FORMULATION OF FLAVOURED NON-DAIRY MILK POWDER

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ABSTRACT
The creation of a flavor-enhanced hemp milk powder is described in this research study. A popular plant-based milk substitute that has several health advantages and a high nutritional value is hemp milk. In order to improve the hemp milk powder's flavors and customer appeal, flavorings were added throughout the formulation process. In this study, honey, vanilla extract, was employed as flavorings. In order to enhance the flavor profile and sensory properties of the hemp milk powder, hemp milk was dried by using spray dryer. The physical, chemical, and sensory qualities of the improved formulation were assessed. The outcomes demonstrated that the taste and general acceptance of the hemp milk powder were greatly enhanced by the addition of natural flavorings. The improved recipe smelled good and tasted sweet and nutty, with a creamy texture. The results of this study show that hemp milk intake may be encouraged as a healthy and enticing dairy-free option by enhancing its flavor and appeal using natural flavorings. In this study 13 formulation were formulated by using response surface methodology (RSM) in central composite design.

Keywords: Hemp seed, Hemp milk powder, Spray dryer, RSM.
VEGAN INDUSTRY

CHAPTER-1 INTRODUCTION

The worldwide vegan food market length turned into USD 23.31 billion in 2020. The marketplace is projected to develop from USD 26.16 billion in 2021 to USD 61.35 billion in 2028 at aCAGR of 12.95% during the 2021-2028 length. The worldwide impact of COVID-19 has been extraordinary and outstanding, with witnessing an effect on call for across all regions amid the pandemic. The growing demand for opportunity dairy analogues and meat analogues from consumers is predicted to enhance product adoption.

Plant based non-dairy milk merchandise have become famous among purchasers. Growing recognition of vegan products among youth gives more contributes to market growth. This aspect has precipitated the corporations to innovate and release new products inside the market. Health and well-being have become the two important elements contributing purchasers to shift their purchase patterns, causing a rise in the income of vegan products inside the international market.

Enthusiasts suggest that this mainstreaming of veganism has the potential to enact significant ethical, ecological and health changes to an agri-food system increasingly understood as broken. Meanwhile critics caution that this mainstreaming risks diluting the radical ethics of veganism, and argue that it is characterized by notable continuities and consolidations of who is developing, producing and profiting from new vegan products. These debates center on the emergence of what we term ‘Big Veganism’: the recent turn by ‘Big Food’ and ‘Big Agriculture’ to veganize their offerings and bring vegan products into mainstream spaces of food production and consumption. This model of veganism’s mainstreaming is evidenced by the kinds of developments described at the opening of this paper that have seen the biggest names in food and agriculture over recent years increasingly incorporate vegan options into their operations through direct investment, acquisitions and even corporate rebranding.1 Regular headlines attest to global networks of commodity plant-based (e.g. soya, wheat, pea) and biotechnological ingredients being mobilized by agri-businesses to meet the growing demand for high-tech, ersatz, ‘ultra-processed’ foods, like the flagship Impossible Burger and plant-based milks. (Alexandra E Sexton., 2022)

This model is grounded in the prevalent neoliberal politics of individual food choice and garnish food cultures, in which fetishized, often expensive products are marketed primarily to privileged audiences that celebrate white ‘shredded’ bodies and the welfare of charismatic animals. Alternative versions of lower-tech, minimally processed and socially embedded vegan foodways are noticeably absent from the Big Vegan model. While examples of these alternative vegan offerings can be found in more niche food and health retailers (e.g. Hodmedod’s Pulses and Grains, and Riverford Organic in the UK), Big Veganism has arguably emerged as the significant driving force of the current mainstreaming of vegan identity, practice and products in Europe and
North America. The considerable cultural and financial power it continues to amass at pace is defining the politics of possibility of what contemporary veganism is, who it is for, the geographies and scales it encompasses, and what kinds of alterity to conventional food systems it can deliver. (Tara Garnett, 2022)

In 2020, the global vegan food market will be worth USD 23.31 billion. The market is anticipated to grow at a CAGR of 12.95 percent from USD 26.16 billion in 2021 to USD 61.35 billion in 2028. The COVID-19 epidemic has had an unprecedented and spectacular global impact, affecting calls in all geographical areas. Consumer demand for meat and dairy alternatives is expected to increase, which will accelerate product adoption. Products made from plants that are non-dairy milk have gained popularity with consumers. More young people are becoming aware of vegan products, which helps the industry expand. This factor has encouraged businesses to innovate and introduce new products to the market. The two main factors influencing customers to change their purchasing behaviors are now health and well-being, which has increased the demand for vegan products on the global market (Christpoher J. Brynt., 2022)

HEMP SEED

Hemp seeds are a wealthy source of vitamins and wholesome fats, such as omega-3 and omega-6 fatty acids. Part of the hemp plant, those seeds are technically anut that may be eaten uncooked or used to make milk, oil, cheese substitutes, or protein powder. While related to the cannabis plant, hemp seeds have none of the psychoactive compound THC observed in marijuana. Hemp seed’s nutty flavor and versatility additionally make them a high-quality alternative for the ranges of protein, essential fatty acids, and different dietary blessings discovered in meat and dairy products. Each of those fats is recognized for improving coronary heart health by way of decreasing cholesterol, blood stain, and triglycerides. Adding hemp oil for your food plan may also reduce risk of heart problems. The minerals and vitamins in hemp seeds can provide a health advantages. As an instance, hemp oil is rich in vitamin E, which is beneficial for helping keep immune system functioning. It also acts as an antioxidant to reduce free radicals that cause cell damage. (R.E. Aluko, 2017).
According to United States Department of Agriculture (USDA) Data from Anamika (2021)

28 grams of hemp seed contains

Table 1.1 Nutritional information of hemp

<table>
<thead>
<tr>
<th>Nutritional Component</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy</td>
<td>161 calories</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>330 milligrams</td>
</tr>
<tr>
<td>Protein</td>
<td>920 milligrams</td>
</tr>
<tr>
<td>Fat</td>
<td>1230 milligrams</td>
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<tr>
<td>Fiber</td>
<td>200 milligrams</td>
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<tr>
<td>Manganese</td>
<td>2.8 milligrams</td>
</tr>
<tr>
<td>Vitamin E</td>
<td>15.4 milligrams</td>
</tr>
<tr>
<td>Copper</td>
<td>0.1 milligram</td>
</tr>
</tbody>
</table>

MILK POWDER

Milk is a nutritious food. Raw milk from healthy cows is supposed to contain relatively few bacteria. Drinking of raw milk of healthy cows reduces the health risk but it is highly perishable. In history, mankind has made various attempts to preserve and concentrate milk so as to extend its shelf life. Milk powder is a perfect solution to those who lack immediate access to adequate refrigeration facilities. Milk powder results from extracting water content out of milk. The main purpose of converting milk into Milk powder is to convert the liquid perishable raw material to a product that can be stored without substantial loss of quality, preferably for some years. (Patel, H. A., 2013)

The Milk powder has various applications in confectionaries, bakeries, infant formulas, nutritional foods, etc. Production of Milk powder has become an increasingly important segment of the dairy industry, which is expected to grow further because of its features such as better keeping quality, less storage space, and lower
transportation costs, which result in attractive economics and convenience during industrial and domestic formulation of composite foods. Non fat dry milk serves the same purpose for milk solids-not-fat that traditionally butter has done for milk fat. The ultimate aim of the industry is to obtain dry products, which if recombined with water give little or no evidence of detrimental change compared to the original liquid product. Various names have been applied to the same dry milk product. For example, non fat dry milk also has been called skim milk powder (SMP), dried skim milk, non fat dry milk solids, and dehydrated skim milk. Dry whole milk, dry cream, dry buttermilk, and others frequently are called dried or powdered whole milk, according to their identities. (Marathe, J. A., 2010)

**DRIYING CHARACTERISTICS**

**SPRAY DRYING**

Spray drying is the continuous transformation of feed from a fluid state into dried particulate form by spraying the feed into a hot drying medium. The feed may be solution, slurry, emulsion, gel or paste, provided it is pumpable and capable of being atomized. It involves bringing together a highly dispersed liquid and a sufficient volume of hot air to produce evaporation and drying of liquid droplets. The air supplies the heat for evaporation and conveys the dried product to the collector; the air is then exhausted with the moisture.

Three types of atomizers are commercially used namely rotary atomizer, pressure nozzle and two-fluid nozzle. (Gao, J., 2007)

The feed droplets while losing its moisture to hot air remain at temperatures much below the hot air temperature for a very short time. Hence spray drying is essentially known as "Low Temperature Drying". The dried product can be in the form of powders, granules, or agglomerates depending upon the physical and chemical properties of the feed, the dryer design and final powder properties desired. It is used for "In-House" manufacturing of critical excipients. (Subramaniyan R., 2010)

The spray-dried products have improved mean particle size and particle size distribution, appearance, texture, flow property, compressibility, bulk density, dispersibility and solubility. It has wide range of non-pharmaceutical and pharmaceutical applications. It is used for preparation of tableting constituents, vaccines, vitamins, blood products, enzymes, hormones, algae, yeast extracts. Spray drying plants are tailor made to suit product to be dried and its properties desired. (Patel R. K., 20)
CHAPTER-2 LITERATURE REVIEW

PLANT BASED MILK ALTERNATIVE

Sustainability, health-related concerns, change in life style and dietary reasons are the reasons for the acceptance of plant based milk alternatives. Fermentation in plant based milk improves sensory profiles, nutritional properties, texture and microbial safety. Plant based milk alternatives are usuallunnutritionally unbalanced, but it can be solved by the addition of required nutrients (Muzi Tangyu et al., 2019)

Non thermal processing techniques are used for the processing of plant based milks. Fortification improves the stability and nutrient content of the plant based milk (Swati Sethi et al., 2016).

Maria Luisa Astolfi compared dairy milk with plant based milk alternatives. Plasma mass spectrometry and cold vapour generation atomic fluorescence are the methods used for comparison. Result shows that dairy milk and plant based milk alternatives are safe from contamination due to toxic trace elements. (Maria Luisa Astolfi et al., 2020)

Large variety of pulses potentially suitable for processing into protein concentrates and isolates and various dry and wet processing options available, there is considerable scope for the development of highly functional protein ingredients (Martin Vogelsang- O’Dwyer et al., 2021).

Veganism is the subject of an increasingly diverse body of social scientific research, focusing on the literature that explores multiple contested modes of veganism. (Tara Garnett et al., 2022)

Due to lactose intolerance and allergies, demand for plant-based milk alternatives increased over recent years. Main challenges in plant based milk alternatives are to enhance stabilization, reduce antinutrients, off-flavor and allergens (F Reyes-Jurado et al., 2021).

The potential of plant-based milk alternates and associated challenges are the aim of the review is to give a scientific comparatives of non-dairy milk beverage (Satish Kumar et al., 2019)

The plant based milk can be a good alternative of cow milk as an emerging segment of functional beverages and researched for the tackling of problems related to shelf life, emulsion stability, nutritional completeness and sensory acceptability of final product (Swati Sethi S. K et al., 2016)
HEMP SEED

R E Aluko (2017) noted high presence of tetrahydrocannabinol [THC] makes the hemp seed suitable for consumption. During storage the moisture content in the hemp seeds will be below 10% to prevent sprouting. Based on the individual type hemp seeds sometimes cause allergies. (R E Aluko et al., 2017)

Claudia Anna Emma Greslehner (2008) investigated the beginning of hemp seed cultivation to the production of hemp milk and other products from hemp seeds. Austria is the first country to produce hemp milk from hemp seeds. Due to high price and difficulty to access, the hemp seed and its products are not yet popular to general public. (Claudia Anna Emma Greslehner et al., 2008)

The study was to identify and test efficiency of antioxidant and antihypertensive peptides present in hemp seed protein hydrolysate (HPH). In vitro and in vivo studies have shown that hemp seed peptides possess the potential to be used as antioxidant and anti hypertensive agent (Sunday Malmo et al., 2013)

The growing of industrial hemp and its market acceptability was high. Hemp seed is a valuable product in its own right for food industry, which can be developed into value-added ingredient (R.A.Burton et al., 2022)

K Sacilik noted that the hemp seed consumption reduces cholesterol and high blood pressure. Linoleic and linolenic acids are added to hemp seed to make it perfectly balanced for human consumption. The dimensions of hemp seeds changes with the change in the moisture content. (K Sacilik et al., 2003)

Hemp seed protein isolate had minimum protein solubility at pH 4.0 and it was increased when pH was increased or decreased. The protein functionality of hemp seeds are highly dependent on structural confrontation (Sunday A Malomo et al., 2014).

HEMP MILK

Lorenzo Nissen concentrated on substituting dairy products. Fermented hemp milk drinks can be introduced as a dairy substitute. This leads to development of food having low glycemic index, low in saturated fats, carbohydrates and additives are successfully developed. (Lorenzo Nissen et al., 2020)

The more knowledge of hemp seed milk can ensure its enlarge market. New research should have occurred about hemp seed milk and hemp seed milk products (Nour Awad et al., 2022)

The nutritional comparision of hemp milk with other plant based milk alternative with taste, texture, cost (U.S.Vahanvaty et al., 2009)

To form physically and oxidatively stable hemp milk, high pressure homogenization (HPH) combined with pH shift treatment is used. Basically plant based milks are unstable so this HPH – pH shift treatment is used (Qingling Wang et al., 2018).
Wet milling and dry milling are the two methods for the production of hemp milks. Both the methods give same yield of milk. Production process and analyzing of hemp milk will understood (Briana Marie Naylor et al., 2021)

CHAPTER-3 MATERIALS AND METHODOLOGY

COLLECTION OF RAW MATERIAL

Hemp seeds were purchased from online website Meesho of brand MOKSA. Other ingredients such as, muslin cloth, sweetener were purchased from the nearby supermarket. These ingredients were used in the development of milk powder.

HEMP SEED

Hemp, or industrial hemp, is a botanical class of Cannabis sativa cultivars grown specifically for industrial or medicinal use. It can be used to make a wide range of products. Along with bamboo, hemp is among the fastest growing plants on Earth. Hemp seeds can be eaten raw, ground into hemp meal, sprouted or made into dried sprout powder. Hemp seeds can also be made into slurry used for baking or for beverages, such as hemp milk and tisanes. Hemp oil is cold-pressed from the seed and is high in unsaturated fatty acids. Hemp seeds are a rich source of nutrients. Part of the hemp plant, these seeds are technically a nut that can be eaten raw or used to make milk, oil, cheese substitutes, or protein powder.

Fig 3.1 Hemp seed

MUSLIN CLOTH

Lightweight and breathable, muslin is a loose plain weave cotton material that dates back to Ancient India. Today, muslin’s value is in its versatility and it is used in everything from photography backdrops to cooking to surgical procedures. Muslin is a loosely woven cotton fabric. It’s made using the plain weave technique, which means that single weft thread alternates over and under a single warp thread. Muslin is known as the material used
in fashion prototypes to test patterns before cutting and stitching the final product.

**EXTRACTION OF HEMP MILK**

Hemp seeds (100g) were soaked for 10 hours. After soaking, by the process of wet milling hemp seeds were grinded and milk was obtained. This hemp milk was filtered using muslin cloth having 8 layers. After that hemp milk was packed in storage bottles and stored under refrigeration condition and utilized for production of milk powder. (Briana Marie Naylor, 2021)

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**SOAKING (12 HOURS)**

| Fig 1 Soaked seed | Fig 2 Blended Hemp seed | Fig 3 Hemp milk |

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**Figure 3.2 Flow sheet for extraction of hemp milk**
FORMULATION OF HEMP MILK POWDER

Hemp milk was taken about 100ml and addition of sweetener and flavoring substances like honey and vanilla essence and the milk was taken into spray dryer for drying purpose. In spray dryer milk was preheated to 90 degree Celsius then preheated milk is taken into drying at 177 degree Celsius for 3 minutes.

(Ismail, F et al., 2021)

![Flow sheet for Formulation of hemp milk powder](image-url)
CHAPTER 4

PHYSIO-CHEMICAL ANALYSIS

PHYSIO-CHEMICAL ANALYSIS

MOISTURE CONTENT

Apparatus:

- Dish made of porcelain, silica or platinum
- Electric oven
- Weighing balance
- Desiccator

Procedure:

5g of the sample is weighed accurately in a previously dried and weighed moisture dish. It is dried for 4 hours at 100±2°C in an oven. It is then cooled in a desiccator and weighed with the lid on. The process of drying, cooling and weighing is repeated at half-hour intervals until the difference in mass between two successive weighing is less than two milligram. The lowest mass obtained is recorded.

Calculation:

Moisture, percent by mass = \( \frac{100(M1-M2)}{(M1-M)} \)

\( M1 = \) mass, in g of dish with material before drying.
\( M2 = \) mass in g of dish with material after drying to constant mass.
\( M = \) mass in g of the empty dish.

ASH CONTENT

Ash content refers to the amount of inorganic residues remains after ignition process. For find out ash content muffle furnace is used. 5g of sample is taken in crucible and placed in muffle furnace for 4 hours at 550°C. After ignition ash was obtained and weighted. (IS 4706 (Part II))

Ash Content (%) = \( \frac{W2 - W}{W1 - W} \times 10 \)

\( W = \) Weight of empty dish, g

\( W1 = \) Weight of dish with fresh material, g

\( W2 = \) Weight of dish with ash, g
**CRUDE FIBRE**

**Apparatus:**
- Beakers
- Gooch crucible
- Air Oven
- Muffle furnace
- Weighing balance
- Condenser
- Filtering cloth
- RB flask of 500ml capacity provided with a condenser

**Reagents:**
- Dilute Sulphuric Acid -1.25% (w/v) accurately prepared
- Sodium Hydroxide solution -1.25% (w/v) accurately prepared
- Ethyl Alcohol- 95% by volume
- Petroleum ether

**Procedure:**

Weigh accurately about 2.5g of sample in a suitable thimble. Place the thimble in the Soxhlet extraction apparatus and extract with petroleum ether for 1 hour. Transfer the fat free material to a 500ml round bottomed flask. Boil 200ml of dilute Sulphuric acid in a beaker. Immediately connect the flask with a water cooled reflux condenser and heat so that the contents of the flask begin to boil within one minute. Rotate the flask frequently taking care to keep the material from remaining on the sides of the flask and out of contact with the acid. Continue boiling for exactly 30 minutes. Remove the flask and filter through fine linen (about 18 threads to the centimeter) held in funnel, and wash with boiling water until the washings are no longer acid to litmus. Wash the residue thoroughly with 200ml of boiling sodium hydroxide solution. Immediately connect the flask with the reflux condenser and boil for exactly 30 minutes. Remove the flask and immediately filter through the filtering cloth. Wash the residue with boiling water and transfer to Gooch crucible prepared with a thin but compact layer of ignited asbestos. Wash the residue thoroughly first with hot water and then with 15ml of ethyl alcohol. Dry the Gooch crucible at 105 ± 2°C in an air-oven to constant weight. Cool and weigh. Incinerate the contents of the Gooch crucible in a muffle furnace at 600 +20°C until all the carbonaceous matter is burnt. Cool the Gooch crucible in a desiccator and weigh.
Calculation:

Crude fiber (on dry basis),
percent by mass = \( \frac{100(M1-M2)}{M\ast(100-W)} \)

Where

\( M1 = \) mass, in g of Gooch crucible and contents before ashing
\( M2 = \) mass in g of Gooch crucible containing asbestos and ash
\( M = \) mass in g of the material taken for test
\( W = \) percent of moisture content

pH

Apparatus:
- pH meter with Glass electrode
- Mortar
- Weighing balance

Procedure:
Weigh accurately about 10g of sample and grind to a fine paste in mortar. Add 100ml of water and mix thoroughly. Allow the mixture to stand for 15 minutes and filter. Determine the pH of the filtrate using a pH meter.

TOTAL FAT

Apparatus:
- Soxhlet Extraction Apparatus
- Electric oven
- Weighing balance

Reagents:
- Petroleum Ether (40-60°C)

Procedure:
5g of the material is weighed accurately in a suitable thimble and dried for 2 hours at 100±2°C. The thimble is placed in the Soxhlet extraction apparatus and extracted with Petroleum Ether (40-60°C) for 8 hours. The extract contained in the Soxhlet flask is dried, the empty mass of which has been previously determined by tarring at 95°C-100°C for one hour. It is then cooled in a desiccator and weighed. The process of drying, cooling and weighing is repeated at half-hour intervals until the difference in mass between two successive weighing is less than two milligram. The lowest mass obtained is recorded.
Calculation:
Fat % by mass = \( \frac{100 \times (M_1 - M_2)}{M} \)

Where,
\( M_1 \) = Mass in g of Soxhlet flask with the extracted fat.
\( M_2 \) = Mass in g of empty Soxhlet flask
\( M \) = Mass in g of the material taken for test.

**TOTAL SOLID CONTENT**

Total solid content is found out using oven drying method. 5g of non-dairy hemp powder sample is taken and kept in hot air oven at 105℃. Weight the sample every one hour till the sample weight becomes constant. Then moisture content is determined by dry basis and subtract it from 100 total solid content is obtained.

**Total Solids (%) = 100 – Moisture Content**

**CARBOHYDRATES**

Carbohydrate is determined by difference method. Total carbohydrates are calculated as follows after determining the percentage of moisture, total protein, fat and total ash.

Calculation:
Total carbohydrates = 100-(A+B+C+D)

Where,

**PROTEIN**

\( A = \text{percent by mass of moisture} \)
\( B = \text{percent by mass of total protein} \)
\( C = \text{percent by mass of fat} \)
\( D = \text{percent by mass of total ash} \)

- Kjeldahl apparatus
- Beaker
- REAGENTS:
  - Concentrated sulphuric acid (nitrogen free)
  - Mercuric oxide or copper sulphate
  - Potassium sulphate or anhydrous sodium sulphate
  - Sodium hydroxide solution-50%
  - Standard sodium hydroxide solution-0.1N or 0.5N
  - Standard Hydrochloric or Sulphuric acid solution-0.1 or 0.5N
  - Methyl red indicator
Procedure:

Digestion

Weigh accurately 0.7 to 2.2 g of the sample into the digestion flask and add 0.7 g mercury oxide or copper sulphate and 15 g powdered potassium sulphate or anhydrous sodium sulphate and 25 ml sulphuric acid. Each gram of fat consumes 10 ml and each gram of carbohydrate 4 ml sulphuric acid during digestion. Place the flask in an inclined position on a heater and heat gently until foaming ceases. Then boil vigorously until the solution becomes clear and then continue boiling it for 1 to 2 hours.

Distillation

Cool the digested sample and add about 200 ml distilled water into it for avoiding complex formation, add 25 ml of the sulphide or thiosulphate solution mix to precipitate the mercury. Add a few zinc granules/glass beads to prevent bumping, incline flask, and add without agitation 25 g of sodium hydroxide as solid or equivalent solution, to make solution strongly alkaline. Immediately connect flask to distillation bulb or trap on condenser, and, with tip of the condenser immersed in a measured quantity standard acid. In the receiver, rotate flask to mix the contents thoroughly, then heat immediately until all ammonia has distilled over (at least 150 ml distillate). Lower the receiver before stopping distillation and wash tip of condenser with distilled water. Back-titrate excess acid with standard 0.1 N Sodium hydroxide using Methyl red as indicator. Conduct a blank determination in reagents.

Calculation:

Calculation of Nitrogen content

Nitrogen content in g = [(a-0.2b)-(c-0.2d)] × 0.007

Where,

a = volume in ml 0.5 N acid measured for main distillation
b = volume in ml 0.1 N alkali used for back titrating a

C = volume in ml 0.5 N acid measured for blank distillation
D = volume in ml 0.1 N alkali used for back titrating C

OR

Nitrogen content in g = [(A-B)-(C-D)] 0.0014

A = volume in ml 0.1 N acid measured for main distillation
B = volume in ml 0.1 N alkali used for back titrating A
C = volume in ml 0.1 N acid measured for blank distillation
D = volume in ml 0.1 N alkali used for back titrating C
ENERGY VALUE

Energy value is determined by calculation

Table 4.1 Energy conversion

<table>
<thead>
<tr>
<th>Carbohydrates</th>
<th>17 KJ/g, 4kcal/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat</td>
<td>38 KJ/g, 9kcal/g</td>
</tr>
<tr>
<td>Protein</td>
<td>17 KJ/g, 4kcal/g</td>
</tr>
</tbody>
</table>

Calculation:
Energy (KJ/g) = (Carbohydrate * 17) + (Fat * 38) + (Protein * 17)
Energy (kcal/g) = (Carbohydrate * 4) + (Fat * 9) + (Protein * 4)

TITRATABLE ACIDITY

Titratable acidity is determined using principles of titration. Weight 2g of sample and crush it using mortar and pestle and mix with 3ml of distilled water. Then the mixture is again mixed with 17ml of distilled water. Two or three drops of phenolphthalein is added as indicator and titrated against 0.1N NaOH till pale pink color appears in the solution. Volume of NaOH used for titration is noted and calculation is done. (IS 548 (Part 1))

Titratable Acidity (%) = \((0.9*v*N)/V\)

v – Weight of sample, g
V – Volume of NaOH used in titration

SENSORY ANALYSIS

Sensory evaluation can be done on the basis of 9 point hedonic scale. It can be evaluated by a set of faculty members and students. The sensory attributes taken for evaluation are color, taste, texture, flavor and overall acceptability. Each trial is evaluated and overall acceptability rate can be determined and noted. The 9 point hedonic scale is given below.
Table 4.2 Point Hedonic Scale

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Rating</th>
</tr>
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<tbody>
<tr>
<td>Like Extremely</td>
<td>9</td>
</tr>
<tr>
<td>Like Very Much</td>
<td>8</td>
</tr>
<tr>
<td>Like Moderately</td>
<td>7</td>
</tr>
<tr>
<td>Like Slightly</td>
<td>6</td>
</tr>
<tr>
<td>Neither Like nor Dislike</td>
<td>5</td>
</tr>
<tr>
<td>Dislike Slightly</td>
<td>4</td>
</tr>
<tr>
<td>Dislike Moderately</td>
<td>3</td>
</tr>
<tr>
<td>Dislike Very Much</td>
<td>2</td>
</tr>
<tr>
<td>Dislike Extremely</td>
<td>1</td>
</tr>
</tbody>
</table>

**Response Surface Methodology (RSM)**

Response surface methodology (RSM) includes optimization procedures for the settings of factorial variables, such that the response reaches a desired maximum or minimum value. The response is in effect modeled by factorial techniques and ANOVA, but these are extended for more detailed modeling of the effects.

**CCD (CENTRAL COMPOSITE DESIGN) FOR OPTIMIZATION PROCESS**

The Box and Wilson design or CCD model comprising of factorial1, factorial2, and factorial3 design. The star point outside the domine and the center point, representing the experimental domine, helps determine the response surface plot. By estimating the precision of surface responses, the value of $\alpha$ can be determined; where star design is $\pm \alpha$. There are three types of CCD; the $\alpha$ can be determined according to the calculation possibilities and the required precision, which can be obtained from surface responses.
Table 4.3 RSM Table CCD (Central Composite Design) for optimization process

<table>
<thead>
<tr>
<th>Std Run</th>
<th>Factor 1 (Hemp gm)</th>
<th>Factor 2 (Water ml)</th>
<th>bonse1</th>
<th>bonse2</th>
<th>bonse3</th>
<th>bonse4</th>
<th>Response 5</th>
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<tr>
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<tr>
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<td>13</td>
<td>225</td>
<td>8</td>
<td>9</td>
<td>8</td>
<td>9</td>
<td>8.5</td>
</tr>
</tbody>
</table>

MICROBIAL ANALYSIS

TOTAL PLATE COUNT

Disinfect the surface of the bottle/pouch/cups containing sample with 70% ethanol. Thoroughly mix the sample by vigorous shaking to achieve uniform distribution. Aseptically inoculate 1 ml of the water sample using sterile pipette into sterile petri plates in duplicate in two sets. The petri plates should be labeled with the sample number, date and any other desired information. Pour into each plate 15–18 ml of the molten sterilized PCA media (cooled to 44°C–47°C). Avoid pouring of molten medium directly onto the inoculum. Mix the media and inoculum by swirling gently clockwise and anti-clockwise so as to obtain homogenous distribution of inoculum in the medium. Allow to cool and solidify. In case, where in sample microorganism having spreading colonies is expected, add 4 ml of overlay medium onto the surface of solidified plates. After complete solidification, invert the prepared plates and incubate one set at 37°C for 24 hr and other set at 20 – 22°C for 72 hr. After specified incubation period count all colonies including pinpoint colonies. Spreading colonies shall be considered as single colony. If less than one quarter of dish is overgrown by spreading, count the colonies on the unaffected part of the dish and calculate corresponding number in the entire dish.

If more than one quarter is overgrown by spreading colonies discard the plate. (IS 4251 : 1967)

\[ N = \text{Colonies Plate 1} + \text{Colonies Plate 2} \]
YEAST AND MOLD COUNT

Aseptically clean the surface of bottle/pouch/cups containing sample with 70% ethanol and filter 250ml (or as specified) of water sample through a membrane filter of 0.45μm pore size using sterile membrane filtration assembly. Place the filter on CGYEA media and incubate at 25±1°C. Observe the plates for colonies of Yeast & Molds on 3rd, 4th and 5th days of incubation. (IS 5403: 1999)

Yeast and Mold Count = Present * ml of sample taken
Absent.

CHAPTER 5 RESULT AND DISCUSSION

This chapter deals with the result obtained from the various experiment conducted on formulation of hemp milk powder and to estimate the nutritional quality by chemical analysis, microbial analysis, physical analysis and sensory analysis.

OPTIMIZATION OF HEMP MILK USING RSM

Response 1: Flavor

Where X1=hemp ,X2=water In this 3D graph values of X varies ranges from 30 to 60 and Y value ranges from 150 to 300 The flavor of hemp milk varied between minimum range of 6 and maximum range of 8. The maximum efficiency of flavor was obtained from the trial that had 45g of hemp and 225 ml of water.
Response 2: Colour

Where $X_1$=hemp , $X_2$=water In this 3D graph values of X varies ranges from 30 to 60 and Y value ranges from 150 to 300 The colour of hemp milk varied between minimum range of 5 and maximum range of 9. The maximum efficiency of color was obtained from the trial that had 45g of hemp and 225 ml of water

Response 3 : Texture

Where $X_1$=hemp , $X_2$=water In this 3D graph values of X varies ranges from 30 to 60 and Y value ranges from 150 to 300 The texture of hemp milk varied between minimum range of 5 and maximum range of 8. The maximum efficiency of texture was obtained from the trial that had 45g of hemp and 225 ml of water

Response 4 : Taste
Where $X_1=hemp$, $X_2=water$ In this 3D graph values of $X$ varies ranges from 30 to 60 and Y value ranges from 150 to 300 The texture of hemp milk varied between minimum range of 5 and maximum range of 9. The maximum efficiency of texture was obtained from the trial that had 45g of hemp and 225 ml of water.

Response 5: Overall acceptability

The overall acceptability of hemp milk varied between minimum range of 6 and
maximum range of 8.5. The maximum efficiency of overall acceptability was obtained from the trial that had 45g of hemp and 225 ml of water.

The optimization of the sample was based on the sensory characteristics of the product as generated by the RSM software. The optimized trial was with 45 gram of hemp seed with 225 ml of water. For the optimized value, for flavor and texture was 8 and for the color and taste was 9.

**PHYSIO CHEMICAL ANALYSIS**

Physio-chemical analysis viz moisture, ash, total solid, fat, protein, fiber, carbohydrate, pH, titratable acidity, bulk density, particle density were done.

Table 5.1 physio chemical analysis

<table>
<thead>
<tr>
<th>S.NO</th>
<th>ANALYSIS</th>
<th>VALUE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Moisture (%)</td>
<td>4.91</td>
</tr>
<tr>
<td>2</td>
<td>Ash (%)</td>
<td>2.09</td>
</tr>
<tr>
<td>3</td>
<td>Total solids (%)</td>
<td>95.09</td>
</tr>
<tr>
<td>4</td>
<td>Fat (%)</td>
<td>9.79</td>
</tr>
<tr>
<td>5</td>
<td>Protein (%)</td>
<td>15.33</td>
</tr>
<tr>
<td>6</td>
<td>Fiber (%)</td>
<td>0.902</td>
</tr>
<tr>
<td>7</td>
<td>Carbohydrate (%)</td>
<td>67.88</td>
</tr>
<tr>
<td>8</td>
<td>pH</td>
<td>5.54</td>
</tr>
<tr>
<td>9</td>
<td>Titratable acidity (%)</td>
<td>1.7</td>
</tr>
<tr>
<td>10</td>
<td>Bulk density</td>
<td>0.664</td>
</tr>
<tr>
<td>11</td>
<td>Particle density</td>
<td>1.047</td>
</tr>
</tbody>
</table>

The Physio-chemical analysis of the hemp milk powder were analyzed in Indian standards methods moisture content of milk powder was 4.91%, ash content of milk powder was 2.09%, total solid content of milk powder was 95.09% fat content of milk powder was 9.79%, protein content of milk powder was 15.33% fiber content of
milk powder was 0.902% carbohydrate content of milk powder was 67.88 gram, pH content of milk powder was 5.58 titratable acidity content of milk powder was 1.7 bulk density and particle density of milk powder were 0.664 and 1.047.

MOISTURE CONTENT

Moisture content of hemp milk powder sample was found to be 4.91% by using oven drying oven drying method soy milk powder has the range of moisture content between 3.6% and 5.9% (Xiaojuan Wang, et al., 2015). While comparing our values with the reference value there is no significant difference in the moisture content. These range of moisture content helps to increase the shelf life of the product.

ASH CONTENT

Ash content of hemp milk powder sample was found to be 2.09% by using muffle furnace. Ash content of soy milk powder was found between 3.62% and 4.88% (W. H. Wang et al., 2015). While comparing values with the reference value there is no significant difference in the ash content.

TOTAL SOLID CONTENT

Total solids of hemp milk powder sample were found to be 95.09% by using oven drying method. Soy milk powder has the range of total solid content between 92.6% to 94.9%, (H.K. Lee and S.K. Sastry 1990). While comparing our values with the reference value there is no significant difference in the total solid content. This result will improves the quality, texture, and consistency of the hemp milk powder.

FAT CONTENT

Fat content of hemp milk powder sample was found to be 9.79% by using Soxhlet method. Similar to study conducted by 2016 (Shakerardekani et al.), fat content of almond milk powder was found 15.9% to 27.5%. While comparing with reference value fat content of hemp milk powder comparatively low which is also unsaturated fat suitable for consumers having fat related health issues.

PROTEIN CONTENT

Protein content of hemp milk powder sample was found to be 15.33% using Kjeldahl method. Protein content of almond milk powder was found to be 10.5% to 14.8% protein. (Babazadeh, A., et al. 2019). While comparing our values with the reference value protein content of hemp milk powder is high suitable for consumers who are seeking for protein.

FIBER CONTENT

Fiber content of hemp milk powder sample was found to be 0.92% using crude fiber method. The fiber in hemp milk powder is primarily soluble fiber, which can help to promote feelings of fullness and support digestive health. Including hemp milk powder as part of a varied and balanced diet can help to increase your overall fiber intake.

CARBOHYDRATE CONTENT

Carbohydrate content of hemp milk powder sample was found to be 67.88% using differential
method. Carbohydrate content of rice milk powder was found to be 86.4 to 91.4 gram. (Babazadeh, A., et al 2019). Carbohydrate of the hemp milk will digest easily as it contains simple form of sugar.

**pH**

pH of the hemp milk powder was found to be 5.54 using digital pH meter. pH of coconut milk powder was found to be 6.2 (Sanjay Yadav et al., 2019). While comparing our values with reference value only a slight difference in the pH was found.

**TITRATABLE ACIDITY**

Titratable acidity of hemp milk powder sample was found to be 1.7% using the principle of titration. Titratable acidity of almond milk powder was found to be 0.57% (Briana Marie Naylor et al., 2019). It is used for determination of acid content in the hemp milk powder.

**BULK DENSITY**

Bulk density of hemp milk powder was found to be 0.664 g/cm³. The bulk density of soy milk powder is 0.37 g/cm³ (N. Man et al. (2011)). While comparing values with the reference value there is no significant difference in the ash content.

**PARTICLE DENSITY**

Particle density of hemp milk powder was found to be 1.047 g/cm³. Particle density of hemp milk powder will improve the consistent quality and performance of the product.

**MICROBIAL ANALYSIS**

Microbial analysis viz standard plate, yeast and mold were done.

<table>
<thead>
<tr>
<th>S.NO</th>
<th>ANALYSIS</th>
<th>VALUE (CFU)/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Standard Plate Count</td>
<td>&gt;10</td>
</tr>
<tr>
<td>2</td>
<td>Yeast and Mold Count</td>
<td>NIL</td>
</tr>
</tbody>
</table>

**TOTAL PLATE COUNT**

Standard plate count value of hemp milk powder was found to be >10 CFU/g. This value is low
when compared to soy milk powder. While comparing the value with reference there is only slight difference in the standard plate count.

**YEAST AND MOLD COUNT**

Yeast and mold was absent in the hemp milk powder sample. This value is very low when compared to study conducted by Budiman C *et al.*, 2013, in the soya powder as he got $2.7 \times 10^2$ to $1.1 \times 10^3$ CFU/g. While our hemp milk powder yeast and mold is absent when compared to reference. It increases the shelf life of the powder.

**COST ANALYSIS OF HEMP MILK POWDER**

Table 5.3 cost analysis

<table>
<thead>
<tr>
<th>S.NO</th>
<th>EXPENSES</th>
<th>COST OF PRODUCTION OF 100g OF MILKPOWDER</th>
</tr>
</thead>
<tbody>
<tr>
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<td>Ingredients (A)</td>
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</tr>
<tr>
<td></td>
<td>Hemp seeds</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Vanilla essence</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Honey</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>115</td>
</tr>
<tr>
<td>2</td>
<td>Utilities (B)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Muslin cloth</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Water</td>
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<td>3</td>
<td>Packaging (C)</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>Profit (D)</td>
<td>30</td>
</tr>
<tr>
<td>5</td>
<td>Total Cost (A+B+C+D)</td>
<td>150</td>
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</tbody>
</table>

**CHAPTER- 6 CONCLUSION**

Formulation and analysis of hemp milk powder was studied. This hemp milk powder is recommended to those who is suffering from lactose intolerance. Hemp seeds are used as source of plant-based protein. Human body
cannot produce essential fatty acids, hemp seeds contains all 9 essential fatty acids which make diet balanced. Nowadays hemp products are suggested to patients undergoing post covid recovery. Hemp milk powder is a promising alternative to traditional dairy milk products. Hemp milk powder are successfully formulated with nutritional improvement when compared to conventional dairy milk powder. The fiber content in hemp milk powder is high when compared to normal milk based paneer. The sensory characteristics show similar results to milk based powder. The physicochemical characteristics like moisture content 4.91%, protein content 15.33%, carbohydrate content 67.88%, fiber content 0.92%.

**REFERENCE**

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