

# **PROTEIN ENRICHMENT IN HERBAL WHEY JELLY THROUGH SPIRULINA INCORPORATION**

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**DEPARTMENT OF FOOD PROCESSING TECHNOLOGY**

**ST. TERESA'S COLLEGE, ERNAKULAM**

**MAHATHMA GANDHI UNIVERSITY**

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## ST. TERESA'S COLLEGE, ERNAKULAM



### CERTIFICATE

This is to certify that the dissertation work entitled "**PROTEIN ENRICHMENT IN HERBAL WHEY JELLY THROUGH SPIRULINA INCORPORATION**" is a bonafide work done by **Ms. KRISHNAVENI A. S (Reg No.VM22FPT017)**, student of ST. TERESA'S COLLEGE, ERNAKULAM, MAHATHMA GANDHI UNIVERSITY, in partial fulfillment of the degree of MASTER OF VOCATIONAL IN FOOD PROCESSING TECHNOLOGY. This dissertation work is carried out by her under my supervision and guidance.

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**CERTIFICATE**

This is to certify that the dissertation work entitled " **PROTEIN ENRICHMENT IN HERBAL WHEY JELLY THROUGH SPIRULINA INCORPORATION** " is an authentic project work carried out by **Ms. KRISHNAVENI A.S (Reg. No. VM22FPT017)** under the supervision and guidance of **Mrs. DIVYA K.B.** Assistant Professor, Department of Dairy Technology, Verghese Kurien Institute of Dairy and Food Technology, Mannuthy, Thrissur, submitted in partial fulfillment of requirements for the award of the degree of Master of Vocational in Food Processing Technology, St. Teresa's College Ernakulam, Mahathma Gandhi University.

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## DECLARATION

I, KRISHNAVENI A.S, do hereby declare that the dissertation "**PROTEIN ENRICHMENT IN HERBAL WHEY JELLY THROUGH SPIRULINA INCORPORATION** " is a bonafide record of the project work done by myself in partial fulfillment for the degree of Master of Vocational in Food Processing Technology, St. Teresa's College Ernakulam, Mahatma Gandhi University.

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## **ABSTRACT**

Paneer whey is a byproduct of dairy industry. It can be utilized for the preparation of jelly, it is a semi-solid dessert, which is popular among individuals of all age groups because of its sweet taste and unique texture. The variety phytochemicals and bioactive compounds present in herbs, spices and spirulina gives additional nutraceutical, medicinal property and health benefits. This spirulina contains high in protein, studies shown addition of spirulina in children's diets are often inadequate in terms of essential nutrients, vitamins, and minerals.

Control sample of herbal whey jelly was standardized by adding different levels of ginger and mint. Here, ginger and mint were taken in the following proportions to prepare T1 (2:2), T2 (2:3) and T3 (2:4) respectively. After sensory evaluation T3 (ginger to mint ratio 2:4) received highest score. Therefore, T3 was selected and hereafter used as control as herbal whey jelly. Spirulina incorporated herbal whey jelly was prepared by spirulina at different levels (viz., 0.5%, 1%, 1.5%, 2%) and it was coded as S1, S2, S3 and S4. Sensory evaluation was done to select the best combination and S3 was selected as it received the highest sensory scores.

The prepared spirulina incorporated herbal whey jelly has good flavor, green color, body and texture, acceptable taste and overall acceptability. Sensory evaluation of the product was done by using a Nine-point Hedonic scale on the basis of mouth feel, color and appearance, flavor and overall acceptability. The physico- chemical analysis of herbal whey jelly was analyzed. The protein, total moisture, ash, fat, antioxidant. The color characteristics were determined using Hunters colorlab and textural attributes using texture profile analyser.

**Keywords:** Ginger, Mint, Spirulina, Whey,  $\kappa$ -carrageenan

# 1.INTRODUCTION

(Andrei C et al., 2015) One of the primary foods ingested by humans is milk, which also serves as a crucial foundation for many productive human endeavours. The composition of milk changes based on the species, location. The components in milk are as rich as a newborn's need for a rapid weight growth; the amount of sugar in milk is linked to brain development, and animals living in cold climates have higher milk fat content.

Whey is a byproduct of paneer production, is a versatile and nutritionally dense liquid with a wide range of applications in the food and beverage industry. Extracted during the paneer making process, whey contains high levels of protein, particularly whey protein, which is renowned for its complete amino acid profile and rapid absorption by the body. This makes whey protein a popular supplement among athletes and fitness enthusiasts seeking to support muscle growth and recovery. Additionally, whey is used as an ingredient in various food products, including protein bars, shakes, baked goods, and infant formula, due to its functional properties such as emulsification, foaming, and gelling.

Jelly, a gelatinous substance typically made from fruit juice, sugar, and pectin, is a beloved sweet treat enjoyed in various forms around the world. Its creation involves boiling fruit juice with sugar and pectin, a natural thickening agent found in fruits, until it reaches a thick, spreadable consistency upon cooling. With a smooth texture and vibrant colors, jelly is often used as a topping for toast, pastries, and desserts, or as a filling in sandwiches, cakes, and cookies. It's also a popular ingredient in homemade jams, preserves, and sauces, offering a delightful burst of fruity flavour to a wide range of dishes. Whether enjoyed on its own or incorporated into recipes, jelly adds sweetness and vibrancy to culinary experiences, making it a cherished satisfaction, for people of all ages.

Spirulina, a blue-green algae cultivated for its remarkable nutritional profile, has gained popularity as a superfood in recent years. Rich in protein, vitamins, minerals, and antioxidants, spirulina offers a wide array of health benefits. It is particularly renowned for its high protein content, making it a valuable source of plant-based protein for vegetarians and vegans. Additionally, spirulina contains essential amino acids, iron, calcium, and vitamins like B12 and beta-carotene, which are beneficial for overall health and well-being. Consumed as a dietary supplement in powdered or tablet form, spirulina is often added to smoothies, juices, and energy bars to boost nutritional content. Its vibrant green color and mild, slightly seaweed-like

flavor make it a versatile ingredient in savory dishes, such as soups, salads, and dips. Beyond its nutritional value, spirulina has been studied for its potential therapeutic properties, including anti-inflammatory, anti-cancer, and immune-boosting effects. it promote good health benefit.

Ginger, a flowering plant with a distinctive spicy and aromatic rhizome, has been used for centuries in various culinary and medicinal applications. Renowned for its unique flavor profile, ginger adds depth and warmth to dishes ranging from savory stir-fries to sweet desserts. Beyond its culinary uses, ginger is celebrated for its potential health benefits. It contains bioactive compounds like gingerol, which have potent antioxidant and anti-inflammatory properties. Ginger is often used to alleviate nausea, motion sickness, and digestive discomfort, and it may also help reduce muscle soreness and alleviate symptoms of osteoarthritis.

Mint, a fragrant herb with a refreshing taste, is widely used in culinary, medicinal, and aromatic applications. With its distinctively cooling flavour, mint adds a burst of freshness to dishes, beverages, and desserts. Commonly found varieties include spearmint and peppermint, each offering its unique flavour variation. Mint leaves are often used fresh or dried in salads, sauces, teas, and cocktails, enhancing their aroma and flavour profile. Medicinally, mint has been utilized for centuries to aid digestion, alleviate indigestion, and relieve nausea.

Objectives:

1. Standardisation of herbal whey jelly incorporated with spirulina.
2. Study the effect of incorporation of spirulina on the protein content of herbal whey jelly.
3. Study the physico-chemical characteristics of developed product.

## 2. REVIEW OF LITERATURE

### 2.1 Jelly

(Shinwari et al., 2018) Studied Fruits are abundant source of bioactive substances that have the potential to act as antioxidants, including vitamins, phenolics, carotenoids, and flavonoids. These fruits are processed into shelf-stable goods like jam and jellies to make them available for consumption during the off-season. A food product jam or jelly is made by heating fruits with sugar and either pectin or acid added. It has an intermediate moisture content, making it semi-solid to raise the amount of total soluble solids (TSS) more than 65%. These are shelf-stable, ready-to-eat foods that are popular among kids and can be transported anywhere, even into space by astronauts. Diet-conscious consumers need can be satisfied by optimizing the entire process, including the ingredients and the time-temperature combinations used to produce a product with the fewest calories and the greatest number of functional qualities.

(Park et al., 2021) studied Jelly is a semi-solid delicacy that appeals to people of all ages due to its distinct texture and sweet flavour. Gelling agents like carrageenan and gelatin, sweeteners like sugar and flavouring and colouring additives are used to make jelly. Jelly is mostly made of sugar and gelling agents changes to these ingredients in the jelly's formulation should have an impact on its quality. Large levels of sugar are found in jelly, but earlier research has tried to lower or replace the sugar in jelly products using sugar alternatives made from fruit powder. There are some gelling agents, which include gelatin, pectin, agar, starch, carrageenan, and gellan gum, are used to stabilize jelly. A naturally occurring polymer derived from edible red algae is called carrageenan. Carrageenan undergoes a coil-to-helix transformation during gelation, which is followed by helical aggregation. These ingredients are frequently employed as gelling agents in the making of jelly because of their gelling qualities. characteristics of the gelling substances. For instance, the textural qualities, stability, and sensory attributes of gel-based foods are significantly influenced by the presence of sugar, pH, ionic strength, and temperature.

(S. Titri et al., 2021) studied use ginger juice to make jelly stick items that are similar in texture to those sold in stores. This product is more chewy and sticky, and it is easy to digest. Ginger can strengthen human immunity, has a high antioxidant content, and is abundantly available in Indonesia. A gel-shaped snack known as jelly is created by adding sugar, acid, or other culinary ingredients to hydrocolloid substances. Jelly comes in a variety of forms on the

market, including jelly drinks, jelly candies, and regular jelly, each having a unique feel. Higher carrageenan content in jelly sticks generally results in higher hardness ratings. The reason for this is that kappa carrageenan can create an elastic and rigid gel by forming a strong double helix structure.

(D.S.Mondhe et al., 2018) studied Jellies are goods produced from the juice and/or aqueous extracts of one or more fruits or vegetables, mixed with foodstuffs with sweetening characteristics with or without the addition of water, and brought to a semi-solid gelled form. jelly is persevered by addition of citric acid. This weak organic acid with the formula  $C_6H_8O_7$ , acts as a natural preservative and also imparts a sour taste to food products. It will provide a good acidifier, chelating agent and flavouring agent

(P.K.Wasnik et al., 2015) conducted amount of paneer whey increased, the nutritional content in the treatments improved and paneer whey was acceptable as a 100% replacement for water. The production of paneer whey jelly offers a compelling substitute for utilizing all of the whey at the point of production. This helps dairy plants operate more economically by lowering the expense of treating wastewater while solving the pollution issues brought on by whey drainage. The findings indicate that paneer whey can completely replace water as the primary ingredient in a jelly confection that is acceptable without appreciably changing the sensory attributes, since all of the sensory ratings varied not statistically. As the amount of paneer whey in the treatment increases, the nutrient levels improve. Increased stiffness in jelly-based confections may be attributed to high acidity. Up to 30 days of storage, it was noted that the jelly confection's look was clear and semi-transparent.

(Hanna et al., 2004) This study demonstrated that it is possible to create a jelly with acceptable sensory qualities that is sugar-free or contains no additional sugar by combining gums with a high-intensity sweetener. The jelly might then be sold to people with diabetes or those who want to lose weight. Three jelly formulations were made utilizing xanthan gum or locust bean gum (LBG), either alone or in combination, sucralose, low methoxyl pectin, and maltodextrin. The formulations were stored at 4C or 43C to assess their shelflife. After the proper examination of jelly Compared to using either gum alone, the combination of xanthan gum and LBG significantly reduced syneresis. When xanthan gum and LBG were used together, the hardness and springiness of the valves was much higher than when either substance was used alone.

## **2.2 Whey**

(Nimish et al.,2019) reported as its a transparent watery liquid which remains after removal of fat and casein from milk. When paneer, cheese, casein, chakka, and chhana are made from the coagulation by acid, bacteria or enzymes. Which result in the greenish-yellow color of the watery portion of milk after curd separation. Whey contains soluble elements, including lactose, whey proteins, vitamins, and minerals, its nutritional valuve is high.

(Ahasanul karim et al.,2022) found high organic content, whey is typically seen as a contaminant to the environment. The growing demand for items made from milk is causing a progressive increase in the production of whey. For the dairy sector, it poses a significant disposal challenge. While a portion of whey is used in the food and animal feed industries and converted into useful products. consisting of water (93–94%), lactose (4–5%, w/v), lipids (0.4–0.5%, w/v), soluble proteins (0.6–0.8%), and mineral salts (0.8–1.0%, w/v) in most cases. Whey waste on agricultural land alters the physicochemical properties of the soil, reducing crop yield and raising major pollution concerns for the surrounding ecosystem. This is a result of the whey and milk proteins' fast breakdown, which sucks up oxygen in the soil.

(Kumari et al., 2020) examined whey components have several great functional qualities, including solubility, foaming, emulsifying, gelling, and water binding, in addition to their nutritional and medicinal benefits. The components of whey are essential to human nutrition. Whey has a good nutritional profile; therefore it can be utilized in drinks, foods for the elderly, and sports nutrition.

### **2.2.1Types of whey**

(Silviya R. et al 2016) studied there are two types of whey: sweet and sour. Rennet, an industrial casein-clotting preparation that contains chymosin or another casein coagulating enzyme, is used to coagulate the casein in whey. When casein is coagulated by rennet at a pH of about 6.5, the resulting whey is known as sweet whey. The production of fresh cheese and most industrial casein uses fermentation or the addition of organic or mineral acids to coagulate the casein, resulting in the second form of whey, known as acid whey (pH < 5). After water, lactose (about 70–72% of the total solids), whey proteins (about 8–10%), and minerals (about 12–15%) make up the majority of both sweet and acid whey. The composition of the whey protein fraction, acidity, and mineral content are the primary distinctions between the two forms of whey.

Components	Sweet whey	Acid whey	Chhana/paneer whey
Total solids (%)	7.0	7.0	6.5
Fat (%)	0.3	0.1	0.5
Protein (%)	0.9	1.0	0.4
Lactose (%)	4.9	5.1	5.0
Ash (%)	0.6	0.7	0.6

Typical composition of whey

## 2.3 Carrageenan

(Dipali et al., 2019) examined gel is a solid substance that resembles jelly and can range in texture from weak and soft to strong and rigid. Gels are described as non-fluid polymer or colloidal networks that have had a fluid expand throughout their whole volume. Carrageenan or carrageenin are a class of polysaccharides that are extracted from different red seaweed species. These are red algal cell wall mucopolysaccharides that thicken.

(Sarah et al., 2019) examined Iota ( $\iota$ ), kappa ( $\kappa$ ), and lambda ( $\lambda$ ) are the three primary extracted carrageenan kinds that are significant. In order to offer textural functioning, carrageenan is widely utilized, mostly for gelling and viscosity in a several uses in food kappa carrageenan is extracted from species of Kappaphycus, primarily *K. alvarezii* but also *K. striatum*. Whereas iota carrageenan is taken from *Eucheuma denticulatum*, lambda carrageenan is primary taken from *chondrus crispus*. Refined kappa carrageenan, has excellent clarity and gel strength. Carrageenan lambda does not have gelling property. It is more frequently utilized because of its ability to thicken and became a creamy texture as opposed to a gel-like consistency. Iota carrageenan refined from *Eucheuma* has a high degree of clarity and purity. Its primary use is in expensive goods like toothpaste.

(Lucia et al., 2022) studied Cooling the mixture of kappa carrageenan causes it to gel. Carrageenan has a melting point of roughly 70 degrees Celsius, a gelling temperature of 40–45 degrees Celsius, and a gelling concentration of 60–65 degrees Celsius, respectively. The lowest gelling concentration is found at 0.7 degrees Celsius. The characteristics of chewiness, gumminess, cohesiveness, and hardness were analysis in terms of texture. Jelly candy's texture was primarily affected by the use of gelling agents. The strength of the gel is determined by gumminess and hardness.

(Ana et al., 2019) Vegetable jelly is made from long-chain polysaccharides called carrageenans that are derived from algae and have the capacity to gel. One of the most common texturizing agents used in the food business is carrageenan. These are all-natural substances that have been safely used in food applications for decades. A significant portion of the carrageenan used in food goods, including frozen desserts, chocolate milk, cottage cheese, and whipped cream, comes from the dairy industry. As a result of the interaction between carrageenan subtypes (K and L), successive studies and experiments on colloids produced gels with an ideal texture that emerged as 100% vegetable origin jelly with a quicker solidification rate.

## **2.4 Spirulina**

(Shafai et al., 2023) reported Spirulina is blue-green algae that is edible to humans. The three species that make up this biomass are *Arthrospira platensis*, *A. fusiformis*, and *A. maxima*. It is high in protein and folic acid, which helps the growing brain grow and stay hydrated. Spirulina is a great source of protein because it contains between 55 and 70 percent protein by dry weight. Since spirulina protein has all of the essential amino acids in the proper amount, it is thought to be of the highest standard. A range of carbohydrates, including glucose, fructose, sucrose, and starch, are found in spirulina. Spirulina has a modest carbohydrate content; about 15-20% of its dry weight. Lipids make up 5–10% of the dry weight of spirulina. This is a very modest proportion. Numerous studies have demonstrated that children's meals frequently lack important vitamins, minerals, and nutrients. Inadequate nourishment might result in several health issues, including reduced immunity, delayed cognitive development, under growth. Spirulina's lipid composition consists of a variety of fatty acids, such as omega-3 and omega-6 fatty acids. The Recommended Dietary Allowances (RDA), according to FDA, for minerals vary depending on age, gender, and other factors such as pregnancy or lactation. Here are the estimated needs of some important minerals for the age group of 4-18 years according to the RDA:

- Calcium: The RDA for calcium for children aged 4-8 years is 1000 mg/day, and for children aged 9- 18 years, it is 1300 mg/day.
- Iron: The RDA for iron for children aged 4-8 years is 10 mg/day, and for children aged 9-13 years, it is 8 mg/day. For boys aged 14-18 years, the RDA is 11 mg/day, and for girls aged 14-18 years, it is 15 mg/day.



- Zinc: The RDA for zinc for children aged 4-8 years is 5 mg/day, and for children aged 9-13 years, it is 8 mg/day. For boys aged 14-18 years, the RDA is 11 mg/day, and for girls aged 14-18 years, it is 9 mg/day.
- Magnesium: The RDA for magnesium for children aged 4-8 years is 130 mg/day, and for children aged 9-13 years, it is 240 mg/day. For boys aged 14-18 years, the RDA is 410 mg/day, and for girls aged 14-18 years, it is 360 mg/day.
- Phosphorus: The RDA for phosphorus for children aged 4-8 years is 500 mg/day, and for children aged 9-18 years, it is 1250 mg/day.

(Amir Ali et al., 2021) found *Spirulina* is a cyanobacteria genus that has achieved the ability to utilize carbon dioxide dissolved in seawater as a nutrient source for their reproduction. *Spirulina*, a genus within the Oscillatoriaceae family, comprises filamentous cyanobacteria that are distinguished by linear cell chains encased in a slender filament. It's a photosynthesizing cyanophyte that thrives in intense sunlight, warm temperatures, and an extremely alkaline environment. *Spirulina* have gained considerable recognition in the health food market and are increasingly being used as a protein and vitamin supplement in aquaculture diets. It is easily harvested and processed, grows in both fresh and salt water, and has very high macro- and micronutrient levels. It has been made industrially for human consumption and distributed as a protein supplement in many Asian nations. Pregnant ladies have a common belief that consuming dihé cakes will protect their unborn child from magicians' gaze. It is the best dietary source for malnutrition and a superfood. It strengthens the immune system, decreases cholesterol, inhibits the buildup of fat in the liver, stops tumors from growing, and safeguards the kidneys. It is well recognized that *S. platensis* is a great source of proteins, carbs, vital fatty acids, minerals, vitamins, calcium, potassium, and carotenes as well as chlorophyll and phytocyanin. As a result, *Spirulina* may be used as a daily nutrient supplement or as medication for illnesses. It contains beta-carotene, gamma-linolenic acid, vitamin B, trace minerals, and all of the essential amino acids. It has 3100% more beta carotene than carrots, 670% more protein than tofu, 5100% more iron than spinach, and 180% more calcium than milk.

(G.Usharani et al., 2012) examined pH range of *Spirulina* upto 9 to 11, where it grows more ideally, there is minimal possibility of other germ contamination. *Spirulina* can be beneficial for energy conservation, wastewater recycling, human and animal nutritional and environmental protection. as an additional source of protein in the diets of underprivileged,

malnourished children in underdeveloped nations. A kilogram of mixed veggies is equal to one gram of spirulina protein. Moreover soy beans, spirulina protein has one of the greatest amino acid compositions of any plant. All of the necessary nutrients are provided by spirulina without adding extra calories or fat. Controlling obesity and premenstrual stress is advised. Athletes use spirulina to get quick energy. Numerous herbal cosmetics, such as hair lotions and biolipstics for the face, have been created using the phycocyanin pigment present in spirulina. The prevention of cancer in humans and the improvement of egg, meat, and ornamental fish colors pigmentation are two potential benefits of beta carotene and other carotenoids. And well-known source of beneficial dietary supplements, including proteins, vitamins, minerals, amino acids, and fatty acids, is spirulina. It is extensively utilized in the cosmetics sector as well as in the nourishment of humans and animals

(Tri Winarni et al.,2016) examined contain chlorophyll and carotenoids in addition to phenolic and flavonoid components, which can function as natural substances. However, several of the bioactive compounds found in *S. platensis* are heat sensitive. Additionally, possible phenolic and flavonoid components are present in *S. platensis* powder. Beta-Carotene, which is the simple forms of carotenoid found in *S. platensis*, is an antioxidant that the body can use to counteract free radicals. This finding indicated that the optimal concentrations of *S. platensis* for soft cheese and ice cream, respectively, were found to be 1% and 1.2% additions. Protein, water, fat,  $\beta$ -carotene, and texture (soft cheese) as well as protein, total solid, fat, total sugar, overrun, melting point, and sensory ice cream were all significantly impacted by the addition of *S. platensis*.

(Burcu Ak et al., 2016) studied dairy products have spirulina added for human nutrition due to its extensive nutritional benefits. sensory evaluation results for the bread enhanced with spirulina were deemed acceptable. Additionally, it was found that bread containing spirulina and kept at room temperature effectively inhibited the growth of mold. Bread protein content and improve its calcium, iron, and magnesium content, this study will add spirulina to bread, which is a staple diet for those with low incomes. Addition of 10% of Spirulina was mixed to the flour to make bread. The moisture, ash, fat, protein, and carbohydrate content of samples collected from the breads were examined. Hence the results, this study shows that adding 10% spirulina has no negative impact on product durability loaves and improved nutritional value.

(Amany M. Basuny, et al., 2023) research show that adding 0.5%, 1%, or 1.5% of Spirulina to yogurt drinks enhanced their nutritious value without compromising customer

approval. The sensory aspects of color, look, flavor, and texture of yogurt drinks with 1% spirulina were graded similarly to controls. Numerous bioactive compounds, some of which are heat sensitive, are present in spirulina. The characteristics of this spirulina vary according to whether it is fresh or processed. Nowadays, dairy products have spirulina added for human nutrition due to its extensive nutritional benefits. Fortification of these items becomes efficient it will minimize the number of diseases caused by food insufficiencies. Dry powder is the only form of spirulina that is currently used by industry. Spirulina can ensure both the alkaline balance of the body and the integrity of the meal. The chemical composition of spirulina offers many health benefits. Many diseases such as kidney failure, cancer and high blood pressure are slowed down.

Ingredients (%)	Spirulina algae
Moisture	4.75±0.05
Protein	62.00±0.08
Fat	8.50±0.07
Fiber	6.49±0.05
Ash	11.40±0.03
Carbohydrates	13.80±0.07

% of Nutritional value of spirulina

(Vinay Kumar Pandey et al., 2023) studied research is to create a vegan snack product that is high in protein, low in fat, and high fiber which strengthen the immune system. Spirulina can be added to snack item to provide plant-based protein source, which makes them appropriate for vegans seeking high-protein snack. Also, contain antioxidant and anti-inflammatory qualities, it is a very beneficial product. Protein denaturation typically happens after frying. However, when spirulina powder is combined with gram flour and finger millet flour, the protein denaturation ratio increases. In addition, the observation indicated that the product would benefit from 90 days of storage for improved sensory qualities.

(Ashoush et al.,2019) studied Cookies were made using wheat flour that had been partially replaced with powdered moringa leaves and dried spirulina biomass. All of the nutrients in the cookies, including ash, protein, lipids, crude fiber, and minerals, were considerably boosted when part of the wheat flour was replaced with blends of spirulina and moringa at varying concentrations. The purpose of this study was to improve the baked cookies by including varying amounts of spirulina and moringa powder, which is thought to be a good source of phytochemical components, health-promoting bioactive compounds, and free radical scavenging activity. It will boost the nutritional and antioxidant qualities of cookies by adding 6% dried spirulina biomass and 9% powdered moringa leaves to the cookie. The fiber levels in cookie samples increased to 5.73, 6.54, and 7.65%, respectively, in 5, 10, and 15% replacement level when moringa and spirulina, two fiber-rich substances (7.68 and 4.92%), were added to the cookie formula. The importance of promoting health has led to a large increase in the production of foods rich in fiber. That's why it was discovered that the cookies enriched with spirulina and moringa powder had a higher nutritional profile than the control group.

(Rajaa et al., 2019) examined algae have been added to animal feed, utilized as food supplements to increase their nutritional content, and even for medicinal purposes. They were sold in a variety of forms, mostly as powder, tablets, straws, capsules, and liquids, but they also have a variety of chemical qualities and can be added to other meals like pasta, gums, and drinks. Although microalgae contain many types of flavonoids and other phenolic classes, polyphenolic chemicals are recognized as significant natural antioxidants. Also connected to bio pharmacological properties were a variety of polyphenolic compounds, which include antioxidant and antibacterial properties

#### **2.4.1 Health benefits**

Consumption of spirulina with combination of dairy products are associated with numerous health benefits

(Kumari et al., 2011) Research on the therapeutic effects of this substance on a variety of disorders, such as cancer, viral infections, cardiovascular diseases, inflammatory diseases, hypercholesterolemia, and hyperglucagonemia, has been intensely focused on recently. Spirulina's anti-inflammatory, hypolipidemic, and antioxidant properties are largely responsible for its cardiovascular health benefits. The nutritional properties of spirulina are genuinely unique. Spirulina is the highest known natural source of protein, with over 71 percent of its structure made upon protein. Its protein content is five times higher than that of meat and

almost three times higher than that of the widely consumed soybean. Other important minerals found in spirulina include carotenoids, B complex vitamins, vitamin E, copper, manganese, magnesium, iron, selenium, and zinc. Because it has been used as food for a long time and has shown a good safety profile in research involving animals, spirulina is regarded as safe for human ingestion.

## 2.5 Ginger

(Aamir et al., 2023) found that Zingiberaceae family, ginger (*Zingiber officinalis* Roscoe) is one of the herbs and spices that are used in a wide range of foods and drinks all over the world. It includes a large number of physiologically active chemicals that have positive effects on health, it is also recognized to have medicinal qualities. Good antibacterial effects against both gram-negative and gram-positive bacteria have been reported for ginger. Through the stimulation of physiological processes, it may aid in the regulation of blood pressure in humans.

(Stoilova et al., 2006) examined that more than 2,000 years, ginger has been used as a spice. Its roots and the extracted materials include polyphenol compounds with strong antioxidant properties. Even though this spice has a stimulating effect on digestion. A significant factor in the intake of fats is bile acids. Ginger can be extracted in a number of ways, however the CO<sub>2</sub> extracts are the most concentrated in polyphenol chemicals and have the most similar composition to the roots. The other important component that is essential to the digestion of fat is lipase. The amount of pancreatic and intestinal lipase was shown to significantly rise when ginger was added to animal diets. The purpose of the study is to evaluate the potential of ginger extract as a natural preservative that may be used in the food and pharmaceutical industries by comparing it with synthetic antioxidants.

(Rika Sepriani et al., 2021) reported based on the size, form, and color of the rhizome, ginger is categorized into three types: red ginger, elephant ginger, and emprit ginger. Red ginger is the smallest, red to orange colour, ginger with a strong aroma and coarse. Medium-sized, white or yellow, slightly flattened, fibrous, and not very harsh ginger is called little (emprit) ginger. Elephant ginger is golden or light yellow in color, soft, low in fiber, and has a subtle scent, despite having the greatest rhizome size. 8.6% protein, 6.4% fat, 6.9% fiber, 66.5 percent carbs, 5.7 percent ash, 0.1 percent calcium, 0.15 percent phosphorus, 0.03 percent sodium, potassium, 1.4 percent, 175 of vitamin A per 100 grams, 0.05 mg of vitamin B per 100 grams, 0.13 mg of vitamin B2 per 100 grams, and 12 mg of vitamin C per 100 grams are found in

ginger. In addition to vitamin E, ginger contains antioxidants in the forms of gingerols, shogaol, and gingerone. Based on the study's findings, water infused with red ginger and soaked for 12 hours had the highest level of antioxidant activity. Even after soaking for 6 hours, the elephant ginger exhibited the least amount of antioxidant activity. Small ginger (emprit) was discovered to have the highest levels of vitamin C infused water after soaking for 12 hours.

(Giacosa et al., 2015) found that addition to being a widely used spice, ginger has long been used as a medicine to treat fever, nausea, vomiting, and dyspepsia. It has recently been possible to identify some of the chemicals causing this activity as well as their mechanisms of action. 6-The presence of alpha, beta-unsaturated ketone moiety is responsible for the most powerful antioxidant and anti-inflammatory actions. essential oil of ginger (0.25-3.3% V/m)<sup>2-5</sup> contains monoterpenes– mainly geranial (citral a) and neral (citral b)– and sesquiterpenes (30.7%) mainly Beta-sesquiphellandrene, Beta-bisabolene, ar-curcumen and alpha-zingiberene. Additionally, strong principles (4.7–5% w/w) like shogaols, gingerols, and similar derivatives of phenolic ketone are present. Diterpenes, 6-ginger sulphonic acid, diarylethene's, and monoacyldigalactosyl glycerol's are some of the other constituents.

(Adrija et al., 2018) studied Ginger is a popular remedy for a number of "stomach problems," such as motion sickness, morning sickness, colic, upset stomach, gas, diarrhoea, nausea after cancer therapy, nausea and vomiting following surgery, and appetite loss. Ginger has been shown in studies to have proteolytic activity. As a result, it is frequently employed as one of the most common proteases to tenderize meat since it acts more on collagen than actomyosin, and the combined action makes the flesh tender.

(Wenhui Si et al., 2018) reported ginger extract has strong anti-inflammatory, anti-cancer, and antibacterial qualities, it is also frequently used as a herbal remedy. Ginger's health-promoting properties are usually linked to its abundance of phytochemicals, which have been shown to stop rats' liver tissue from apoptosing when exposed to lead. decrease aortic atherosclerotic plaque, total plasma cholesterol, and liver cholesterol in hamster models and stop mice from becoming obese due to a high-fat diet.

Constituent	Value
Moisture	15.02 ± 0.04
Protein (g)	5.087 ± 0.09 (5.98)
Fat (g)	3.72 ± 0.03 (4.37)
Insoluble fibre (%)	23.5 ± 0.04 (30.0)
Soluble fibre(%)	25.5 ± 0.04 (30.0)
Carbohydrate (g)	38.35 ± 0.1
Ash (g)	3.85 ± 0.61 (4.53)
Vitamin C (mg)	9.33 ± 0.08 (10.97)

Nutritional composition of ginger (per 100 g) (Mohamad et al., 2019)

## 2.5.1 Health benefit

### 2.5.1.1 Antioxidant property

Antioxidants are substances that bind these active compounds with their own active sites, eliminating them and lowering the risk of various health issues. Plant-based diets are regarded as a rich source of antioxidants in this scene. Including ginger in your diet strengthens your body's defenses. Numerous in vitro studies have demonstrated the link between oxidative stress and a number of chronic diseases. Because its chemical components have a high level of antioxidant activity, ginger can help avoid these illnesses. Gingerols are among the active ingredients in ginger and are thought to be the most potent antioxidant in nutraceuticals.

### 2.5.1.2 Gingers Hyperglycaemic/ Anti-Diabetic Effects

Numerous lifestyle-related illnesses, including blood pressure, obesity, cardiovascular disease, and many more, are caused by dietary changes and insufficient physical activity. Ginger has been the subject of numerous research in recent years, demonstrating its safety as a herbal remedy and its usefulness in preventing diabetes. Research has shown that giving rabbits oral ethanolic ginger extract considerably lowers their blood glucose levels. To investigate the potential of ginger extract to lower cholesterol. The haematological studies showed that the level of fasting blood glucose is subsequently lowered by ginger extract. The amount of 6-gingerol in ginger extract determines its anti-diabetic properties. The haematological studies showed that the level of fasting blood glucose is subsequently lowered by ginger extract. The amount of 6-gingerol in ginger extract determines its anti-diabetic

properties. Throughout the duration of the treatment, the diabetic rats given ginger maintained their starting weights and drank less water. Consequently, ginger may be useful in helping human subjects manage the consequences of diabetes problems (Muhammad Shoaib et al., 2016)

## 2.6 Mint

(Fischer, B et al., 2020) found mint referred to as "hortelã" in Brazil, is used as a natural seasoning, tea flavouring, and culinary flavouring. Certain mint species may have strong antioxidant and antibacterial capabilities, according to some research. Peppermint is a strong, pleasantly peppery flavour that can be found in teas, chewing gum, and culinary goods including drinks. In order to find an alternative use for whey and to innovate functional products for small-, medium-, and large-sized dairy companies, this work aimed to develop and characterize a functional milk-based beverage with coffee flavour and mint extract added, utilizing readily available industry equipment in a straightforward process. The phenolic total content ( $R = -0.979$ ) and flavonoid content ( $R = -0.972$ ) showed a strong association with the EC<sub>50</sub> of the extracts in this investigation, suggesting a direct relationship between the extract obtained and its antioxidant capacity. The concentration, affinity, and selectivity of the solvent all affect the compounds' ability to be extracted with antioxidant quality

(Padmini et al., 2008) current studies compares and evaluates the phytochemical composition and antioxidant activity of the aqueous extracts of black tea, mint, and black tea enhanced with mint. The mint plant, *Mentha spicata*, produces leaves that are aromatic, warm, fresh, sweet, and have a refreshing aftertaste. It has diaphoretic, stomachic, stimulant, carminative, and antispasmodic properties. Mint extract has long been used as a digestive aid and a treatment for severe colds. Similarly, it has been suggested that the components of black tea, such as polyphenols, flavones, and gallate esters, have antioxidant properties and lower the risk of heart disease, cancer, and hypertension. Additionally, the study suggests that the combined effects of the tea and mint extracts might not always be cumulative. However, increasing the intake of tea, tea supplemented with mint, or tea itself may help to boost antioxidant defence and prevent the number of degenerative diseases brought on by oxidative stress Living a healthy life.

(Mahmoud et al., 2023) examined concentrates of bioactive phytochemicals derived using extraction techniques such as solvent extraction, steam distillation, or cold pressing with or without pretreatment are known as plant extracts. Depending on the species and variation,



essential oils can also be extracted from different plant sections. The goal of extraction is to extract as much of the plant's bioactive components as possible. Plant metabolites that are soluble are separated from their insoluble components with the aid of solvents. food can incorporate plant extracts at low amounts that are uniformly distributed and effective. Encapsulation shields the components from light, heat, moisture, and oxidation while enabling scientists to regulate the release of the substances and cover up offensive smells.

(G Larionov et al., 2020) examined to develop zero waste and low waste technology for private and collective farms and agricultural holdings and as a result, provide the solution to whey disposal issue. One of the promising areas for the use of whey is the production of beverages. Herbal ingredients are used to increase the nutritional value of whey drinks. Peppermint is a valuable raw material for the production of mint-whey drink. The results showed that adding three grams of peppermint to one liter of whey had a major impact on the drink's organoleptic qualities. A whey drink containing 1 g of peppermint was chosen based on the organoleptic evaluation results. The beverage was kept for a period of twelve days at  $4\pm 2^{\circ}\text{C}$  in the refrigerator. The use of peppermint syrup in whey drink production technique is what makes this work novel. The goal of the study is to employ peppermint syrup to produce whey drink in individual homes, communal farms, and agricultural holdings.

(Asmaa A. E et al., 2023) found Mint leaves contain bioactive compounds that are used in the food sector. Mint is hence a good choice for a range of culinary dishes. Because of its therapeutic benefits and health claims. It has a long history of usage, both therapeutically and as a food preservative. It is a significant source of polyphenolic chemicals such as flavonoids, hydroxycinnamic acids, and hydroxybenzoic acids. A rising number of people, they interested to obtaining bioactive chemicals from natural sources, like spices, herbs, and medicinal plants, because of the risks to one's health and the limitations associated with using synthetic additives.

Chemical composition	Mint leaves (g/100)
Ash	18.6
Total lipids	4.9
Crude protein	19.8
Total carbohydrates	55.7

Chemical composition of mint leaves per (100g) (Sakr et.,al 2019)

## 2.6.1 Health benefit

### 2.6.1.1 Antibacterial and antifungal effects

Both menthol or peppermint can have anti-bacterial and anti-fungal properties against a variety of common species. Typically, menthol or peppermint are used as crude extracts or essential oils; however, some research has looked at the specific constituent chemicals. Studies can also make a distinction between different plant components. Although leaves are frequently used, some groups use methodologies that involve the entire plant or only the leaves and stems.

### 2.6.1.2 Nociception, Migraine and Headache

Pain is defined as the interpretation of a nociceptive input, that may result in perceived or actual tissue damage, or the experience of such damage. Both menthol and peppermint have been shown to have analgesic effects, particularly when given after resistance exercise or other activities that intentionally cause tissue injury, or in clinical situations like pain associated with arthritis or neuropathy. Mechanisms linked to a decrease in pain could include vasoconstriction of peripheral blood vessels and decreases in arterial blood flow. These elements are especially crucial to take into account when thinking about possible migraine applications. It is believed that there is a hereditary component to migraine that may be exhibited partially through TRPM8 receptors.

### 2.6.1.3 Respiratory health

Applying menthol and peppermint to the respiratory system also smoothe muscles and relaxes them. Application through respiration, inhalation, or ingestion causes feelings of greater nasal flow inside the upper respiratory system, however this hasn't been demonstrated to be physically changed. Due to these methods, using menthol and peppermint during exercise also increases airflow and may have additional positive effects during activity. These are

covered in the sections that follow under the headings of athletic performance and thermal comfort and feeling.

## **2.7 Other Human Benefits**

### **2.7.1 Taste of sensitivity**

Products containing menthol and peppermint can nevertheless cause sensations in people with anosmia and ageusia, most likely by way of chemesthetic and trigeminal pathways. However, a few subjects in related studies have mentioned sustained loss of olfactory sensitivity. Because of their extra-oral connections to mucosal immunity, polymorphisms in bitter taste receptors have also been identified as potential candidates that may be relevant in relation to COVID mortality. Age also affects or impairs this system, even if menthol or peppermint sensitivity may be a proxy for infection and subsequent healing in persons with low degrees of infection.

### **2.7.2 Alertness**

Research has shown that the odors of peppermint and menthol both have an impact on reaction times and cognitive functions, although these effects and the perceived qualities of these odors seem to diminish with repeated exposure. This suggests that either menthol-containing stimuli might cause quick habituation or that repeated effects need to occur at a significant interval. Similarly, both healthy participants and those with upper respiratory tract infections find that eating peppermint/menthol gum increases alertness. Significant changes in oxygenated and deoxygenated hemoglobin were noted when military troops using menthol as a mouthwash experienced increases in brain metabolism while completing cognitive tasks in the heat (Russ best 2022)

## **2.8 Citric acid**

(Angumeenal et al., 2012) studied Citrus and pineapple juices include citric acid, a tricarboxylic acid ( $C_6H_8O_7 \cdot H_2O$ ), which is a common metabolite of both plants and animals. Pure citric acid has a molecular weight of 210.14 g/mol, is colorless, and dissolves easily in water. It is a cheap, safe, environmentally friendly, biodegradable, and multipurpose chemical for cleaning, dispersing, buffering, wetting, and sequestering. The acid is mostly used to make confections, soft drinks, effervescent salts, and medicinal citrates. Renewable and less priced substrates can be used to manufacture citric acid.

(Rejeb et al., 2020) current study involved the development of reduced sugar jelly items employing citrus, a fruit high in antioxidants. Citrus juices' effects on the antioxidant qualities, color, consistency, and sensory assessment of the jelly were also studied. Producing citrus fruit-derived goods like juice, jam, jelly, and marmalade extends the shelf life of fruits and makes them more widely available to consumers throughout the year. The primary ingredient needed to make fruit jellies is sugar. It provides good sweetener and is crucial to the gelation process and the achievement of the required texture. Every citrus jelly had a considerable amount of phenolic compounds and was a great source of phytochemicals with a host of health benefits.

### **3.MATERIALS AND METHODS**

#### **3.1 Materials**

**3.1.1** Pasteurised milk used for the preparation of paneer whey.

#### **3.1.2 Paneer whey**

Paneer was prepared using standardized pasteurized milk (4.5 per cent fat and 8.5 per cent milk solid not fat).

#### **3.1.3 Sugar**

Commercially available Fine crystalline sugar (Madhur brand) obtained from the local market was used in the preparation of jelly.

#### **3.1.4 Kappa Carrageenan**

This is purchased from online brand name ‘‘bake king’’and it is used as gelling agent in jelly preparation.

#### **3.1.5 Spirulina**

Most of the biomass of Spirulina being produced today is consumed as a nutritional supplement promoted as a ‘‘superfood’’ and sold as a dried powder, capsule. Spirulina has been certified as Generally Recognised as Safe (GRAS) by the United States (US). Spirulina was purchased from online brand name grenera spirulina powder.

#### **3.1.6 Citric acid**

The citric acid used in present study was of food grade quality

#### **3.1.7 Ginger**

The ginger was procured from local market, ginger juice extract is used for making herbal jelly

#### **3.1.8 Mint**

The mint was procured from local market, mint juice extract is used for making herbal jelly

#### **3.1.9 Equipment**

The equipment available at Varghese Kurian institute if dairy and food technology were used for the present study.

## **3.2 METHODS OF PREPARATION**

### **3.2.1 Preparation of paneer whey**

Paneer whey is used for the preparation of herbal whey jelly. Paneer whey was prepared using pasteurized milk by the addition of 2% of citric acid coagulation. (Kakan AV et al., 2017) Whey may be defined as broadly watery part of milk remaining after separation of the curd, which results from the coagulation of milk proteins by acid or proteolytic enzymes.

### **3.2.2 Preparation and extraction of ginger juice**

Ginger is procured from local market. It is cleaned and peeled ginger is extracted by mortar and pestle. This juice is filtrated using a sieve. So fresh juice collected in a beaker.

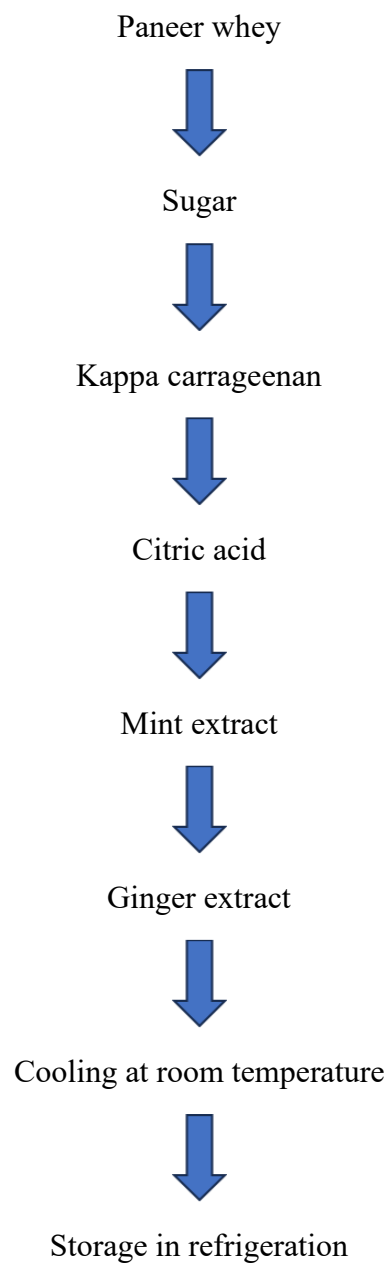
### **3.2.3Preparation of extraction of mint juice**

Mint is procured from local market. Fresh mint properly wash with portable water and juice is extracted by using mortar and pestle. This juice is filtrated using sieve. So fresh juice is collected in a beaker.

### **3.2.4Preparation of control jelly**

Control jelly is prepared with paneer whey, sugar, kappa carrageenan (1.5%), citric acid (0.5%). 100ml of paneer whey is boil add 45g of sugar and continuously stir the mixture. Add 1.5% of kappa carrageenan is mixed with 5g of sugar. then add 0.5% of citric acid. Then add 4% mint extract and 2% of ginger extract in the mixture. Then the whole mixture is poured into a bowl for settling. after cooling control jelly is cut into unique shapes. and stored at room temperature

### 3.2.5 Flow chart of control jelly



### 3.2.6 Flow chart of jelly preparation

Paneer whey boil at (60°C to 70°C for 15 to 35 minutes)



Addition of Sugar



Spirulina



Addition of sugar mixed with carrageenan whey jelly



Citric acid



Ginger Extract



Mint Extract



Cooling at room temperature



Storage in refrigeration



### 3.3 Standardization of herbal whey jelly

Jelly was prepared using different ratio of spirulina for the optimization of best combination. Paneer whey and spirulina is mixed in the ratios of 100:0 (control), 100:0.5 (S1), 100:1 (S2), 100:1.5 (S3), 100:2 (S4) then jelly was prepared and subjected to sensory evaluation.

sample	Whey (ml)	Spirulina(g)	Carrageenan(g)	Citric acid(g)	Mint(g)	Ginger(g)
Control	100	nil	1.5	0.5	4	2
S1	100	0.5	1.5	0.5	4	2
S2	100	1	1.5	0.5	4	2
S3	100	1.5	1.5	0.5	4	2
S4	100	2	1.5	0.5	4	2

Control jelly was prepared by adding 100 ml of paneer whey, 45g of powdered sugar, stir continuously at it completely dissolve. Then sprinkle the combination of sugar and carrageenan with level of 5:1.5. Adding 0.5% of citric acid. Add the combination of mint extract and ginger extract at level of 4:2 is added into mixture. This mixture is poured into bowl for setting and stored at refrigeration.

Preparation of jelly made by adding different levels of spirulina to increase the protein content. It was prepared by adding 100ml of paneer whey, 45g of powdered sugar, stir continuously at it completely dissolve. Then add *S. platensis* powder was added at different concentration (0.5%, 1%, 1.5%, 2%) into mixture and stir it. Sprinkle the combination of sugar and carrageenan with level of 5:1.5. Then 0.5% of citric acid. The combination of ginger and mint is added at 2:4% into the mixture. This whole mixture is poured into bowl for setting and stored at refrigeration.

### 3.4 SENSORY EVALUATION OF JELLY

Sensory evaluation is the science related to the senses. Human senses include touch, smell, vision, hearing and taste. These organoleptic characteristics are judged by different organs of our body. In this evaluation product is judged for different sensory attributes such as flavour, color and appearance, body and texture as well as overall acceptability. All the jelly samples prepared in the present study were evaluated for their sensory attributes on Nine Point Hedonic scale by a panel of seven judges.

### 3.5 PROXIMATE COMPOSITION AND PHYSICO CHEMICAL ANALYSIS OF JELLY

#### 3.5.1 Moisture



The moisture content of jelly samples were estimated by procedure given by Ranganna(2012).A flat bottom metallic dish was dried at 110 °C for 1 hr; cooled and weighed. Then 10g jelly was uniformly spread in dish. It was then dried in a hot air oven at atmospheric pressure. Temperature was maintained at 100 °C. After drying the sample was cooled in desiccator and reweighed. Sample was then reheated, and reweighed until the consecutive weighing did not vary by more than 1mg. The per cent moisture in sample was calculated using the formula.

$$\text{Moisture content (\%)} = \frac{\text{loss in weight of sample (g)}}{\text{weight of the sample (g)}} \times 100$$

#### 3.5.2 Total solids

The total solids content includes both the suspended and dissolved salts. The total solid content also used to determine a sludge dry weight expressed as a percentage. The total solid

content is expressed as a ratio of weight obtained before and after drying process. percentage is calculation using formula.

$$\text{Total solids\%} = 100 - \text{moisture\%}$$

### 3.5.3 Total soluble solids (TSS)

TSS of jelly samples were determined using hand refractometer



### 3.5.4 Fat



Two gram of jelly sample was weighed accurately into a dried beaker followed by addition of 2 ml of absolute ethyl alcohol. The content was stirred well followed by addition of 7 ml HCl and 3 ml distilled water. Whole content was heated on water bath maintained at 80 °C for 30 min. The content was cooled and 10 ml ethyl alcohol slowly added and transferred quantitatively into a Mojonnier fat extraction tube. Diethyl ether was added (25 ml) through the beaker. The tube was closed and shaken vigorously for 1 min. Now, 25 ml of petroleum ether was added through the beaker into the fat extraction tube. Again the tube was shaken vigorously for 1 min after stoppering. Now tube was allowed to stand on the flat bottom of the lower bulb until

ethereal layer was cleared and completely separated from the aqueous layer for not less than 30 min. The supernatant layer was carefully decanted into a suitable reweighed dish by gradually bringing the cylindrical bulb of the tube into horizontal position. Extraction and subsequent operation was repeated twice by using 15 ml ether and 15 ml petroleum ether. Flask containing combined ethereal extract was kept on boiling water bath for complete evaporation of the ethereal portion. The flask was then transferred to the oven maintained at  $100 \pm 2^\circ \text{C}$  for about 1 h. ° Cooled in desiccator and weight was recorded. The procedure of heating, cooling and weighing was repeated till the two consecutive weighing do not differ by more than 0.5 mg.

$$\% \text{ Fat content} = \frac{W_2 - W_1}{W_3} \times 100$$

Where,

W1= Weight of empty dish

W2= Weight of dish after evaporation

W3= Weight of sample

### 3.5.5 Protein



Total nitrogen content of jelly samples were determined by macro Kjeldahl method given by Ranganna (2012) with slight modification (using Kelplus digestion system, model-KES 20L VA DLS, M/s. Pelican Instruments, Chennai and Kelplus semiautomatic distillation system model-Distil M, M/s. Pelican Instruments, Chennai). In the digestion tube 3 g of the sample was accurately weighed and then 2.4 g of digestion mixture (potassium sulphate: copper sulphate: selenium dioxide; 1.0: 0.1: 0.1) was added. To this, 25 ml of nitrogen-free

concentrated sulphuric acid was added. The tubes were transferred to the digestion block, where the contents were digested at a temperature of 350 °C until the digest is clear or pale blue for about 30 min. The cooled digested contents were loaded in the Kjehl-plus distillation unit and 60 to 70 ml alkali (50.0 % sodium hydroxide) was added automatically for neutralization. The sample was distilled for 9 min. The liberated ammonia was condensed and collected in 50 ml of saturated boric acid solution, containing mixed indicator (equal volume of saturated solution of methyl red and 0.1 % methylene blue solution). The distillate was titrated against 0.1 N H<sub>2</sub>SO<sub>4</sub>. A reagent blank was simultaneously run. The per cent total Nitrogen was calculated using the formula:

$$\% \text{ Nitrogen} = \frac{(V_1 - V_2) \times \text{normality of H}_2\text{SO}_4 \times 14 \times 100}{W_1 \times 1000}$$

Where,

W= Weight of sample,

V<sub>1</sub> = Volume of 0.1 N H<sub>2</sub>SO<sub>4</sub> used in titrating the sample,

V<sub>2</sub> = Volume of 0.1 N H<sub>2</sub>SO<sub>4</sub> used in blank titration.

The per cent total protein was calculated by multiplying nitrogen with a factor of 6.25.

### 3.5.6 Ash



The Ash content of whey based and control jelly was estimated by procedure described in Ranganna (2012). Fifteen-gram jelly sample was weighted into silica dish (7-8 cm dia). Silica dish was used for ashing. The contents were ignited on a Bunsen Burner. Then material was

ash at not more than 525 C for° 10 h in a muffle furnace. The dishes were cooled in a desiccator and weighed. The total ash content in per cent was calculated as follows:

$$\text{Total ash \%} = \frac{100 \times (W3 - W1)}{W2 - W1}$$

Where,

W1= Weight in gram of the empty crucible

W2= Weight in gram of the crucible with sample

W3= Weight in g of the crucible with ash

### 3.5.7 Total carbohydrate

The carbohydrate content was estimated by difference.

### 3.5.8 Titratable acidity

A known weight of jelly (10 g) was dissolved in distilled water and heated on water bath to dissolve the sample. It was then cooled and made up to a known volume (50 ml). It was titrated against 0.1 N NaOH using few drops of 1% phenolphthalein solution as indicator. Titre value was noted and calculated as per cent anhydrous citric acid (Ranganna, 2012)

$$\% \text{ of titrable acid} = \frac{0.64 \times \text{titre value (ml)}}{\text{weight of sample (g)}} \times 100$$

### 3.5.9 pH



The pH values of jelly samples were determined using a digital pH meter. after mixing the samples with equal quantity of distilled water (Ranganna, 2012).

### 3.5.10 Crude fibre

Transfer qualitatively fat free dry material in sample after either extraction into spout less beaker of 1L capacity. Add 200ml of 1.25% H<sub>2</sub>SO<sub>4</sub>. Half drop of higher alcohol is added to prevent frothing. Keep bulb condenser in position and reflux for exactly half an hour after the mixture in the beaker started boiling. Filter the content of beaker through filter cloth kept in funnel. Wash the residue in cloth with hot water till washing in acid free. Transfer residue quantitatively into same tall beaker. Add 200ml 1.25% NaOH and few drop of octyl alcohol. Boil it half hour for acid extraction. Filter the content and wash with hot distilled water to make it alkali free. Pour little acetone over residue to remove adhering moisture. Transfer the content into silica dish. Dry the content in hot air oven at 100 to 105°C and cool it in dessicator. Record the weight of dish with residue. Note the weight of ash. Incinerate it in muffle furnace at 600-620°C

$$\% \text{ of crude fibre} = \frac{\text{loss of weight residue on incineration}}{\text{weight of sample}} \times 100$$

Where,

W<sub>1</sub>= Weight of bottle

W<sub>2</sub>= Weight of bottle with material

W<sub>3</sub>= Weight of dish with residue after drying

### 3.5.11 Antioxidant

The antioxidant activity was determined using DPPH method.

Take 1g of sample and dissolve in 10ml of methanol. place the solution in water bath for 2hour with continuous stirring. After 2hours, centrifuge the solution in 5000rpm for 20mins. Filter the centrifuge using Whatman filter paper. Take 0.5 ml of sample solution and add 3.5ml methanol and 1ml DPPH reagent. blank is also prepared by adding 1ml DPPH solution and 4ml of ethanol. Incubate both sample and blank at 37°C for 30mins. Analysis both using spectrophotometer at 515nm.

Calculation

$$\text{Antioxidant activity (\%)} = \frac{\text{absorbance of blank} - \text{absorbance of sample}}{\text{ansorbance of blank}} \times 100$$

### 3.6 PHYSICAL PROPERTY

#### 3.6.1 Color characteristics



Color of the herbal whey jelly was measured by reflectance spectroscopy technique employing reflectance meter, color flex (hunter lab colorimeter) along with the universal software. Before the test instrument was calibrated as specified by the manufacturer. The light source was dual beam xenon flash lamp. Data was received from the software in terms of L\*[lightness, ranges 0(black) to 100 (white)], a\*[Redness, ranges from +60(red) to 60 (green) and b\*(yellowness range from +60(yellow) to -60(blue) in value of international color system.



### 3.6.2 Texture Profile Analyzer



Various textural characteristics such as hardness, cohesiveness, springiness, gumminess & chewiness were measured by texture profile analyzer (TA-X T2 texture analyzer - stable microsystem). The equipment was set in the back extrusion mode with the following settings: Pretest speed: 1.00mm/sec, Test speed: 1.00mm/sec, Post test speed: 10mm/sec, Distance: 30mm, Trigger force: 2g. Probe with a diameter of 35 mm was used for the test. The mode was set to measure the force in compression.

### 3.7 SENSORY EVALUATION

The sample was evaluated on a 9-point hedonic scale, for their sensory attributes of flavour, color and appearance, body and texture, sweetness and overall acceptability.

Liked extremely	-9
Liked very much	-8
Liked moderately	-7
Liked slightly	-6
Neither liked nor disliked	-5
Disliked slightly	-4

Dislike moderately -3

Dislike very much -2

Dislike extremely -1

## **SCORE CARD FOR SENSORY EVALUATION OF HERBAL WHEY JELLY**

Name of judge:

Date:

<b>ATTRIBUTES</b>					
<b>flavour</b>					
<b>color</b>					
<b>Body&amp;texture</b>					
<b>Overall acceptability</b>					

Remarks:

signature:

## 4.RESULT AND DISCUSSION

In this study an attempt was carried out for the standardisation of a procedure for the preparation of herbal whey jelly incorporated with spirulina, herbs and spices also, to identify the effects of spirulina in physico-chemical properties of herbal jelly. Herbal whey jelly is prepared by addition of sugar, spirulina,  $\kappa$ -carrageenan, citric acid, ginger and mint into the paneer whey. It is found to provide good nutritive value, texture and aroma. The spirulina is added in different levels (viz., 0.5%, 1%, 1.5%, 2%) to herbal whey jelly and treatment control, T2, T3, T4 and T5 respectively. It is selected on basis of preliminary trials. The optimized product was evaluated for its physico-chemical analysis, textural and colour attributes. Herbal whey jelly with no added spirulina is taken as control. So, addition of spirulina with different ratio will increase protein content in herbal whey jelly.

### 4.1 Effect of levels of Ginger Extract

Addition of ginger to observe sensory attributes like flavour, colour and appearance, addition of 2% of ginger enhances good flavour and aroma. Also, it will mask the flavour of spirulina. So that after sensory evaluation 2% of ginger is fixed as constant.

### 4.2 Effect of levels of Mint Extract

Addition of mint at different levels to observe sensory attributes like flavour, colour and appearance. There are three levels are taken for the selection of mint 2%, 3%, 4%. After sensory evaluation. Addition of 4% of mint extract enhances good flavour and aroma so that this is fixed as constant.

samples	T1	T2	T3
Whey	100ml	100ml	100ml
Sugar	50g	50g	50g
$\kappa$ carrageenan	1.5g	1.5g	1.5g
Ginger	2g	2g	2g
Mint	2g	3g	4g
Citric acid	0.5g	0.5g	0.5g

### 4.3 Effects of levels of spirulina powder

Addition of spirulina will improve the protein content in herbal whey jelly. So addition of different levels of spirulina powder. Spirulina are prokaryotic or eukaryotic photosynthetic microorganisms that use the process of photosynthesis to create lipids, proteins, and carbohydrates. Grown microalgae are a good source of antioxidants, pigments, polyunsaturated fatty acids, and biologically active substances. In herbal whey jelly, addition of spirulina at different levels such as, 0.5%, 1%, 1.5%, 2%. After sensory evaluation 1.5% of spirulina enhance good acceptability. After physico-chemical analysis small amount of protein content and antioxidant content is increasing.

sample	Whey (ml)	Spirulina(g)	Carrageenan(g)	Citric acid(g)	Mint(g)	Ginger(g)
Control	100	nil	1.5	0.5	4	2
T2	100	0.5	1.5	0.5	4	2
T3	100	1	1.5	0.5	4	2
T4	100	1.5	1.5	0.5	4	2
T5	100	2	1.5	0.5	4	2

### 4.5 Sensory Evaluation

The organoleptic quality of product was determined with the help of a panel of judges using 9-point hedonic scale. The results are depicted in Table 1 below

**Table 1: sensory score of herbal whey jelly control prepared by the addition of ginger and mint extract at different levels.**

	Attributes			
samples	flavour	Body & texture	Colour & appearance	Overall acceptability
S1	7.68±0.38	7.76±0.55	7.72±0.31	7.70±0.26
S2	7.96±0.25	8±0.37	7.72±0.19	7.95±0.40
S3	8.1±0.36	8.25±8.25	8.24±0.20	8.16±0.28

**Table 2: Sensory scores of herbal whey jelly prepared by the addition of spirulina at different levels**

	Attributes			
Samples	flavor	Body & Texture	Color & appearance	Over acceptability
control	8.3±0.34	8.2±0.2	8.26±0.30	8.4±0.17
S2	7.73±0.30	8.13±0.23	8.36±0.32	8.16±0.2
S3	7.66±0.05	7.76±0.20	8.06±0.49	7.96±0.11
S4	7.5±0.45	7.96±0.40	7.93±0.46	7.76±0.49
S5	6.7±0.1	6.4±0.2	7±0.17	6.83±0.40

Where,

Control- Have no addition of spirulina. Here combination of ginger and mint at the ratio of 2:4%

S1-Addition of spirulina at the ratio of 0.5%. Here combination of ginger and mint at the ratio of 2:4%

S2- Addition of spirulina at the ratio of 1%. Here combination of ginger and mint at the ratio of 2:4%

S3- Addition of spirulina at the ratio of 1.5%. here combination of ginger and mint at the ratio of 2:4%

S4- Addition of spirulina at the ratio of 2%. Here combination of ginger and mint at the ratio of 2:4%

**Flavour:** - Control had good flavour and aroma of ginger and mint extract. Here T3 imparts a good flavour and aroma while adding spirulina to the product. Ginger and mint extract contain volatile components it imparts good flavour and aroma.

**Colour and appearance:** - The mean sensory scores for colour and flavour of herbal whey jelly. The mean highest sensory score T3(having 2:4% of ginger and mint extracts). A good

quality jelly had uniform green colour and uniform distribution of spirulina made by product of whey respectively. Other have intense green colour.

**Body and texture:** - Body and texture of jelly will losses by increasing the concentration of spirulina. Because of binding property of  $\kappa$  carrageenan will decrease by increasing spirulina content. So highest sensory score is T3.

**Overall acceptability:** - It is based on multiple organoleptic quality parameters like colour, flavour, texture shows accumulative perception and acceptance by the panelists. So based on that maximum score was awarded for T3. Such that the product with 1.5% spirulina, 2:4% ratio of ginger and mint extract was selected as the best product. Similar study about effect of spirulina.



## 4.6 Physico-chemical Analysis

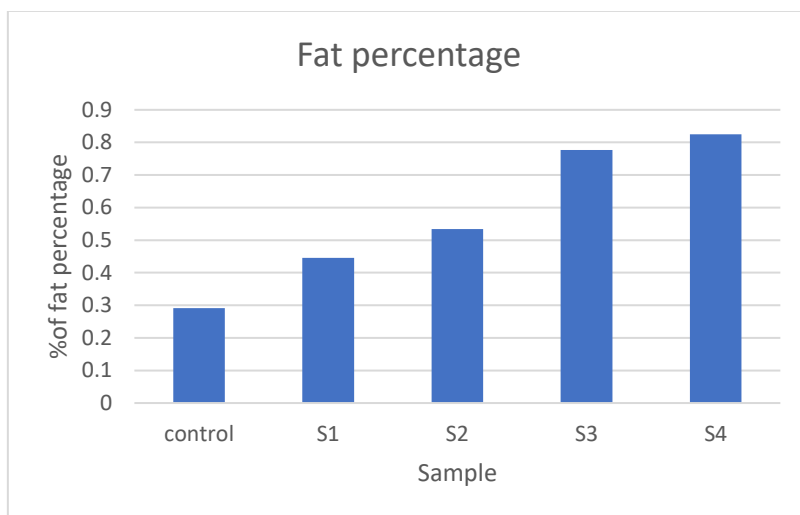
The optimized product was taken for further analysis along with the control.

### 4.6.1 Proximate composition values of Herbal whey jelly and control

Proximate analysis	control	S1	S2	S3	S4
MOISTURE	18.166189%	18.82813%	19.30675%	20.40094%	22.45069%
ASH	0.54%	0.62%	0.65%	0.66%	0.71%
FAT	0.291262%	0.445545%	0.533981%	0.776699%	0.825243%
PROTEIN	0.52537%	1.401%	1.57612%	1.75125%	2.1015%
TITRABLE ACIDITY	0.32%	0.32%	0.32%	0.32%	0.32%
pH	3.96%	3.96%	3.96%	3.96%	3.96%

#### 4.6.2 Fat

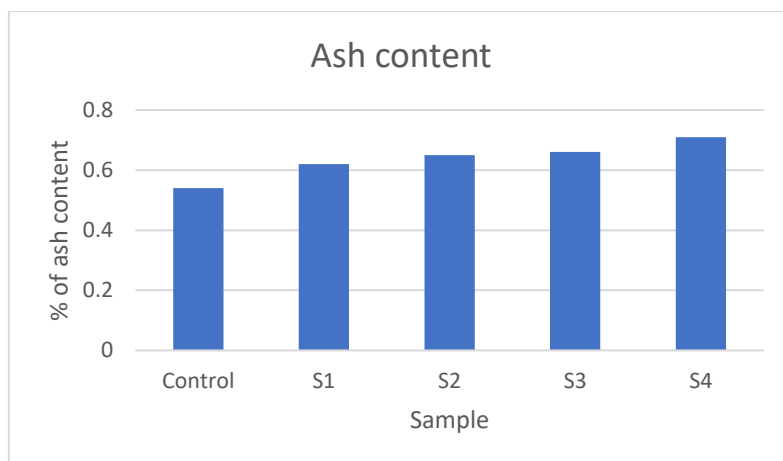
The body uses fat as a fuel source and fat is the major source of energy in the body. The fat percentage was determined by Mojonnier method. The fat percentage in herbal whey jelly was 0.29%(Control), 0.44%(T2), 0.53%(T3), 0.77%(T4), 0.82%(T5). Fat percentage is increased by adding spirulina. Spirulina have small amount of fat content. In related study herbal whey beverage noted that fat content was found to be 0.45 – 0.43%, (Dhananjay et al., 2018).



Fat composition of herbal whey jelly

#### 4.6.3 Ash

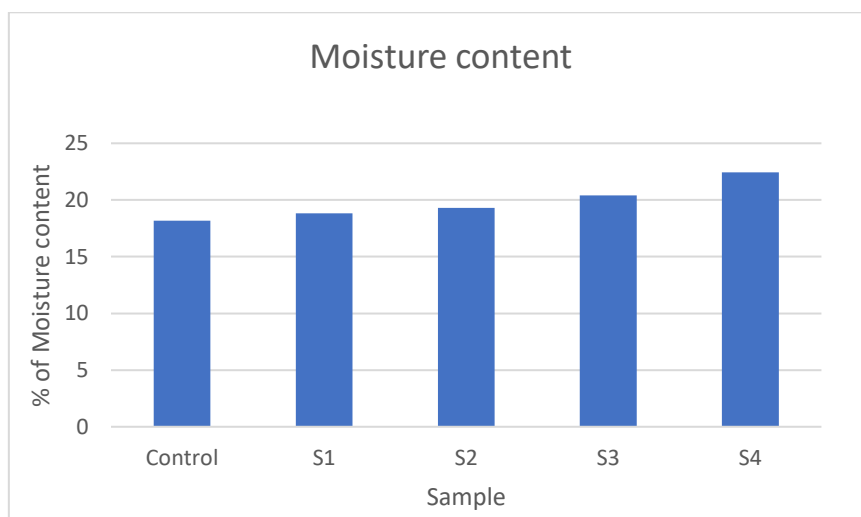
It refers to all non-aqueous, nongaseous residues that remains after something is burned. Ash is the inorganic residue remaining after water and inorganic matter have been removed by heating. The ash content in herbal whey jelly was 0.54%(Control), 0.62%(T2), 0.65%(T3), 0.66%(T4), 0.71%(T5). Small amount of ash content is present in all samples. In a study related ash content was found to be 0.38 – 0.57% (Rohit et al., 2019)



Ash composition of herbal whey jelly

#### 4.6.4 Moisture

Moisture content influence taste, texture, weight appearance shelf life of food products. Moisture content is one of the most commonly measured property of food materials. The moisture percentage in herbal whey jelly 18.1% (control), 18.8% (T2), 19.3% (T3), 20.4% (T4), 22.4% (T5). here, Moisture content was increases.

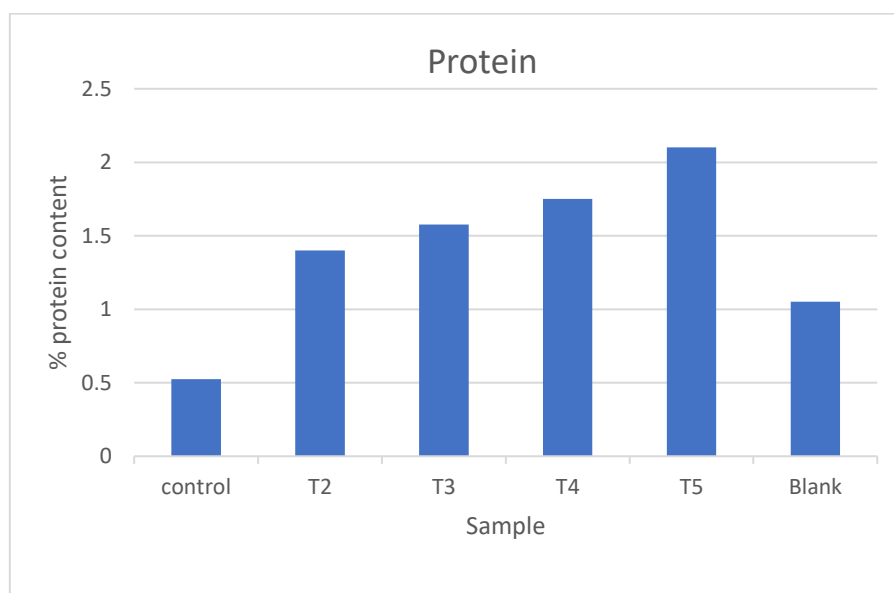


Moisture content of herbal whey jelly



#### 4.6.5 Protein

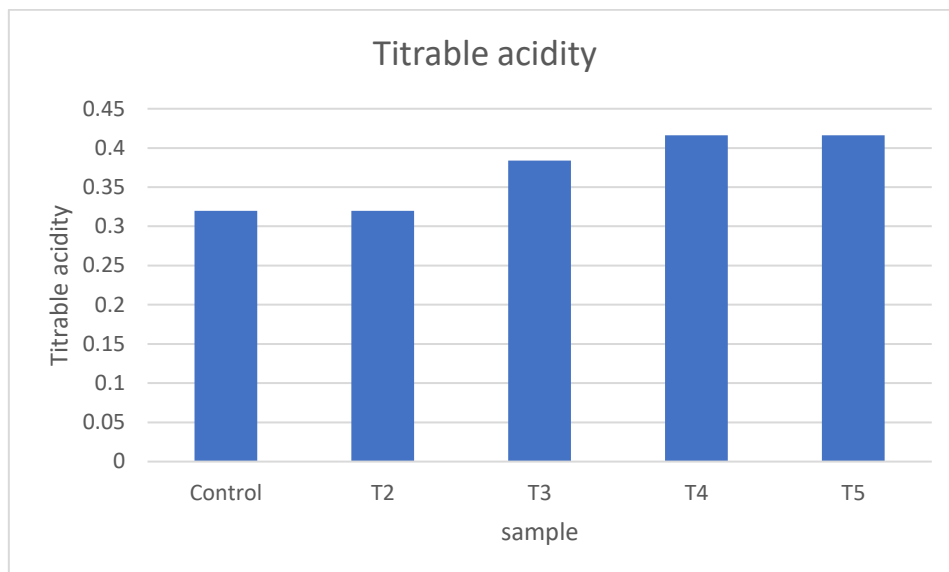
Protein is essential nutrient for those want to keep lean and build muscle. If you don't get enough protein in your diet, then your body breaks down its own muscle so it can be converted into energy. Addition of spirulina and whey increasing the protein content in final product. 60-70% of protein present in spirulina. The develop herbal whey jelly will be helpful for who following diet. Protein content is increasing by the addition of spirulina. The protein percentage was calculated by kjeldhal method. The protein content in herbal whey jelly was 0.52% (control), 1.40%(T2), 1.57(T3), 1.75%(T4), 2.10%(T5), 1.05% (Blank). Percentage of Protein content is increases. In a related to protein content study was found to be 1.90% - 2.45% (Asrul et al., 2021)



% of protein content in herbal whey jelly

#### 4.6.6 Titrable acidity

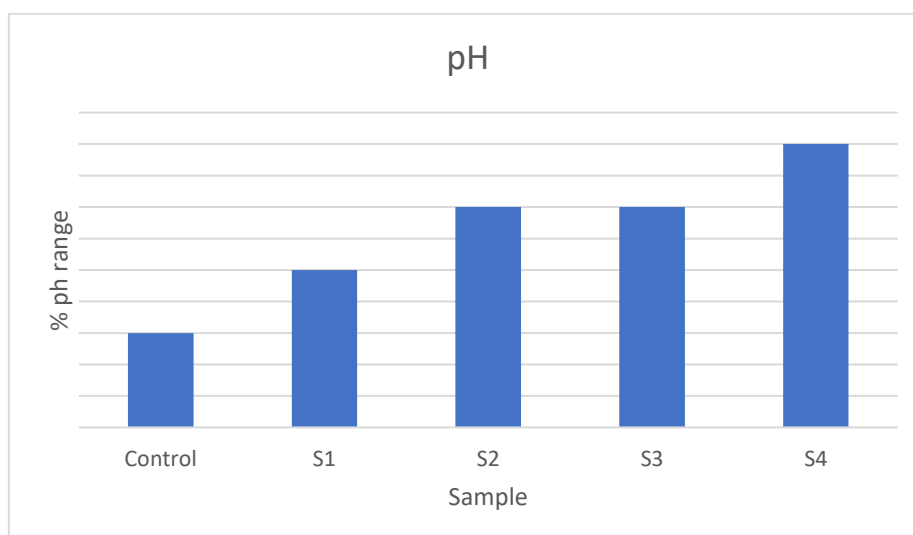
Titrateable acidity is a better predictor of how organic acid in food sample impact flavour. It measures the total acid ion determined by titration of intrinsic acid with standard base. The herbal whey jelly having acidity 0.32%(control), 0.32%(T2), 0.38%(T3), 0.41% (T4), 0.41% (T5). In this related study acidity was found to been 0.20-0.43%. (Sharma et al., 2020)



Titrable acidity of herbal whey jelly

#### 4.6.7 pH

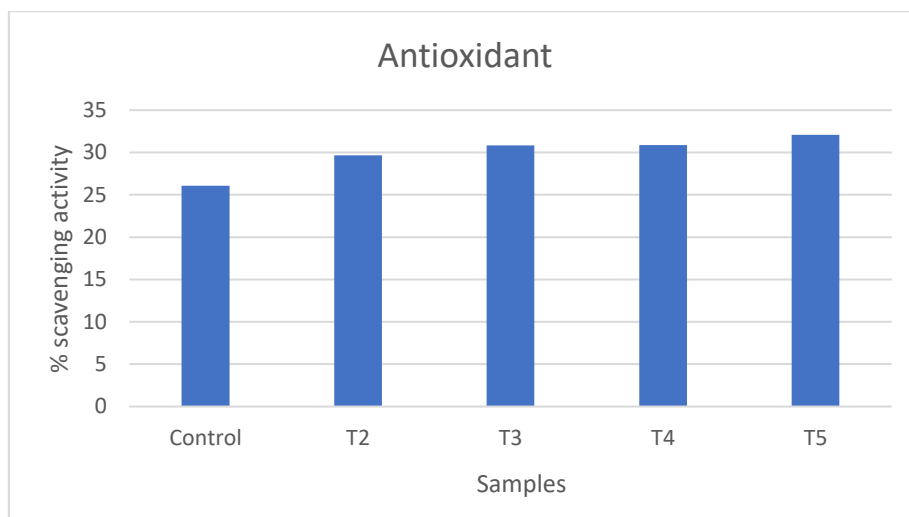
pH is important to assess the ability of microorganisms to grow in a specific food. It is the -ve logarithm (base 10) of hydrogen ion concentration is measured with pH meter. Here the pH of herbal whey jelly 3.96% (control), 3.97% (T2), 3.98% (T3), 3.98% (T4), 3.99% (T5). In this related ph was found to been 3.7-3.35%. (Deswandi et al., 2021)



% pH of herbal whey jelly

#### 4.6.8 Antioxidant

Antioxidants are compound that inhibit oxidation (usually occurring as autoxidation), a chemical reaction that can produce free radicals. Autoxidation leads to degradation of organic compounds, including living matter. Which is measured using DPPH method. 26.08% (control), 29.65% (T2), 30.84% (T3), 30.89 (T4), 32.08%(T5).



% of scavenging activity

#### 4.7 Physical methods

##### 4.7.1 Color characteristics

Color in food is an important attribute that affect consumer perception. This color will change according to the product. The color characteristics of herbal whey jelly was measured instrumentally in terms of L\* (lightness), a\* & b\* (color opponment dimension) values determined by Hunters jab

Color characteristics	control	T2	T3	T4	T5
L*	43.18	22.67	14.36	16.34	15.65
a*	1.18	-0.96	-2.03	-1.96	-1.52
b*	27.58	14.77	6.46	3.74	2.45

Color characteristics of herbal whey jelly control, T2, T3, T4 and T5 are mentioned above

L\* value ranges from 0 to 100 in which 0 indicates black & near to 100 represents whiteness. And the a\* represents redness if it has positive value and 0 indicates grey and if it has negative value, it represents greenness and its value ranges from -60 to +60. And the b\* is the degree of yellowness if it has positive value and 0 is the indication of grey and blueness if it has negative value and its value ranges from -60 to +60. These are the value representation of international color system.

#### 4.7.2 Texture profile analysis

It is known that various properties (texture, functionality and appearance) of foods were greatly affected by their structure. Rheological and textural jelly. The textural attribute of herbal whey jelly was analysed by using Texture Profile Analyzer. The observation on rheological attributes of herbal whey jelly of control, T2, T3, T4, T5 represented in table:

TEXTURAL ATTRIBUTES	Control	T2	T3	T4	T5
HARDNESS(N)	1974.73	136.707	653.838	825.281	2009.56
SPRINGINESS (mm)	0.468	0.926	0.507	0.406	0.312
COHESIVENESS	0.081	0.122	0.121	0.074	0.064
GUMMINESS (kgf)	159.28	16.703	79.233	60.682	128.463
CHEWINESS(Kgf.mm)	74.516	15.463	40.205	24.633	40.065

## 5.SUMMARY

Whey is a byproduct in the process of paneer and cheese production. Composition and characteristics of whey are depending on the production technology, the end product and the quality of used milk. Liquid whey consists of approximately 93% water and contains almost 50% of total solids present in the milk of which lactose is main constituent. Its disposal as waste poses serious pollution problems for the surrounding environment. The aim of this study was to utilize this byproduct and to enrich protein content in herbal jelly. The present study was undertaken to develop herbal whey jelly incorporated with spirulina and to study the effect of addition of spirulina on the physico-chemical parameters. Spirulina is a blue-green algae that grows naturally in saltwater lakes and oceans. It is rich in protein, vitamins, minerals, carotenoids, and antioxidants that can help protect cells from damage.

Control sample of herbal whey jelly was standardized by adding different levels of ginger and mint. Here, ginger and mint were taken in the following proportions to prepare T1 (2:2), T2 (2:3) and T3 (2:4) respectively. This control is prepared by adding 100ml of water, 50g of sugar, 0.5% of citric acid and 1.5 g  $\kappa$  carrageenan. The ginger and mint were also added as described above. This mixture is poured into a bowl and kept in refrigeration for setting. It was cut into desired size and shape. After sensory evaluation T3 (ginger to mint ratio 2:4) received highest score. Therefore, T3 was selected and hereafter used as control as herbal whey jelly.

Spirulina incorporated herbal whey jelly was prepared by spirulina at different levels (viz., 0.5%, 1%, 1.5%, 2%) and it was coded as S1, S2, S3 and S4. Sensory evaluation was done to select the best combination and S3 was selected as it received the highest sensory scores.

From present investigation, it may be concluded that ginger, mint extract and spirulina can be successfully utilized for preparation of herbal whey jelly. Even though spirulina imparted an unfavourable flavour to the product it improved nutritional quality of the product. Addition of ginger and mint extract improved flavour and aroma of the jelly. Also, it masked the tang of spirulina. It is well known that ginger and mint can help to boost the immune system and fight off illnesses.

Physico-chemical analysis of all treatments was conducted which include estimation of total moisture content, protein, fat, ash, acidity, pH, TSS, antioxidant content. The color

characteristics were determined using Hunters colorlab and textural attributes using texture profile analyser.

Addition of spirulina at higher levels cause  $\kappa$ -carrageenan to loss its binding property. The optimized product was found to have 18.16%(control), 18.82% (T2), 19.30% (T3), 20.40% (T4), 22.45% (T5) of moisture, Ash content (control) 0.54%, (T2) 0.62%, (T3) 0.65%, (T4) 0.66%, (T5) 0.71%, The protein content in jelly was found to have 0.525% (control), 1.401%(T2), 1.576%(T3), 1.751%(T4), 2.101%(T5), Total solids was found to have (control) 81.9%, (T2) 81.8%, (T3) 80.7, (T4) 79.6, (T5) 77.5%. Titrable acidity was found to have (control) 0.32%, (T2) 0.32%, (T3) 0.384%, (T4) 0.416%, (T5)0.416%, The optimized product has (control) 26.08238%, (T2) 29.65201%, (T3) 30.84821%, (T4) 30.89157%, (T5) 32.08054%. it was found to been increase protein content in herbal whey jelly. Because of addition of spirulina powder. Ginger, mint extract and spirulina enhance the color and flavor to final product. Addition of k carrageenan provide good soft body and texture. So, there is no side effect by adding these ingredients. Overall acceptability was good.

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