSM. According to Nair *et al.* 2020, the protein content of millet milk was found to be 8.5g. and the results obtained by Shrivastav *et al.*, 2022 found to be 6.75g. The soaking process led to an increase in the total protein of seeds (El-Adawy *et al.*, 2000). Hence it can be concluded that DLSM is a rich source of protein and also possess higher protein concentration than cow's milk.

B) Fat

The fat content of DLSM was found to be 0.2g±0.1 and FLSM shows 0.13g±0.34. A slight increase in the fat content of DLSM was observed. The results obtained by Vahini and Mary, 2021 from soy milk were found to be 2.29g, and from the results obtained by Nair *et al.* 2020 the fat content of millet milk was 0.74g. Furthermore, the low level of fat in LSM will lead to the reduction of cholesterol content, and the inclusion of heart-healthy unsaturated fatty acids, and is ideal for people suffering from obesity, hypercholesterolemia, and other heart diseases.

C) Ash

The ash is an inorganic deposit obtained after the removal of water and organic matter by heating which is carried out in the presence of oxidizing agents. The amount of ash

content present in the sample accounts for the total amount of minerals present in the food (Ahsan *et al.*, 2015). The ash content of DLSM has a concentration of 0.79g±0.1 and FLSM has 0.2g±0.12. In this analysis, the ash content of DLSM increased due to the soaking process of dry lotus seed (El-Adawy *et al.*, 2000). According to De *et al.*, 2022, the ash content of soy milk extract was found to be 0.84g and the results obtained by Vahini and Mary, 2021 in the sesame seed milk extract were found to be 0.71g. the above results indicate that DLSM has higher mineral composition than FLSM.

D) Carbohydrates

In this present study, the carbohydrate content of DLSM was found to be 0.82g±1.0 and FLSM was found to be 1.5g±1. However, the results obtained by Vahini and Mary, 2021 showed that the carbohydrate content present in the sesame seed milk extract was found to be 2.45g and the soymilk was found to be 3.21g. The total nutrient quality of the seed was affected by the soaking process which resulted in the reduction of carbohydrate content (El-Adawy *et al.* 2000).

Antioxidant activity of FLSM and DLSM

Table 3 Antioxidant activity of the samples

| Sample | OD Value | Scavenging % |
|------------------------------|----------|--------------|
| Fresh lotus seed milk (FLSM) | 0.600 | 30% |
| Dry lotus seed milk (DLSM) | 0.637 | 31.87% |

Reactive oxygen species (ROS) and reactive nitrogen species (RNS) in biological systems, such as superoxide, hydroxyl, and nitric oxide radicals, can damage DNA and cause lipid and protein oxidation in cells. Normally, the antioxidant system in the human body can scavenge free radicals, keeping the oxidation-antioxidation balance in check. Fruits, vegetables, cereals, mushrooms, drinks, flowers, spices, and traditional medicinal herbs are all sources of exogenous antioxidants (Xu *et al.*, 2017). The DPPH assay is a common method for determining an antioxidant molecule's ability to scavenge free radicals and is regarded one of the most common and straightforward colorimetric methods for determining the antioxidant capabilities of pure compounds (Cheng *et al.*, 2006). The results of antioxidant activity were recorded in (Table 3). The DLSM contain DPPH scavenging activity of 31.87% and FLSM has antioxidant activity of 30% and both the samples exhibits similar results. Khan *et al.*, 2017 reported that cow's milk exhibited DPPH scavenging activity of 24.3%, and

buffalo milk exhibited 31.5% of DPPH activity. Thus, the presence of antioxidants can scavenge free radicals and protects our body cells providing various health benefits.

Physical properties of DLSM custard powder

A) Bulk density

The bulk density of powder is a crucial factor in determining how easily it can be handled during processing and transportation (Adejuyitan *et al.*, 2009). Bulk density of formulated custard powder mix was found to be 0.54 ± 0.28 , the bulk density of the control was 0.40 ± 0.34 . Alake *et al.*, 2016 reported that cassava enriched custard powder exhibits a bulk density of 0.90 ± 0.85 . Akinwale *et al.*, 2017 mentioned that bulk density of cassava-based starch to be 0.37 ± 0.00 . Sarkar, 2022 observed the bulk density of custard prepared from Dioscorea to be 0.667 ± 0.02 . This implies that formulated LSM powder will occupy less space during packaging and more products can be transported.

Table 4 physical properties of the powder

| Physical properties | Bulk density | Moisture content | Water absorption capacity (%) |
|------------------------|-----------------|------------------|-------------------------------|
| Control | 0.40 ± 0.34 | 7.54 ± 0.40 | 78.4 ± 0.56 |
| LSM custard powder mix | 0.54 ± 0.28 | 6.54 ± 0.30 | 81.3 ± 0.43 |

Values represent Mean \pm Standard deviation

B) Moisture content

Moisture content of LSM custard powder mix has 6.54 ± 0.30 followed by control 7.54 ± 0.40 . The moisture content of custard powder prepared from sweet potato starch and jackfruit seed starch 9.88 ± 0.12 and 10.43 ± 0.04 (Sarkar, 2022). El-Said and Ali, 2020 mentioned the moisture content of custard powder made from red beetroot, golden berry, and dates to be 6.68, 7.57, and 7.62. The LSM custard powder exhibits lower content than the control which is due to spray drying. Spray drying reduced the amount of water for evaporation, resulting in decreased moisture content of the powder and low moisture content aids in storage stability and avoids microbial contamination.

C) Water absorption capacity

The amount of water the protein matrix can hold at room temperature is measured by its water absorption capacity (Akinwale *et al.*, 2017). The LSM custard powder mix results are found to be 80.3 ± 0.43 followed by the control of 78.4 ± 0.56 . Both the samples showed similar results. The water absorption capacity of cassava-based starch was found to be 81.83 ± 0.59 (Alake *et al.*, 2016). The groundnut isolate custard by Arise *et al.*, 2022 also mentioned the value of water absorption to be 83.2 ± 0.6 . The mentioned results indicate that both the findings match with the results obtained from the LSM custard powder mix. The result may be due to the presence of hydrophilic amino acids in the lotus seeds.

Microbial quality of DLSM custard powder

Microbial analysis of any food product, new or existing, aids in recognizing the quantitative and qualitative existence of microorganisms in the given food product.

Table 5 Microbial analysis of the powder

| Type of sample | Total bacterial count (cfu/g) | Yeast and mould count |
|------------------------|-------------------------------|-----------------------|
| Control | 15×10^2 | Nil |
| LSM Custard powder mix | 5×10^1 | Nil |

The total bacterial count technique and nutrient agar were used for the microbial analysis. By examining the colonies

that formed, particularly those of bacteria, the bacterial count was estimated (Bonnet *et al.*, 2020). The microbial quality was determined by assessing its total bacterial count which is present in (Table 5), the total bacterial count for custard powder was found to be 5×10^1 cfu/g, and the control exhibited total bacterial count of 15×10^2 cfu/g and there was no growth found in yeast and mould growth for both the samples. The total bacterial count of the custard powder mix and the control was within the recommended limit prescribed by the International Microbiological Standards Recommended limit of bacteria contaminants for food of less than 10^6 cfu/g as well as that of the center for food safety (10^5 to $<10^6$ cfu/g) (Shobha *et al.* 2011).

Proximate composition of DLSM custard powder

A) Determination of energy content

Food energy is the energy released from proteins, carbohydrates, lipids, and other organic components. When the three primary calorific components of a diet are entirely burned with enough oxygen and measured in kilojoules or kilocalories, energy or food calories are released (Bo *et al.*, 2020). The energy released from the formulated custard powder was found to be 227.61 kcal. The control sample exhibited the range of 275.72 kcal.

Table 6 Proximate composition of LSM custard powder

| Parameters | Control (per 100g) | LSM custard powder mix (T ₃) (per 100g) |
|------------------------|---------------------------|---|
| Energy (Kcal) | 275.72 | 227.61 |
| Protein | 30.8 ± 1.35 | 33.84 ± 1.01 |
| Fat | 1.392 ± 0.11 | 1.402 ± 0.15 |
| Carbohydrate | 35 ± 1.48 | 19.91 ± 1.0 |
| Ash | 2.83 ± 1.13 | 2.474 ± 1.0 |
| Calcium | 1.257 ± 0.54 | 3.947 ± 1.0 |
| Magnesium | $72 \text{mg} \pm 0.87$ | $94 \text{mg} \pm 1.0$ |
| Potassium | $82.6 \text{mg} \pm 0.50$ | $70.5 \text{mg} \pm 1.17$ |
| Vitamin C | $2 \text{mg} \pm 0.45$ | $20.33 \text{mg} \pm 1.3$ |
| Vitamin B ₆ | Nil | $2.48 \text{mg} \pm 1.11$ |

Values represent Mean \pm Standard deviation

B) Determination of protein content

The protein content of LSM custard powder and control was mentioned in table 6 and found to be 33.84 ± 1.01 followed by control of 30.8 ± 1.35 . The LSM custard powder exhibits higher range of protein control than control. Awoyole *et al.*, 2016 mentioned the protein content of cassava-based custard powder to be 7.92g. Cassava-enriched custard powder exhibits a protein range of 15.45g. Ezegbe *et al.*, 2021 reported the protein content of custard to be 9.91g. Goldenberry custard powder showed a protein content of 14.65g. Thus, the higher protein content of LSM custard powder is suitable for adding to food compositions in order to increase the protein content. Hence it can also be used to fortify various food products.

C) Determination of fat content

The fat composition of LSM custard powder exhibits the range of 1.402 ± 0.15 and 1.392 ± 0.11 range was observed in the control. Alake *et al.*, 2016 reported the fat content of cassava-enriched custard powder to be 2.75g. The cassava-based custard powder by Awoyole *et al.*, 2017 has fat content of 8.38g. Ezegbe *et al.*, 2021 showed the fat results to be 2.03g custard powder developed from sweet potato and corn starch. Both the control and LSM custard powder showed similar fat content. This implies that the custard powder will not exhibit a rancid flavor and lower fat content will lead to the reduction of cholesterol content, and the inclusion of heart-healthy unsaturated fatty acids, and is ideal for people suffering from obesity, hypercholesterolemia, and other heart diseases.

D) Determination of carbohydrate content

The LSM custard powder showed the carbohydrate composition to be 19.91 ± 1.0 and the control has 35 ± 1.48 . Higher content of carbohydrates was found in the control due to the presence of lactose. The cassava-based starch showed a range of 72.35g (Alake *et al.*, 2016). Ezekbe *et al.*, 2021 exhibit a carbohydrate result of 68.87g. The low carbohydrate concentration of LSM custard powder was due to soaking process which affected the total nutrient quality of Lotus seed that resulted in the reduction of carbohydrate content (El-Adawy *et al.*, 2000).

E) Determination of ash content

The term ash content describes the inorganic byproduct left over after the removal of water and organic matter by heating which is carried out in the presence of oxidizing agents. The amount of ash content present in the sample accounts the total amount of mineral present in the food (Ahsan *et al.*, 2015). The ash content of LSM custard powder found to be 2.474 ± 1.0 and the control exhibits the range of 2.83 ± 1.13 . Ezegbe *et al.*, 2021 also reported similar results of 2.35g and cassava-based custard powder has a range of 1.68g (Awoyole *et al.*, 2016). The amount of ash content present in the sample accounts for the total amount of minerals present in the food. Although minerals represent a small proportion of dry matter, often less than 7% of the total, they play an important role from a physicochemical, and nutritional composition of food.

F) Determination of minerals and vitamins

The microminerals were subjected to analysis and the results are found to be the calcium of LSM custard powder 3.947 ± 1.0 and control 1.257 ± 0.54 . The LSM custard powder exhibits a higher range of calcium than the control. The calcium content of whole prickly pear and red beetroot custard powder showed calcium content to be 237.53mg and 241.16mg (El-said and Ali, 2020). Calcium is linked to strong bones and teeth, and also plays a critical role in blood clotting, assisting with

muscular contraction, regulating regular heartbeats and nerve activity, lower blood pressure (Cormick and Belizán, 2019).

The *potassium* of LSM custard powder is 70.5 ± 1.17 and control 82.6 ± 0.50 . The control exhibits a higher potassium range. Potassium is very important for the human body and acts as the main enzymatic co-factor maintaining the acid-base balance.

Magnesium of LSM custard powder 94 ± 1.0 and control 82.6 ± 0.50 . The LSM custard powder exhibits higher Mg content than the control. Magnesium is useful for preventing depression, renal calculi and cataract development (Schwalfenberg and Genius, 2017).

Vitamin B6 of LSM custard powder exhibits the range of 2.48 ± 1.11 and control shows nil concentration and vitamin C content in LSM custard powder of 20.33 ± 1.3 and the control shows the result of 2 ± 0.45 . The vitamin B6 and vitamin C content of lotus seed was found to be 3.03g and 39.4g (Zheng *et al.*, 2003) which shows that the obtained results were similar to the exhibited value.

Table 7 DPPH activity

| Name of the sample | Scavenging % |
|--------------------|--------------|
| Control | 39.38 |
| LSM custard powder | 80.45 |

Antioxidant activity of DLMS custard powder

Reactive nitrogen species (RNS) and reactive oxygen species (ROS) in biological systems, can oxidize lipids and proteins in cells and damage DNA. Free radicals may typically be scavenged by the body's antioxidant system, which helps to maintain the proper balance between oxidation and antioxidants. Exogenous antioxidants are found in a variety of foods, including fruits, vegetables, cereals, mushrooms, beverages, flowers, spices, and conventionally used medicinal plants (Lobo *et al.*, 2010). The DPPH assay is a common method for determining an antioxidant molecule's ability to scavenge free radicals and is regarded as one of the most common and straightforward colorimetric methods for determining the antioxidant capabilities of pure compounds (Cheng *et al.*, 2006). The results of antioxidant activity for LSM custard powder and control were recorded in (Table 7). LSM custard powder was 80.45% and control has 39.38%. The antioxidant activity of custard powder prepared from dates was found to be 72.81% (El-said *et al.*, and Ali, 2020). The LSM custard powder exhibits higher antioxidant activity than the control. The higher antioxidant activity is capable of reducing degenerative diseases such as arteriosclerosis, cardiovascular disease, and cancer.

Table 8 phytochemical test of DLMS custard powder

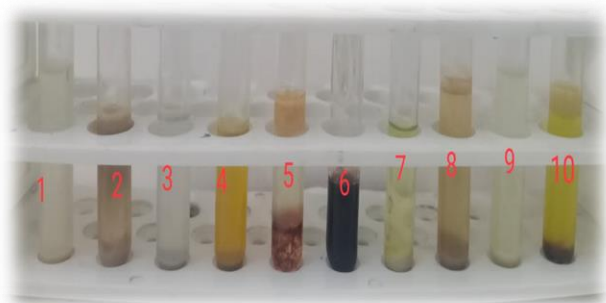
| Phytochemical Tests | Results |
|---------------------|---------|
| Alkaloids | ++ |
| Phenol | - |
| Flavanoids | - |
| Coumarins | +++ |
| Terpenoids | ++ |
| Quinones | +++ |
| Aminoacid | +++ |
| Anthracyanine | - |
| Saponin | - |
| Cardiac Glycosides | ++ |

Phytochemical analysis of DLMS custard powder

Phytochemicals are non-nutritive plant compounds with defense or disease-fighting effects. They are non-essential nutrients produced mostly by plants to protect them.

Phytochemicals in the diet may provide health benefits including protection against degenerative diseases like cancer, cardiovascular disease, and neurological disease (Zhang, 2015).

The obtained results indicates that the LSM custard powder has higher coumarin, quinone and aminoacid content with moderate presence of alkaloids, terpenoids and cardiac glycosides. Coumarins have antioxidant characteristics that may contribute to their anti-inflammatory (Sharifi-rad *et al.*, 2021). Madeo *et al.*, 2013 reported that Quinones play a variety of roles in medicine, including arteriosclerosis, anti-aging, and anti-cancer agents. Lotus seed contains high amount of amino acid including both essential and non-essential amino acid contributing to the increase of protein content (Zhao *et al.*, 2018). The presence of alkaloids, terpenoids and cardiac glycosides. Alkaloids play an important role in the defence systems against pathogens and animals (Patel *et al.*, 2012). Terpenoids have been discovered to have antibacterial, antifungal, antiparasitic, and immunomodulatory effects that can help prevent and treat a number of illnesses, including cancer (Thoppil and Bishayee, 2011). DLSM custard powder are rich in phytochemical and the presence improves the health benefits.



Physical characterization of custard developed from DLSM custard powder

Texture analysis

Texture is a criterion by which quality is judged and an important factor when selecting or rejecting products (Rosenthal, 1999).

The springiness of food is inversely proportional to hardness (firmness increases, elasticity decreases) (Kreungngern and Chaikham, 2016). As the springiness increases, more mastication energy is required to chew the food.

Table 10 Sensory evaluation for different treatment

| Treatment | Colour | Flavour | Appearance | Taste | Overall acceptability |
|--------------|----------|----------|------------|----------|-----------------------|
| Control | 8.0±0.81 | 7.7±0.48 | 7.6±0.59 | 7.3±0.72 | 7.0±0.67 |
| DLSM custard | 7.8±0.76 | 8.1±0.61 | 8.0±0.50 | 8.8±0.67 | 8.0±0.55 |

Values represent Mean ± Standard deviation

Colour

The colour of the custard sample prepared from the control and DLSM custard powder was evaluated through sensory attributes and results are provided in (Table 10). The custard exhibited a colour range of 7.8±0.76. The highest range of colour was found in control followed by T₁. Arise *et al.*, 2022 showed the colour attribute developed from groundnut custard to be 6.93±0.7. Ezegbe *et al.*, 2021 reported that custard exhibits the range of 8.10±1.21. The lower colour attribute in T₃ may be due to the presence of yellow-brown coloured peel of the seeds.

Flavour

Food and beverage acceptance is mostly determined by their flavour. It is a dynamic sense that encompasses the

The springiness of LSM custard was found to be 0.912±0.3 and Shaikh *et al.*, 2017, also reported similar results of springiness of pearl millet custard to be 0.99±1.0 and Ogundele *et al.*, 2020 found the springiness of cassava root custard to be 0.60. It indicates that the custard possesses a good texture and is non-sticky.

Cohesiveness is a measure of how quickly a material deforms when subjected to mechanical force, which is correlated to the internal structure's strength and how hard it is to break down the internal links (Hamed *et al.*, 2018). The cohesiveness of LSM custard was found to be 0.74±0.10. Ogundele *et al.*, 2020 reported that the cohesiveness of cassava root custard to be 0.67±0.14 which was lower than LSM custard. This shows that the developed custard is easy to chew and swallow when it has a low cohesiveness value and the results indicates that both possess close results. Chewiness is one of the crucial textural qualities for a food product and it symbolizes the effort needed to masticate a solid item into a state that is ready for swallowing (Calvarro *et al.*, 2016). The chewiness of the LSM custard was reported as 0.59±0.3N. Ogundele *et al.*, 2020, based on cassava root custard results reported that two variations exhibit the results of 0.58±0.43N and 1.96±1.77N. Shaik *et al.*, 2017, also reported the chewiness of millet custard to be 0.46±1.0N. The obtained LSM custard results are closer to the results mentioned above and it can be attributed to the presence of high-amylose starch content.

Table 9 Texture analysis of custard developed from DLSM custard powder

| Texture property | Springiness | Cohesiveness | Chewiness |
|------------------|-------------|--------------|-----------|
| Custard sample | 0.912 ±0.3 | 0.80±0.1 | 60.4±0.3 |

Values represent Mean ± Standard deviation

Sensory evaluation of the custard developed from DLSM custard powder

The sensory evaluation was carried out for the custard prepared from the custard powder by a semi or untrained panelist of science graduate on the nine-point hedonic scale with respect to color, flavor, appearance, taste, and overall acceptability prepared at room temperature. The sample was prepared using two different samples and subjected to sensory evaluation and the best variation was chosen. The data collected from the sensory is statistically analyzed with mean and standard deviation.

product's smell and taste. From Table 10, it is revealed that the scores for the flavour of the custard are prepared from DLSM custard powder is 8.1±0.61 and control exhibits range of 7.7 ± 0.48. Akinwale *et al.*, 2017 showed the flavour attribute in the custard to be 6.82 ± 1.47. Spada *et al.*, 2017 also reported that soy-based custard exhibits the result 6.88 ± 1.78. Both the findings possess lower flavour value than LSM custard. The aromatic flavour maybe due to the presence of vanilla bean which improves the flavour of custard.

Appearance

The appearance of food consists of its colour, size, dullness or gloss, transparency or turbidity. A consumer takes those qualities and selects a food because these are the indeed qualities of food that are taken into account (Vaclavik and

Christian, 2007). The appearance of prepared custard from control and the variation were evaluated through sensory and the results were provided in table 10. The control has 7.6 ± 0.59 and DLSM custard showed of appearance of 8.0 ± 0.50 . Spada *et al.*, 2017, found the soy-based custard to be 7.06 ± 1.42 and ogundele *et al.*, 2020 mention the obtained outcome to be 7.53 ± 0.86 from cassava enriched custard which has similar result value of LSM custard. Arise *et al.*, 2022 reported the results to be 5.83 ± 0.1 . The jelly-like appearance is majorly due to the presence of high starch content of lotus seeds.

Taste

The taste sensitivity depends on the length of time permitted to taste food. Sensitivity to a specific taste also depends upon the concentration of the substance responsible for taste (Vaclavik and Christian, 2007). The above table 10 mentions the taste of custards made from LSM custard powder and control. DLSM custard results are found to be 8.8 ± 0.67 and the custard prepared from control showed 7.3 ± 0.72 . The T_3 showed higher taste sensitivity than the control. According to Alake *et al.*, 2016 the taste of cassava-enriched custard possessed a value of 7.52 ± 1.05 . Ezegbe *et al.*, 2021 exhibits the taste value of custard to be 7.20 ± 1.47 . Shaik *et al.*, 2017

reported the result of pearl millet custard to be 6.13 ± 1.55 . The presence of sugar and vanilla bean and the sweet content of lotus seed has improved the taste of the custard.

Overall acceptability

The overall acceptability includes all the mentioned sensory attributes together. The LSM custard exhibits the range of 8.0 ± 0.55 followed by control of 7.0 ± 0.67 . Spada *et al.*, 2017 reported the overall acceptance of soy-based custard to be 6.92 ± 1.47 . Shaik *et al.*, exhibited the value of pearl millet custard to be 7.06 ± 1.09 . Ogundele *et al.*, 2020 reported the value of cassava-based custard to be 7.07 ± 0.94 . Thus, the overall acceptability of LSM custard showed good results and has higher acceptance than the mentioned results.

Shelf-life analysis for the DLSM custard powder

Microbial analysis

The shelf life of a food product may be defined as the time between the production and packaging of the product and at the point at which it becomes unacceptable under defined environmental conditions. It is important to find out the shelf life of developed LSM custard powder and thereby ensure the safety of the product (Ellis, 1994).

Table 11 Shelf-life analysis of LSM custard powder

| Days | Control | | LSM Custard powder mix | |
|----------------------|----------------------------|----------------------------|----------------------------|----------------------------|
| | Bacterial count (CFU/g) | Yeast and mould (CFU/g) | Bacterial count (CFU/g) | Yeast and mould (CFU/g) |
| 0 th day | 15×10^2 | Nil | 5×10^1 | Nil |
| 1 st day | 18×10^2 | Nil | 10×10^1 | Nil |
| 10 th day | 30×10^3 | Nil | 25×10^2 | Nil |
| 20 th day | 50×10^3 | Nil | 40×10^3 | Nil |
| 30 th day | 70×10^4 | Nil | 60×10^3 | Nil |

On the 0th day, there is a bacterial count of 15×10^2 in the control and LSM 5×10^1 cfu/g exhibits yeast and mould count, on the 1st day there is a bacterial count of 18×10^2 in control and LSM custard powder mix exhibits 10×10^1 cfu/g. No growth of Yeast and mould was examined for both control and LSM custard powder, on the 10th day there is bacterial count of 30×10^3 cfu/g for control and 25×10^2 cfu/g for LSM custard powder. Yeast and mould count observed to be NIL, and on the 20th day, control shows 50×10^3 and LSM custard powder shows 40×10^3 cfu/g. There is no yeast and mould count found in both the samples.

On the 30th day, the bacterial count of the LSM custard powder (100g of sample) was 60×10^3 cfu/g respectively and the control showed the results of 70×10^4 cfu/g. Though both the sample exhibited bacterial count, the microbial growth does not exceed the permitted limit. It can be concluded that the total bacterial count, yeast and mould count of the LSM custard powder was within the permitted limit due to the decreased moisture content, which does not support the growth of microorganisms. The total bacterial count of the custard powder mix and the control was within the recommended limit prescribed by the International Microbiological Standards Recommended limit of bacteria contaminants for food of less than 10^6 cfu/g as well as that of the center for food safety (10^5 to $<10^6$ cfu/g) (Shobha *et al.*, 2011).

CONCLUSION

The presence of various micronutrients and macronutrients in lotus seed is the important parameter for the selection of value-enhanced custard powder. Commercial custard powder has been loaded with various flavouring agents and high amount of sugar with additives. Vegan products lack in various nutrient quality in plant-based milk food products and it is important to enhance the overall quality. The presence of nutraceutical and bioactive component makes the lotus seed to be unique therapeutic product. The LSM custard powder exhibits higher nutrient values with higher sensory attributes. The microbial growth of the custard powder was also found to be within the safer range and the presence of low fat indicates it exhibits low rancid flavour of the formulated custard powder. It provides various health benefits including antioxidant activity, anti-adipogenic, antibacterial, antifungal, antiparasitic, and immunomodulatory effect etc. These findings infer that, LSM custard powder can be used to provide alternative, cheap and nutritious food that can be used to uplift social economic condition of people associated with protein energy malnutrition. Some of the functional properties (such as the bulk density and water holding capacity) of the LSM custard powder has industrial potential to become a new source of raw materials for the production of lotus seed-based products, confectionery products, and various other food products.

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