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An Observational Study on Enhancing the Nutritional Benefits by Addition of Germinated and Sprouted Flaxseeds

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Abstract: *Flaxseeds, or linseed (*Linum usitatissimum*) popularly known as Alsi in Hindi is establishing importance in the food chain as a functional food. Due to its excellent nutritional profile and health benefits it has become an attractive ingredient in the diet across the world. The main functional component of flaxseed was alpha-linolenic acid, PUFA (polyunsaturated fatty acid). The aim of the study was to study germination and sprout of the flaxseeds. The germination and sprouting was done using organic flaxseeds which are free of gluten. Normal red soil was used for germination. Germination was successfully completed within 6-7 days under favorable conditions, whereas sprouting was completed within 3 days using a muslin cloth. A musk melon flavored smoothie was prepared using germinated and sprouted flaxseeds. The smoothies were subjected to sensory evaluation by 20 panelists. The sprouted flaxseed smoothie was accepted by the panelist in all the sensory attributes.*

Keywords: *Flaxseed, Functional Food, Health benefits, Alpha Linolenic-Acid, Poly unsaturated fatty acid, Sensory Attributes.*

I. INTRODUCTION

Flaxseed or linseed (*Linum usitatissimum* L.) has been used as food and medicines in many countries. Flaxseed comes from the flax plant, which is an annual herb. It has been used in various forms such as flour, oil and seed. Flaxseed and flaxseed oil is considered as healthy due to presence of various bioactive compounds in it.[1] Flaxseed, or Linseed (*Linum Usitatissimum*), popularly known as Alsi, Jawas, Aksebija in Indian languages, is a blue flowering rabi crop and a member of family Linaceae. Annual production of flax was 3.06 million tons and Canada is the world's largest producer of flax (about 38% of total production) (Anonymous, 2000). Globally, Flaxseed is grown as either oil crop or a fiber crop with fiber linen derived from the stem of fiber varieties and oil from the seed of linseed varieties [2].The seed is a healthy source of oil containing poly-unsaturated fatty acids, digestible proteins, and lignans. Major nutritional components of flaxseed include ALA rich oil, protein, minerals and a greater proportion of non-nutritional lignan-rich dietary fiber. On the dry weight basis, flaxseed contain 20% protein, 27% total dietary fiber 41% oil, 4% ash and 8% moisture. Albeit, flaxseed proteins are not complete in nature but the deficiency may be overcome effectively by enrichment with products containing amino acids that form complete proteins. [1] The spherical fruit capsules contain two seeds in each of five compartments. The seed is flat and oval with a pointed tip. It have smooth glossy surface. It varies in color dark brown to yellow. The texture of flaxseed is crisp and chewy possessing a pleasant nutty taste. Beyond its oilseed crop ability, proximate composition of flaxseed makes it more promising for its utilization in different food products. Flaxseed is one of the richest vegetarian source of α - linolenic acid (omega 3 fatty acid) and soluble mucilage. In present era, consumer's trend towards functional food has increased significantly as health awareness rose. Flaxseed can be one stop for novel high quality source of nutrition. [2]

The life of every organism begins with a reproduction phase; developmental processes such as growth and organ formation are following. Further, these are followed by reproductive phases, which lead to the next generation. The cycle ends with this phase. The main interested objects here are also seeds.[3] The whole flaxseed is flat and oval with pointed tips and contains a seed coat or true hull (also called testa), a thin endosperm, two embryos and an embryo axis (Morris, 2007)[4][5]. Every part of the linseed plant is utilized commercially, either directly or after processing. Flax seed sprouts are edible, with a slightly spicy flavor. Whole flax seeds are chemically stable, but ground flaxseed can go rancid at room temperature in as little as one week, although there is contrary evidence. Refrigeration and storage in sealed containers will keep ground flax from becoming rancid for a longer period [4] Flaxseed is establishing importance in the world's food chain as a functional food. Functional food can be defined as the food or food ingredients that may provide physiological benefits and helps in preventing and/or curing of diseases. Presently, flaxseed has new prospects as functional food because of consumer's growing interest for food with superb health benefits. Owing to its excellent nutritional profile and potential health benefits, it has become an attractive ingredient in the diets specially designed for specific health benefits (Oomah 2001. [6][7])

A. Physiological Properties

Flaxseed is establishing importance in the world's food chain as a functional food. Functional food can be defined as a food or food ingredients that provides physiological benefits and helps in preventing and/or curing of diseases. Presently, flaxseed has new prospects as functional food because of consumer's growing interest for food with health benefits. Owing to its excellent nutritional profile and potential health benefits, it has become an attractive ingredient in the diets specially designed for specific health benefits. ALA is one of the essential polyunsaturated fatty acid with anti-inflammatory, anti-thrombotic and anti-arrhythmic properties. Nutritionists all over the world suggest incorporation of omega 3 fatty acid sources in the diet. Flaxseed serves as the good alternative source of omega-3 fatty acid in population who do not eat fish. Edible flaxseed products include the whole flaxseed, ground meal and extracted oil or mucilage.

B. Nutritional Composition

Among the functional foods, flaxseed has emerged as a potential functional food being good source of alpha-linolenic acid, lignans, high quality protein, soluble fiber and phenolic compounds.

C. Protein

The protein content in flaxseed has been reported to between 10.5% and 31%. Like all vegetables, flaxseed proteins have functional properties that affect their behavior in a food system through interaction with other ingredients. These properties are mainly dependent on their hydration mechanisms for solubility and water oil retention capacity. The amino acid pattern of flax protein is similar to that of soybean protein, which is viewed as one of the most nutritious of the plant proteins (Rabatafika et al., 2011)[9]. Differences in protein can be attributed to both genetics and environment. The proximate protein content of de hulled and defatted flaxseed varied considerably depending upon cultivar growth location and seed processing. Hull fraction contains lower protein levels and that de hulling increases protein level of flaxseed protein level from 19.2% to 21.8%.(Oomah and Mazza , 1997)[8] Flaxseed proteins have similar nitrogen extractability at varying pH and ionic strength with other oilseed sources of proteins. Albumin and globulin type proteins are the major proteins in flaxseed. Nutritional value and amino acid profile of flaxseeds are comparable to that of soya proteins. Flaxseed albumin comprised 20% of meal protein (Madhusudhan and Singh, 1983)[10]. Globulin fraction makes up to 73.4% and the albumin constitutes about 26.6% of total protein (Marcone et al., 1998)[11]. Flaxseed proteins are relatively high in arginine, aspartic acid and glutamic acid whereas lysine, methionine and cysteine are limiting amino acid. Total amino acid content of the flaxseed after 8 days germination increased by 15 times with greatest increase (i.e. 200 times) being observed in glutamine and leucine compared to the original seed (Wanasundara et al., 1999).[13]

D. Omega-3- Fatty Acids

Omega-3 fatty acid is known as essential fatty acid because humans cannot introduce a double bond beyond the ninth carbon from carboxyl end of fatty acid. The metabolism of essential fatty acids is depicted in Fig. 1. ALA serves as the precursor for the synthesis of polyunsaturated fatty acids EPA (Eicosapentaenoic acid) and DHA (Docosahexanoic acid). During the transformation of ALA into EPA and DHA, a series of fatty acids belonging to n-3 PUFA family are also synthesized via desaturation and elongation reactions in the presence of specific desaturases and elongases. Similarly, linolenic acid is also synthesized using similar enzymatic reactions. It has been reported that the conversion of ALA to EPA and DHA is not very efficient in humans and animals and there exist competition between both the fatty acids for the same enzyme. Long chain PUFAs, EPA and DHA are further metabolized by the enzymes cyclooxygenase and lipoxygenase to eicosapentnoids, prostaglandins, leukotrienes. Among these eicosanoids, E2 series prostaglandins, leukotrienes B4 derived from linoleic acid are the key metabolites which are responsible for many inflammatory diseases like cardiovascular diseases and arthritis, while eicosanoids and E3 series prostaglandins derived from linolenic acid have anti-inflammatory responses (James et al. 2000; Funk 2001; Barcelo-Coblijn and Murphy 2009; Kaur et al. 2012)[13]. Therefore, it is advised that human beings should consume a diet that contains a balanced ratio of omega-3 and omega-6 essential fatty acids. The two groups of essential fatty acids compete with each other for placement within cell membranes. If the intercellular environment has a higher proportion of one type of fatty acid as compared to the other, it is likely that the predominant fatty acid will be incorporated into cell membrane, resulting in adverse effects in the fluidity of the cell membrane affecting cellular functions and overall health of the cell. If there is an equal proportion of both the essential fatty acid in the intercellular environment, there is selective preference for omega-3 fatty acid. Both these fatty acids have opposing, yet necessary, influences over physiological functions [13] (Lunn and Theobald 2006; Kaur et al. 2012s [14]

E. Dietary Fiber

Flaxseeds serve as a good source of both soluble and insoluble dietary fiber. Flaxseed holds a unique place among the oilseeds due to the presence of mucilage located in the outer layers of the seed (Singh et al. 2011a,b)[12]. Flaxseed mucilage has gained momentum due to its superb health benefits and potential functional properties (Susheelamma 1987; Mazza and Biliaderis 1989). It contains 35–45 % of fiber and two-thirds is insoluble and one-third is soluble fiber. Insoluble fiber consists of cellulose, hemicellulose, and lignin (Morris 2007; Oomah and Mazza 1993). Most of the soluble fiber of flaxseed appears to be the mucilage of the seed coat. It makes up 7–10 % of seed weight (Mazza and Biliaderis 1989). Soluble fiber in the form of mucilaginous material consists mainly of water-soluble polysaccharides. [12]

F. Lignans

Lignans are phytoestrogens, which are abundantly available in fiber-rich plants, cereals, legumes, vegetables, fruits, berries, tea. Flaxseed contains about 75–800 times more lignans than cereal grains, legumes, fruits, and vegetables.

Secoisolariciresinol diglycoside (SDG) is the major lignan of flaxseed, along with minor contents of matairesinol, pinorensinol, lariciresinol, and isolariciresinol (Meagher et al. 1999; Sicilia et al. 2003; Krajcova et al. 2009)[14]. Secoisolariciresinol (SECO) is produced by acid hydrolysis of secoisolariciresinol diglycoside. Structures of the flaxseed lignans compiled from different sources (Toure and Xueming 2010; Meagher et al. 1999)[15]. SDG is metabolized by bacteria in the colon of humans to synthesize mammalian lignans known as enterodiols (END) and enterolactone (ENL) (Chen et al. 2007)[16]. In the human body, the lignans are acted upon by the gastrointestinal microflora to release secoisolariciresinol (SECO), the non-sugar moiety of SDG. Further hydroxylation and demethylation by the microflora lead to the production of mammalian lignan-enterodiols (END), which is then oxidized to give enterolactone (ENL). Bacteroides as well as Clostridia have been identified to release the glucosyl moieties from SDG to yield SECO (Clavel et al. 2006; Struijs et al. 2009)[17]. *Peptostreptococcus productus*, *Eubacterium callanderi*, *Eubacterium limosum*, and *Bacteroides methylotrophicum* are found to be responsible for carrying out demethylation reactions, while dehydroxylation reactions are carried out by *Eubacterium lentum* (Wang et al. 2000; Clavel et al. 2007)[18]. The dehydrogenation of END into ENL has been carried out by several Clostridia and *Ruminococcus* sp. (Clavel et al. 2007; Jin et al. 2007)[19]. The END and ENL, so formed, can be excreted in faeces or are absorbed by the human colon and enter the circulation.[12]

II. HEALTH BENEFITS OF FLAX SEEDS

A. Flaxseed in Cardiovascular Disease

Flaxseed has recently gained attention in the area of cardiovascular disease primarily because it is the richest known source of both alpha-linolenic acid (ALA) and the phytoestrogen, lignans. ALA may prevent ASCVD through several mechanisms, including decreased inflammatory response, inhibition of platelet aggregation and thrombosis, decreased blood pressure, improved serum lipids, and prevention of cardiac arrhythmias (Lanzmann-Petithory D)[23].

The bulk of the evidence from nine clinical trials suggests that whole flaxseed or its powder (15–50 g/day) can modestly reduce total cholesterol and LDL by 1.6 to 18% in both hypercholesterolemic and normocholesterolemic patients without a significant effect on HDL or triglycerides (TG).

The results have not all been consistent, however, with two most recent trials showing no significant effect of flaxseed on LDL in healthy patients with hypercholesterolemia. Data on extracted flaxseed oil is much less consistent, with a few studies finding a modest reduction in TG with large doses (60 mL -4 tablespoons/day), but most finding no effect. Flaxseed oil given alone does not appear to have any beneficial effects on total cholesterol, LDL, or HDL. While there are no human studies investigating the effects of extracted flax lignans on lipids, Kuroda et al. [21] evaluated the lipid effects of a series of synthetic lignans (arylnaphthalene lignans), and reported they significantly lower TC and LDL and raise HDL.

Arjmandi et al [22]. conducted a double-blind, randomized, crossover trial to compare the lipid effects of whole flaxseed with sunflower seed (control) in 38 moderately hyperlipidemic postmenopausal women. Subjects were randomly assigned to receive 38 g of either whole flaxseed or whole sunflower seed baked into breads and/or muffins for 6 weeks.

At the end of 6 weeks, subjects entered a 2-week washout period and then received the other treatment regimen for the remaining 6 weeks. The LDL results in this study were impressive, but may have been influenced by limitations of this study. The researchers did not report potential differences in dietary saturated fat intake between treatment periods, which might have confounded the results. [20]

B. Flaxseed in Diabetes

Flaxseed also impacts another major disease that is growing in incidence across the globe: diabetes. Flaxseed supplementation reduced blood glucose in subjects with type 2 diabetes [24,25] and lowered blood glucose in subjects with prediabetes [26]. Flaxseed derived gum and lignan supplement also decreased blood glucose in subjects with Type 2 diabetes [27,28]. Pre-clinical studies have reported anti-hyperglycemic effects of the flaxseed lignan SDG in animals with Type I diabetes [29]. Whether SDG or flaxseed supplementation improves glycemic control in human subjects with Type 1 diabetes remains unknown and could be a topic of experimentation in future.

C. Flaxseed in Cancers

Flaxseed is already used extensively in animal studies to treat a variety of cancer [30]. Perhaps the most studied cancer with respect to the impact of dietary flaxseed is breast cancer. In both experimental animal studies [30,31] and in human trials [30,32], dietary flaxseed has significant protective effects against breast cancer. A systematic review of 10 human trials led to conclusion that flaxseed reduced tumor growth in women with breast cancer [30,32]. They also found evidence in support of flax associated protection against primary breast cancer as well as a reduced risk of mortality in women living with breast cancer [32]

III. METHODOLOGY

A. Product Development

The process of creating, processing, and commercializing a new food product which is high in terms of nutrient and other health benefits.

The value added products developed in this study are “Flax seed smoothies” which are highly nutritious and can be consumed by all age groups. The flaxseeds were incorporated as sprouted and germinated so as to enhance the nutritive value.

B. Procurement Of The Sample

Organic flax seed which was gluten free and grown without synthetic pesticides and GMOS, natural source of soluble and insoluble fibers from 24 mantra brand was used for the preparation of the smoothies. It was available in some of the supermarkets.

C. Method Used For Sprouting Flax Seeds

A clean muslin cloth was used for sprouting flax seeds. The muslin cloth was soaked in water and then squeezed so that it is moist but not soaked.

About 1 teaspoon of flax seeds (organic) was spread on the surface of muslin cloth without touching each other. This muslin cloth containing organic flax seeds was placed in a dark room to protect them from direct light.

The seeds were sprinkled with clean water anytime the muslin cloth looks a little dry as the seeds must be moist to sprout. The seeds were checked at least once in a day. On the second day the seeds started breaking out. The next day, tiny sprouts were observed. On the fourth day, it was observed that sprouting was completed.

D. Germination Of Flax Seeds

Normal red soil was used for the germination of flax seeds (organic). About 2 inch of soil was taken in a pot and the soil was sprayed with water to make it moist. About 1 teaspoon of organic flax seeds were sprinkled on the soil with a little space between them and another spray of water was given to the seed to make it wet. Then the pot containing seeds was placed in a dark room.

On the second day, the seeds are broken out of the shell and these seeds were moist with little water because the soil at the bottom is still moist.

On day three, small sprouts were coming out of the seeds. On the fourth day the pot containing yellow micro plants was sprinkled with water and kept in natural light. On the fifth day, the yellow micro plants completely turned into green. A spray of water was given once in a morning and once in evening. On the sixth day, there was complete germination of flax seeds and the micro greens were ready.



IV. METHODS OF PREPARATION

A. Muskmelon Flavoured Flax Seeds Smoothies

1) Musk Melon Flavoured Flax Seeds Smoothie

a) Ingredients

Musk melon -50g

Yoghurt- 25 g

Flax seeds (organic)-2 teaspoons

Honey – 2 teaspoons

b) Method

Peel the musk melon and cut it into small pieces in a bowl. To this add yoghurt, flaxseeds and mix well. Then add honey and transfer the mixture into a jar and grind it till the desired consistency. Garnish with roasted flax seeds powder and serve chilled

2) *Musk Melon Flavoured Germinated Flax Seeds Smoothie*

a) *Ingredients*

Musk melon -50 g

Yoghurt -25 g

Germinated flax seeds – 10g

Honey- 2 teaspoons

b) *Method*

Peel the musk melon and cut it into small pieces in a bowl. To this add yoghurt, germinated flax seeds (micro plants) and mix well. Then add honey and transfer the mixture into a jar and grind it till the desired consistency. Garnish with roasted flax seeds powder and serve chilled.

3) *Musk Melon Flavoured Sprouted Flax Seeds Smoothie*

a) *Ingredients*

Musk melon -50 g

Yoghurt – 25 g

Sprouted flax seeds – 2 teaspoons

Honey – 2 teaspoons

b) *Method*

Peel the musk melon and cut it into small pieces in a bowl. To this add yoghurt, sprouted flax seeds and mix well. Then add honey and transfer the mixture into a jar and grind it to the desired consistency. Garnish with roasted flax seeds powder and serve chilled



B. *Standardization*

Standardization is a process where a recipe is tested in a number of trials and found to be consistent in quality. It is a trial and error process. The use of standard recipe is a major factor in developing a product. The basic recipe is prepared as per the method of preparation and the difference of smoothies using sprouted flax seeds and germinated flax seeds are used depending upon the quantities required. The prime ingredient in smoothies is flax seeds and we presented the 3 smoothies

1) Standard musk melon flavored flax seeds smoothie.

2) Musk melon flavored sprouted flax seeds smoothie.

3) Musk melon flavored germinated flax seeds smoothie.

The ingredients used were muskmelon, yogurt, honey, flax seeds (organic), sprouted flax seeds and germinated flax seeds.

C. Sensory Evaluation

It is defined as “a scientific discipline used to invoke, which, analyze and interpret reactions to characteristics of foods and materials as they are perceived by the senses of smell, taste, touch according to Mounts and Warner sensory evaluation, which is a necessary part of both product development and quality control. Once the standardization is complete, 20 panelists were selected for both the trials to evaluate sensory attributes of smoothies. The panelists included both students and teachers. The same procedure was followed for both the trials. To minimize any kind of changes in the preparation that might affect taste, texture, color and odor, the sensory evaluation was done as soon as the preparation was served to panelist.

Samples were placed together. A score card was given to rate the smoothies. A glass of water was also provided. To drink in between the assessment of smoothies, so that it becomes easier for the panelist to get the exact taste of smoothies.

D. Shelf Life

Shelf life of the product is done by keeping the product at room temperature for a day. The product was evaluated in terms of sensory attributes like appearance, flavor, color, texture and taste. The smoothie was spoiled in 1-2 hours at room temperature 4-6 Celsius whereas it took 12 hours for the smoothie to spoil at refrigerated conditions.

E. Score Card

The developed products were tasted and scored on a card on a scale of 5 based on the following sensory attributes ,taste ,odor, color, texture , overall acceptability ,score card consisted of name of judge , date , time , signature

V. DISCUSSION

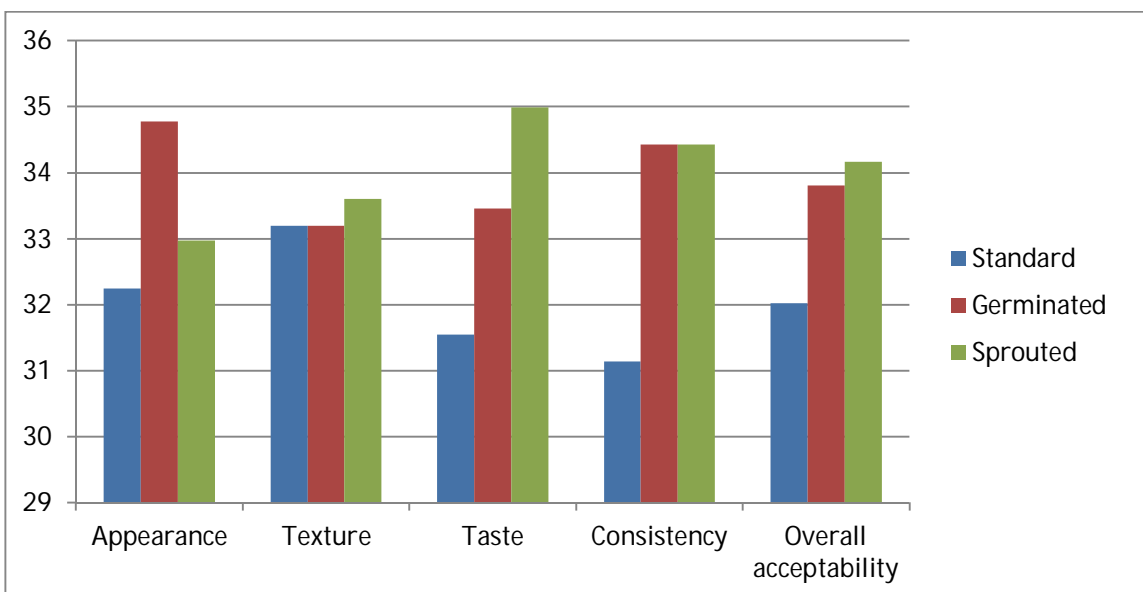
The organic flaxseeds were subjected to sprouting and germination. Sprouting was successfully completed within 3 -7 days were required for complete germination of flaxseeds.

Smoothies were prepared using sprouted and germinated flaxseeds which were evaluated based on the sensory attributes like appearance, texture, taste, consistency and overall acceptability. Two trials were done and the number of panel members was 20.

The appearance of germinated flaxseeds smoothie was found to be acceptable by the panelists and the texture, taste of sprouted flaxseed smoothie was more acceptable by the panelist. In terms of consistency both the sprouted and germinated flaxseeds smoothie were equally accepted.

The overall acceptability of sprouted flaxseeds smoothie was the highest.

VI. RESULT OF SENSORY EVALUATION FORMUSKMELON FLAVOURED STANDARD, GERMINATED AND SPROUTED FLAXSEED SMOOTHIES.



From the above graph, it was observed that the appearance of germinated flaxseed smoothie was more acceptable than standard and sprouted flaxseed smoothie. The texture of sprouted flaxseed smoothie was slightly more acceptable than standard and germinated flaxseed smoothies. The taste of sprouted flaxseed smoothie was dominating the other two smoothies. The consistency of standard flaxseed smoothie was less acceptable than the germinated and sprouted smoothies. The overall acceptability of sprouted flaxseed smoothie was higher than other two smoothies.

VII. CONCLUSION

Flax or Linseed (*Linum usitatissimum*) is a blue flowering rabi crop and a member of family Linaceae, popularly known as Alsi, Jawas, Aksebija in India. It varies in color yellow to dark brown. It is a richest source of alpha-linolenic acid having anti-inflammatory, anti-thrombotic and anti-arrhythmic properties. The major proteins in flaxseeds are albumin and globulins. Flaxseeds are a good source of dietary fiber, both soluble and insoluble fiber. The omega 3 fatty acid, poly unsaturated fatty acids and dietary fiber present in the flaxseed has protective effect against diabetic, anti-atherogenic and Ischemic heart disease. The risk of ovarian cancer can be reduced due to the presence of lignans in flaxseeds which has antioxidant properties. Flaxseeds improve immune system due to presence of arginine and glutamine.

The study was to germinate and sprout flaxseeds. The organic flaxseeds were used for germination and sprouting. Normal red soil was used for germination. Within 6-7 days under favorable conditions germination process was successfully completed.

The sprouting of the flaxseeds was done using a muslin cloth and it was complete within 3 days.

A musk melon flavored smoothies was enhanced using germinated and sprouted flaxseeds. The smoothie was subjected to sensory evaluation based on the sensory attributes such as appearance, texture, taste, consistency and overall acceptability. Two trials were done and the number of panelists was 20. In trial 1 appearance of germinated flaxseeds smoothie was more accepted than the sprouted flaxseeds smoothie. In terms of taste and texture sprouted flaxseeds smoothie was found to be better. The consistency of germinated flaxseeds smoothie was more acceptable. The overall acceptability was higher for sprouted flaxseed smoothie. In trial 2, the appearance of sprouted flaxseeds smoothie was higher and with regards to texture, the germinated flaxseed smoothie was more acceptable. The taste and the consistency of sprouted flaxseed smoothie were dominating. The sprouted flaxseeds smoothie was accepted in overall sensory attributes.

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