STUDIES ON DEVELOPMENT OF VEGAN MILK POWDER (OAT MILK POWDER) FORTIFIED WITH DATES

By

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B. Tech. (Food Technology)

DISSERTATION

Submitted to the
Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani
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MASTER OF TECHNOLOGY

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COLLEGE OF FOOD TECHNOLOGY

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PARBHANI - 431 402 (M.S.) INDIA

2020

Affectionately dedicated to

Almighty Allah,

My Parents,

Guide,

Co- Guides,

Family

&

Friends.

CANDIDATE'S DECLARATION

I hereby declare that the dissertation or part thereof has not been previously submitted by me for a degree of any other University or Institute.

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This is to certify that **Ms. SYED SAMAR JAVED** has satisfactorily prosecuted her course credit and research work for a period not less than four semesters and that the dissertation entitled **"STUDIES ON DEVELOPMENT OF VEGAN MILK POWDER (OAT MILK POWDER) FORTIFIED WITH DATES"** submitted by her is the result of original research work and is of sufficiently high standard to warrant its presentation to the examination. I also certify that the dissertation or part thereof has not been previously submitted by her for the award of a degree of any university.

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LIST OF ABBREVIATIONS

%	:	Percent
°Bx	:	Degree Brix
µg	:	Microgram
µL	:	Microlitre
ANOVA	:	Analysis of Variance
CD	:	Critical Difference
CFU	:	Colony Forming Unit
CO_2	:	Carbon Dioxide
cP	:	Centipoise
CRD	:	Completely Randomised Design
CV	:	Cardiovascular
et al.	:	*et alibi* (and associates)
FAO	:	Food and Agriculture Organization
FeS	:	Iron Sulfide
FSSAI	:	Food Safety and Standards Athority of India
g	:	Gram
GI	:	Gastrointestinal
GRAS	:	Generally Recognized As Safe
H2O2	:	Hydrogen Peroxide
H_2S	:	Hydrogen Sulfide
H_2SO_4	:	Sulfuric Acid
HCl	:	Hydrochloric Acid
i.e.	:	That is
IU	:	Internaional Unit
kcal	:	Kilo calories
kg	:	Kilogram
L	:	Liter
LDL	:	Low Density Lipoprotein
meq	:	Mili equivalent
mg	:	Milligram
mL	:	Milliliter
mm	:	Millimeter
MPN	:	Most Probable Number

NaOH	:	Sodium Hydroxide
NH_3	:	Ammonia
nm	:	Nanometer
No.	:	Number
NS	:	Non-significant
°C	:	Degree Celsius
pH	:	Power of Hydrogen
RH	:	Relative Humidity
rpm	:	Revolutions Per Minute
RTS	:	Ready to Serve
SE	:	Standard Error
TPC	:	Total Plate Count,
TSS	:	Total Soluble Solids
TTA	:	Total Titratable Acidity
TVC	:	Total Viable Count
Var.	:	Variety
viz.	:	*Videelict* (namely)
W/V	:	Weight/Volume basis
WHO	:	World Health Organization

INTRODUCTION

CHAPTER-I

INTRODUCTION

Over the past decade, major research emphasis in all sections of food product development is to address the changing needs of consumers and to meet the present demands of consumers by creating newer alternatives of health foods. There is growing awareness that the daily diet is an important determinate for a healthy life. Consumers judge food products not only in terms of how tasty or delicious they are, but also on the fact about how nutritious and beneficial they are to their bodies.

Increasing urbanization, globalization and more emphasis on healthy foods and diet control have accelerated these demands. One such effort is the tremendous shift in consumption of sodas, soft drinks and artificial flavored drinks to more nutritious fruit juices, probiotic drinks and milkbased beverages. In modern world, beverages are no longer considered simply to be thirst quenching or energising; consumers look for some specific functionality in their drinks, which can address to their changing lifestyles or their current health need. Recently, these changes and developments have led to newer products in the beverage market. One such major functional requirement is milk alternatives to cater to the problems of cow milk allergy, lactose intolerance, calorie concern and prevalence of hypercholesterolemia (Valencia-Flores *et al.*, 2013).

The demand for non-dairy based milk is on the rise due to a variety of reasons. Most importantly, the prevalence of lactose intolerance and milk allergy is amongst the major percentage of population (Rathee V., 2013). Lactose intolerance is a major concern mostly in the western countries but is also increasing considerably in India, besides the fact that most celebrities today, tend to the follow it as a trend.

It has also been observed that milk processors around the globe, face an uphill struggle, including the dominance of private brands; growing consumer concerns over the presence of growth hormones in the locally available milk; fluctuations in milk prices and supply shortages for organic milk. With this long list of challenges and concerns, the market has had only a few growth opportunities even though consumers continue to value milk for its nutritional benefits (Deora and Deswal., 2018).

Plant-based milk alternatives or 'vegan milk' are a rising trend, which tend to serve as an inexpensive substitute to poor economic groups of developing countries and in places, where bovine milk supply is insufficient or scarce. While majority of these milk alternatives lack a well nutritional balance when compared to bovine milk, health conscious consumers find it attractive that these milks contain functionally active components with health promoting properties.

Plant-based milk alternatives are fluids that results from breakdown of plant material (cereals, pseudo-cereals, legumes oilseeds, nuts) extracted in water and further homogenized, resulting in particle size distribution in the range of 5–20 μm which resembles cow's milk in appearance and consistency. Although, there is no exact definition and classification of these plant-based milk alternatives in literature, a general classification of the plant based milk alternatives into five categories is attempted, which is as follows:

a. Cereal based: Oat milk, Rice milk, Corn milk, Spelt milk
b. Legume based: Soy milk, Peanut milk, Lupin milk, Cowpea milk
c. Nut based: Almond milk, Coconut milk, Hazelnut milk, Pistachio milk, Walnut milk
d. Seed based: Sesame milk, Flax milk, Hemp milk, Sunflower milk
e. Pseudo-cereal based: Quinoa milk, Teff milk, Amaranth milk

(Swati Sethi *et.al.,* 2016)

Owing to its potential therapeutic benefits, oat milk is the recent emergent in the market. Oats have received wide interest due to the presence of dietary fibres, phytochemicals and high nutritive value. Health benefits of oats are associated with dietary fibres such as β-glucan, functional protein, lipid and starch components and phytochemicals present in the oat grain and that is why it is one of the promising raw materials for preparation of functional plant-based milk (Swati Sethi *et.al.,* 2016). β-glucan is a soluble fibre, which has the ability to increase the solution viscosity and can delay gastric emptying time, increases gastrointestinal transit time, thereby resulting in a reduced blood glucose level (Welch, 1995).

In the market, oat milk is available under various brand names like Oatly (Sweden), Pureharvest (Australia), Alpro (UK), Bioavena drink (Italy), Simpli (Finland), Pacific (USA) etc.

The milk is available in convenient tetra packs of various sizes and is UHT treated (Swati Sethi *et.al.*, 2016).

The oats, commonly known as *Avena sativa* belongs to the family *Poaceae*, are generally regarded as a minor cereal crop when considered in terms of grain produced annually, or areas sown for production (Wani *et.al*., 2013).

Most of the oat crop has been used as an animal feed traditionally. However, UK figures on the usage of oats, sees slightly more of the crop (44%) going towards human and industrial uses, while 38% of the feed is now going towards animal use (HGCA, 1999).

Oat requires lesser nutrients i.e. NPK (Sodium, Phosphorous and Potassium) to cultivate than that required for wheat or maize. Oat grows well in cool and moist climate and requires more moisture to produce a given unit of dry matter than all other cereals except rice (Forsberg and Reeves, 1995). Oat is predominantly grown in American and European countries, mainly Russia, Canada and USA (Ahmad *et.al*., 2010).

Oat is known for its well balanced composition. Oat is a good source of carbohydrates and high quality protein with good amino acid balance. Oat contains high percentage of oat lipids especially unsaturated fatty acid, minerals, vitamins and phytochemicals (Head *et.al*., 2010). The oat starch constitutes about 60 % of the oat grain. It is mainly a constituent of the endosperm. Oat is a potential source of low cost protein with good nutritional value. Oat is also a good source of lipids i.e. about 5.0 to 9.0 %. It contains much higher levels of lipids than other cereals which are excellent sources of energy and unsaturated fatty acids. The lipids of oats are majorly concentrated in the endosperm (Rasane *et.al*., 2015). The interest in oats is mainly aroused due to the presence of functionally active component, β-glucan which possesses nutraceutical properties. Oat fibres are also known for their choloesterol reducing effect by reducing total and LDL cholesterol (Truswell, 2002).

The oat processing include a number of steps. The oats are dehulled and groats are subsequently separated and decontaminated, prior to its processing into products. It is performed to get good quality appearance and taste. The oat milling operations include cleaning, grading, hulling, 'hull, fine and groat separation' and kilning (Zwer, 2004). After milling, pearling technology, which is also referred to as debranning and pre-processing, was originally used for polishing of rice and wheat. The pearling technology to oat facilitates separation of β-glucan-rich fractions from pericarp, aleurone and sub aleurone layers of oat (Wang *et.al*., 2007). Later, oat groats are mainly flaked. Oats for food purpose are heat treated in order to deactivate the enzymes responsible for changes in oat lipids, owing to the high amounts of lipases. The developed characteristic oat flavour in steamed oats also results in deactivation of enzymes including lipases. In a study, flaking of intact oat groat produced rolled oats of 0.5–0.8 mm thickness (Deane and Commers, 1986).

The date (*Phoenix dactylifera L.*), is a high-energy fruit, and is regarded as a popular food for thousands of years in Egypt, the Arabian Gulf peninsula and its neighboring region (Sohaimy and Hafez, 2010). Palm date is an important fruit and it is one of the oldest fruit trees to be known in the world (Marzouk and Kassem, 2011). Dates are religiously important for Muslims throughout the world (Al-Shahib and Marshall, 2003).

The date fruit is composed of a seed and fleshy pericarp constituting between 85-90% of the date fruit weight (Hussein and Alhadrami, 1998). The important quality criteria for consumers in the selection of dates is the appearance including color, size and shape, physical parameters and absence of defects, mouth feel or texture, flavor, and nutritional value (Wills *et.al*., 1998). Dates are consumed in a variety of ways. They are mainly consumed as fresh (30%-40%) or in the dried form (60%-70%) at Rutab (semiripe) and Tamar (fully ripe) stages with little or no processing (AlHooti *et.al*., 1997).

The Arabian Peninsula produces about 30% of the world's date production with an estimated area of 33% of global world acreage occupied by date palm. However, due to the inferior quality, damage, and undersized fruit of unattractive appearance, a significant portion of dates is wasted in date-producing countries (loss is about 30% of total production in Tunisia) (Besbes *et.al*., 2009).

Dates are a rich source of sugar, protein, dietary fiber, minerals and some vitamins. A high percentage of sugar in the date fruit provides a good source of rapid energy (Al Shahib and Marshall, 2003). In the recent studies it has been indicated that date fruit contains significant amount of flavonoid glycosides which include quercetin, apigenin, p-coumaric acid, ferulic acid, and sinapic acids (Hong *et.al*. 2006, Abdelhak *et.al*. 2005, Biglari *et.al*. 2008). The most important components of dates are the carbohydrates mainly sugars, which can constitute up to 78% (Makki *et.al*., 1998).

Dates contain high levels of protein compared to most of the other fruits (Al-Hooti *et.al*. 1997). The amino acid analysis of dates revealed that it contained all the essential amino acids. Date proteins are found to be rich in acidic amino acids and poor in sulfur containing amino acids such as methionine and cysteine. The amino acid content varies significantly within the same stage of maturation. The water reduction increases the amino acids content in the dried varities (Auda *et.al*., 1976).

Fiber or dietary fiber (DF) is the solid insoluble part of the date flesh, which is mainly composed of cellulose, hemicellulose, lignin, and insoluble proteins. In addition, other noncarbohydrate components like polyphenols, waxes, saponins, cutin, phytates, and resistant protein are also associated with it. The fiber content of the date fruit is very high in the early stages. However, when the fruit ripens, insoluble polymers are broken down by cellulase and pectinase enzymes present in the fruit to smaller soluble molecules (Shafiei *et.al*., 2010).

The pulp of the date contains vitamins such as riboflavin, thiamine, biotin, folic acid, and ascorbic acid that are essential for the body. Dates are also rich in B-complex vitamins, like thiamine (B1), riboflavin (B2), niacin (B3), pantothenic acid (B5), pyridoxine (B6), and folate (B9) and vitamin K (Al-Farsi and Lee, 2008). Dates contain essential minerals.

Date has been considered as a rich source of antioxidants. Antioxidants inhibit oxidative mechanisms that lead to do generative diseases such as heart disease, brain dysfunction and arthritis (Prior *et.al*., 1999). Antitumor activity, antimutagenic properties, and can lower the rate of cancers, especially pancreatic cancer and activate immune system and regulate the role of antibiotics is attributed to dates. (Ishurd and Kennedy, 2005, Mansouri *et.al*., 2005, Vayalil, 2002).

The many medicinal benefits of dates have been valued and studied for many decades. In addition to its benefits as an ideal high-energy food, in the literature, the dates are believed to have many medicinal properties such as to provide strength, fitness, and relief against a number of ailments and pains including fever, stomach disorders, memory disturbances, nervous disorders, as well as an aphrodisiac and to boost the immunity. Dates are also used for the treatment of abdominal troubles because of their high phenolic contents which acts as an astringent in intestinal troubles. Date products in the form of infusion, decoction, syrup, or paste are administered as a treatment for sore throat and colds due to the antioxidant and anti-inflammatory properties of the date fruit. Dates are also used as a renal restorative and their regular consumption can prevent the formation of renal calculi due to its diuretic and anti-inflammatory actions. The most common medicinal use of dates and its products is as a tonic, especially for women who are close to delivery and at postpartum stage which can act as a tonic to strengthen the uterine muscles. They not only help in activating the delivery process but also prevent the post delivery bleeding due to the presence of some constricting substances (Amanat Ali *et.al.,* 2012).

This study is aimed to develop an instant RTS mix of oat milk powder fortified with dates. The aim is to increase its nutritional value and acceptability under the following objectives

OBJECTIVES

1. To study the physicochemical properties of oat milk powder fortified with dates
2. To carry out standardization and characterization of the oat milk powder fortified with dates as well as the developed oat milk
3. To evaluate the organoleptic characteristics of the developed oat milk powder fortified with dates
4. To study the shelf life and packaging requirements of the developed oat milk powder fortified with dates
5. To access the techno-economical feasibility of oat milk powder fortified with dates

REVIEW OF LITERATURE

Chapter- II

REVIEW OF LITERATURE

In recent years, demand for oat based products has been increased due to rapidly increasing knowledge about the many health benefits of oats. Increased consumer awareness towards health has greatly emphasized on the intake of high fibre diets. A comprehensive review was conducted to gather information about the development of technology for production of oat milk powder fortified with dates.

Previous researches conducted on oat milk showcased the development of liquid oat milk produced from oat slurry by enzymatic treatment on the oat starch. The developed oat milk was pasteurised and packed in aseptic packages like tetra packs which may become a source of obstacle for small scale industries. Moreover, despite of its heath benefits, oat milk lacks calcium which is an essential nutrient for the growth and development;therefore, it needs to be fortified.

In the present research, project work is focused on the development of complete standardized method for preparation of an oat based milk powder formula which is fortified with dates to make an instant mix.

For accomplishment, efforts have been taken in this direction like identification of varieties of dates, standardization of production process, physicochemical and microbiological characteristics of final product etc. This chapter deals with comprehensive review of literature relevant to study. A review of literature is an essential part of any scientific investigation with the sole purpose to determine the previous work done and to assist in the description of the objectives of hypothesis and research procedures to be followed. The work done on this topic elsewhere in scientific literature is reviewed here:

1. Physicochemical properties of oats
2. Physicochemical properties of dates
3. Standardized process technology for oat milk powder fortified with dates

4. Organoleptic characterization of oat milk powder fortified with dates
5. Health benefits of oat milk powder fortified with dates
6. Shelf life study of oat milk powder fortified with dates
7. Microbial analysis of oat milk powder fortified with dates
8. Packaging requirements of oat milk powder fortified with dates

2.1 Physicochemical properties of oats

The soluble fibre of oats, as reported by Anderson (1986) to reduce high blood cholesterol levels, triglyceride, and glucose levels. Oats also good sources of insoluble fibre functions as a water holding capacity agent and can reduce intestinal transit time when present in adequate amounts.

Oat has a well-balanced nutritional composition. Hoover and Vasanthan (1992) carried out a comparative study on the characteristics of oat starches with other cereal starches and reported that the oat starches showed higher swelling factor, decreased leaching of amylose, higher peak viscosity and set back, low gel rigidity, greater acid hydrolysis susceptibility, greater α-amylase action resistance and high free-thaw stability.

Karppinen *et.al*., (2000) studied that arabinoxylans and β-glucans are the most important cereal polysaccharides which are partially water-soluble and have an effect on various food preparations. These polysaccharides are known to reduce symptoms of diseases like diabetes, atherosclerosis and colon cancer.

Skendi *et.al*., (2003) reported that oats are an excellent source of different dietary fibre components of mixed-linkage (1→3), (1→4)-β-D-glucan, arabinoxylans and cellulose and the neutral cell wall of β-glucan has great functional and nutritional properties.

Fulvia *et.al*., (2013) determined the chemical composition of some oat samples and concluded that oat contain moisture about 10.39-11.10%, a protein content of 14.31-21.31%, ash content of about 1.50-2.34%, crude fiber 1.90-4.36%, total fat ranges from 4.46-6.70% and total carbohydrates from 60.92-68.78%.

According to Ovando-Martinez *et.al*., (2013), on the basis of digestion rate oat starch has been classified into three fractions, i.e. rapidly digestible starch (RDS), slowly digestible starch (SDS) and resistant starch (RS) where the slow rate starch digestibility is important for human health to maintain balanced blood glucose levels. According to this study, oats contain approximately 7% RDS, 22% SDS and 25% RS of the total starch.

According to the work carried out by Brunava *et.al*., (2014), The β-glucans content of oat grain samples ranged from 2.7g to 3.5g per 100 grams of oat grains.

Rasane *et.al*., (2015) reported that oat contains 60% starch, 11-15% total protein, 5-9% lipids, 2.3-8.5 % dietary fibre and 0.54 % calcium. The study further states that oat is considered to be a potential source of low cost protein with good nutritional value. The study also showed that oat contains smaller quantities of prolamins (15%) in relation to the higher amounts of globulins (80%) of the total oat protein.

According to Vita Sterna *et.al*., (2016), oat is well known as a healthy food containing significant amounts of soluble dietary fibre, β-glucans, vitamin E and PUFAs in the world. The aim of her study was to characterise the biochemical composition of husked and naked oat varieties. The content of protein, fat, vitamin E and also the composition of amino acids, fatty acids and dietary fibre were determined. The obtained results showed a wide range of fat content among varieties, which varied from 4.9 to 10.5g per 100 grams, whereas the α-tocopherol content in oat grain was found to be 4.5-2.3mg/kg, the sum of essential amino acids was 35-45g/kg and unsaturated fatty acids was estimated to be 78-81.5% of total fatty acids content.

Solanki *et.al*., (2019) conducted the physical analysis of oat grain samples and found out that with increase of moisture content, the average length from 7.5354 to 8.6252 mm, width 2.3144 to 2.5446 mm, thickness 2.3144 to 2.5446 mm, volume from 8.0972 to 28.7557 mm^3 and geometric mean diameter from 2.99 to 4.5766 mm of oat grains increased significantly.

2.2 Physicochemical properties of dates

According to Spiller (1993), in the Handbook of Dietary Fiber in Human Nutrition the dietary fibre of dates was reported as being 4.4% (3.2% insoluble bran and 1.2% soluble fiber).

Al-Hooti *et.al*., (1997) investigated that dates contain high levels of protein compared to most other fruits where the highest content is observed during Kimri phase (5.5-6.4%), which gradually decreases to 2-2.5% during the Tamar stage or fully ripened satge. According to the study, the flesh of date also contains 0.2-0.5% oil.

As per the USDA National nutrient database (2007), a 100g of dates contain on average of 0.8μg selenium, 0.3μg copper, 864 mg potassium, and 43 mg magnesium in comparison with other dried fruits.

El-Sohaimi *et.al*., (2010) reported that matured fruits of dates contain a low moisture percentage ranging from 15 to 20% and high percentage of sugar 65 to 70% where sucrose is the major component.

Aslam *et.al*., (2011) carried out quantitative analysis of water-soluble vitamins (B1, B2, B3, B5, B6, B9, B12) which showed a significant variation within the different cultivars and the developing stages of date fruit. Vitamins B1, B3, B5, B6 were observed to be the highest in mature stages, whereas the vitamins B2, B9, B12 were detected in immature fruit.

The chemical composition of three date palm varities (Salma, Magdy and Khalas) were determined by Ahmed *et.al*., (2017). The obtained results showed that the moisture content ranged between 43.5-45.5%. The protein content, ash content, reducing sugars and non-reducing sugars content of dates ranged between 2.21-3.02, 1.69-1.85, 23.03-28.18 and 51.87-59.22%, respectively.

Mahawar *et.al*., (2017), determined the physical properties of two date varities; Khadrawy and Medjool. The three principal dimensions i.e. length, width and thickness of date palm fruits of both Khadrawy and Medjool were determined to be 41.75±2.41mm, 19.46±1.08 mm, 19.54±1.09 mm and 48.26±2.49 mm, 29.05±1.75 mm, 28.80±1.56 respectively. Further, the bulk density of Medjool was 0.59±0.014g/cm^3 which was slightly higher than of that of Khadrawy i.e. 0.56±0.017 g/cm^3. True density of Khadrawy was about 0.93±0.002 g/cm^3 while that of Medjool was 0.85±0.015g/cm^3. The porosity values of date palm fruits decreased from 0.40±0.01 to 0.34±0.007 with Khadrawy and Medjool date palm, respectively.

2.3 Standardized process technology for milk powder

Mohd. AH (1985), carried out the production of coconut milk powder. According to the study, the production of coconut milk powder can be carried out despite the high fat content of coconut milk. The spray drying process was carried out with the inlet temperature of 190°C and outlet temperature of 90°C with the addition of additives like skim milk powder and maltodextrin and can be reconstituted easily with water.

Inglett GF (1991), stated that a US patent has disclosed the preparation of water-soluble oat dietary fiber compositions by treating ground oat products with α-amylases which serves to thin the oat starch. The produced liquid dietary fiber compositions are used as additives in food products, such as fat substitutes.

Ahlden I *et.al.,* (1997), carried out an enzymatic process, wherein whole oat groats are converted into liquid milk like product involving a number of steps like flaking, wet milling, amylase hydrolysis, decanting, formulation, UHT treatment and aseptic packaging.

According to Ahlden I *et.al.*, another US patent (No. 5,686,123) revealed a homogeneous and stable cereal suspension is a milky product, which can be used as an alternative to milk, especially for lactose-intolerant people, having the taste and aroma of that of natural oats.

Aastha Deswal *et.al.*, (2012) conducted experiments in which processing of oat milk was made by rolled oats which were ground into a laboratory food processor to produce finely granulated oat flour and an oat slurry was formed using water. Calcium chloride was added as a catalyst for the enzyme at a concentration of 0.04 % (w/w). The samples were treated with different enzyme concentration and liquefaction time for optimization of the oat milk manufacturing process at 75 °C. It was then filtered through muslin cloth to get the oat milk. The enzyme was inactivated by heating at 100°C for 5 min at the end of the treatment. The percentage of filtrate obtained was estimated as the total yield.

A process of producing soy milk powder was developed by IL Pardeshi *et.al*., (2014), were the purpose was to avoid the need of bottling, pasteurization, and refrigerated storage of soymilk. In this process, the soybeans were soaked in water and rinsing for 6 to 8 times at regular interval of 6 hours. The beans were then grinded with addition of water followed by heating for 20 mins. The feed rate was kept at 230-480 g/h, and an inlet drying temperature of 160-200°C was used for spray drying of soymilk. It was observed that the optimal spray dried soymilk powder could be prepared at feed rate 350-375 g/h and 178-182°C inlet drying air temperature.

Bernat N *et.al.,* (2015), process involved soaking and grinding of whole oats followed by homogenization.

Sangami and Radhai (2018), carried out extraction technique of oat milk using various forms of oats including whole grain, split grain and oat meal. The emphasis was put on varying the quantity of oats to be used, soaking method and duration of soaking and the samples were subjected to yield analysis. The yield of oat milk obtained ranged from 55 to 70 per cent. Dietary fiber content ranged from 18.36 – 18.70 g and β-glucan content ranged from 2.28 - 3.44 g per cent. The product had a pH of 4.38, titrable acidity was around 0.34 per cent, total solids of 32.04 per cent whereas the viscosity ranged from 3 – 3.5 cPS.

2.4. Organoleptic characterization of milk powder

Singh and Bains, (1998) observed that blending of plant-based milk with cow milk had better taste scores than plant-based milk alone.

The extraction process of oat milk carried out by Sangami and Radhai (2018), obtained mean scores for organoleptic evaluation of the oat milk extract samples which indicate that the variation with both roasted and unroasted split oats grain recorded highest scores of 7.7, 8.7 and 8.8 in the respective attributes of appearance, odour and texture/consistency respectively. The scores indicate that the extracted oats milk was found to be in the range of Liked moderately (Score 7) to Liked very much (Score 8).

2.5. Health benefits of milk powder

Önning *et.al*., (1998), observed that oat milk has been reported to decrease the plasma cholesterol and Low Density Lipoprotein (LDL) concentrations of healthy individuals after a consumption period of 4 weeks.

Biörklund M *et.al.*, (2005), reported that compared to a control beverage, 5g of β-glucans from oat milk significantly lowered total-cholesterol by 7.4% ($p < 0.01$), and post prandial concentrations of glucose and insulin.

Lyly M *et.*al., (2009), reported that a group of scientists investigated the effect of oat fibres on observed satiety of beverages and reported that the beverage with added oat fibers showed a trend of having a higher satiety index and decreased the 'desire to eat something', increased fullness and showed a trend of more than the beverage without fiber.

Juvonen KR *et.al.*, (2009), also found out similar results and reported that the consumption of viscous fibers, including β-glucan in oat bran, favorably affect satiety as well as post prandial carbohydrate and lipid metabolism.

2.6. Shelf life study of milk powder

Civille and Szczesniak, (1973), observed that stability of plant-based milk is dependent on the size of dispersed phase particles.These colloidal systems formed by large sized dispersed particles such as fat globules, solid particles from raw materials, proteins and starch granules make it difficult to obtain a stable product which can be stored, not for very long time due to sedimentation or settling of solid particles. The presence of large size particles results in a sandy, gritty or chalky mouth feel and lack of creaminess due to their low fat content which renders the milk instable.

Kwok and Niranjan (1995), reported that plant-based milk are a rich source of nutrients and serve as an ideal medium for growth of micro-organisms, and therefore, its quality may be adversely affected by the rapid growth of micro-organisms. To extend the shelf life of food products thermal treatment has been used as a processing method to eliminate or reduce spoilage and pathogenic micro-organisms. Heat treatment was used to increase the shelf life of plant-based milk along with the objectives of increasing total solids yield and improvement of flavor.

Cruz *et.al.*, (2007); Singh (2013), inferred that thermal treatments may be well applied to plant-based milk such as soy milk and peanut milk but due to the presence of high starch concentration in oat milk, rice milk etc. its application is limited, and therefore, non-thermal processing technologies must be applied to extend the shelf-life.

Cruz *et.al.,* (2007), reported that apart from achieving size reduction of colloidal particles, a simultaneous destruction of micro-organisms by UHPH treatment due to the effects of highpressure on micro-organisms can be achieved simultaneously. Two UHPH treatments, 200 and 300 MPa, were applied to the soy milk at an inlet temperature of 40 °C and was compared with normal and UHT treated soy milk. Results indicated that UHPH treatment significantly improved the emulsion stability by disrupting the colloidal particles and also showed a reduction in the microbial load.

According to Valencia-Flores *et.al*., (2013), Ultra High Pressure Homogenization (UHPH) is one of the promising processing technologies which resulted in smaller and more uniform sized particles, and can be effectively utilized to improve the stability of plant-based milk.

According to Swati Sethi *et.al.*, (2016), the stability of plant-based milk can be improved by reducing the size of dispersed phase particles by using various techniques. Traditionally, colloid mill was used to reduce the size of dispersed phase particles in preparation of soy milk and peanut milk. But, microbial spoilage can be a limiting factor in the use of colloid mill for size reduction and there is a need for the milk to be pasteurized further from food safety point of view. It was also reported that the stability of plant milk can also be improved by using emulsifiers and stabilizers.

2.7. Microbial analysis of milk powder

Kajs *et.al*., (2006), reported that spray-dried and intermediate moisture products prepared from coconut skim milk contained low levels of viable microorganisms (3,100–13,000, mean 6,600/g). There was little change in the bacterial counts of these products during storage for 90– 120 days at either 21 or 35°C. When five microbial species were inoculated individually into an intermediate moisture product with a moisture level of 30.7%, no increase in population took place at 35°C for 25 days.

B.C. Adebayo-Tayo *et.al,* (2008), evaluated branded and unbranded powdered soymilks for microbial quality, physicochemical parameters and aflatoxin level. The total bacterial count ranged from 4×10^4-1.1×10^5 cfu/g for branded samples and 2.0×10^4-7.2×10^4 cfu/g for unbranded powdered soymilk samples. There was no presence of Coliform bacteria in both branded and unbranded powdered soymilk samples. The fungal count ranged from 2.1×10^4 to 4.9×10^4 cfu/g for branded soymilk powder and 1.5×10^4 to 2.6×10^4 cfu/g for unbranded soymilk powder. The branded soymilk powder included isolated microorganisms like Micrococcus sp., *Staphylococcus aureus*, *Bacillus subtilis*, Pseudomonas sp., Streptococcus sp., *Aspergillus flavus*, *Candida pseudotropicalis*, *Saccharomyces cerevisae*, *Penicillium citrium* and Cladosporium sp. while Pseudomonas sp., *B. subtilis*, *Yersinia enterocolitica*, Streptococcus sp., *A. flavus*, *Aspergillus niger*, *C. pseudotropicalis*, Saccharomyces sp. were isolated from unbranded samples.

Thombare D.T *et.al.*, (2015), carried out IMVIC test, Catalase test and gram staining for the microbial analysis of soymilk which showed presence of bacteria like Escherichia spp and Streptococcus spp.

Ravjindamba *et.al*., (2016), evaluated stability of a fermented oat beverage. The viable cell counts were about 6×10^9 CFU/ml after the fermentation process which reduced to 4×10^7 CFU/ml after a storage period of 21 days.

Edet *et.al.,* (2017), carried out a study evaluating the microbiological and biochemical content of soymilk samples from five different areas. The probable bacterial isolates identified were Pseudomonas, Klebsiella and Bacillus species, while the fungi species were that of Yeast and Aspergillus. The bacterial counts ranged from $4.80\pm0.04x10^8$ cfu/ml to $6.90\pm0.01x10^8$ cfu/ml while the mean fungal count ranged from $3.50\pm0.22x10^8$ cfu/ml to $6.50\pm0.17x10^8$ cfu/ml.

2.8. Packaging requirements of milk powder

Hassan (1985), carried out production of coconut milk powder. In his preliminary storage studies, it was found that after storage for three months in ordinary polyethylene packaging under room conditions, samples were found to be stable and there was no separation of phases.

Laurie *et.al*., (2015), analysed five different packaging treatments were over a 36-day period to determine the protection of soymilk from photo-oxidation. The packaging bottles made of different packaging materials were stored in a refrigerated display case for 36 days and evaluated weekly. It was found that soymilk packaged in high LPA (Light Protective Additives) bottles protected from developing light-oxidized off-flavors and odors for a period of 15 days and also provided protection for riboflavin and controlled development of photooxidative products for approximately 29 days.

MATERIALS AND METHOD

Chapter- III

MATERIALS AND METHOD

The present investigation entitled "Studies on development of vegan milk powder (oat milk powder) fortified with dates" was carried out in the Department of Food Chemistry and Nutrition, with collaboration of Department of Food Process Technology and Department of Food Microbiology and Safety, Niche Laboratory, College of Food Technology, and Department of Soil Science, College of Agriculture, V.N.M.K.V., Parbhani during the academic year 2019-20. Materials used and methods adopted for the present investigation are presented under suitable headings and subheadings.

3.1 MATERIALS

3.1.1 Raw materials

Rolled oats were purchased online and fresh dates were procured from the local market of Parbhani, and other materials like maltodextrin, were procured from the Department of Food Chemistry and Nutrition, College of Food Technology, Parbhani.

3.1.2. Chemical and Reagents

Most of the chemicals used in this investigation were of analytical grade. They were obtained from Department of Food Chemistry and Nutrition, Department of Food Process Technology, Department of Food Engineering and Department of Food Microbiology and Safety, College of Food Technology, VNMKV, Parbhani.

3.1.3. Equipments and Instruments

Different equipments required for the preparation of oat milk powder fortified with dates and its physicochemical analysis were Texture Analyzer, Digital Model of Vernier caliper and Electronic balance with the accuracy of 0.0001g for weight measurements, Digital pH meter for pH measurement. These were made available from the Department of

Food Chemistry and Nutrtion Lab and other Departments of College of Food Technology, V.N.M.K.V., Parbhani.

The various other processing and analytical equipments included Hot air oven, Soxhlet apparatus, Micro Kjeldhal apparatus, Spray dryer, Muffle furnace, Grinder, Spectrophotometer, laminar air flow cabinet and glasswares which were used from Department of Food Chemistry and Nutrition, Department of Food Engineering, Department of Food Microbiology and Safety and Department of Food Process Technology, Niche area laboratory, College of Food Technology, VNMKV, Parbhani.

3.1.4 Packaging material

The suitable packaging material such as High Density Poly Ethylene (HDPE) bags and Aluminated standing pouches for containment and safe storage of products were procured from local market of Parbhani.

3.2. METHODS

Oats, dates and prepared milk powder were analyzed for various quality attributes including physical attributes and proximate chemical composition. Physicochemical characteristics were estimated using recommended standard of Association of Official Analytical Chemists (AOAC) methods (2000) as mentioned below.

3.2.1. Physical properties of oats and dates

3.2.1.1. Thousand kernel weight

Thousand kernel weight was measured by counting 100 randomly selected grains and weighing them using an electronic balance having an accuracy of 0.001 g and then multiplied by 10 to give mass of 1000 seeds.

3.2.1.2. Bulk density (ρ_b)

25 g of sound grains will be weighed on the digital weighing balance and filled into the measuring cylinder earlier filled with reference solution of kerosene or toluene. The increase in the level of liquid will be measured after adding the grains. It is bulk density represented in g/ml (Dutta *et.al*., 1988).

$$\text{Bulk Density } (\rho_b) = \frac{\text{Weight of grains}}{\text{Volume of grains}}$$

3.2.1.3. True density (ρ_t)

25 g of grains will be filled into the measuring cylinder and volume occupied by them will be measured. It will be then calculated by following formula and represented in g/ml (Rooney *et.al*., 1982).

$$\text{True Density } (\rho_t) = \frac{\text{Weight of grains}}{\text{Volume occupied}}$$

3.2.1.4. Angle of repose

The frictional property such as angle of repose (θ) was calculated from the height and diameter of the naturally formed heap of the food grains on a circular plate as method described by Singh *et.al.,* (2013).

$$\text{Angle of repose } (\theta) = \tan^{-1}\{h/r\}$$

Where, h = Height of heap

r = Radius of base of heap

3.2.1.5. Porosity (ε)

The porosity (ε) of the bulk grain was defined as the fractions of the space in the bulk grain that is not occupied by the grain. The porosity, ε was calculated using formula

$$\varepsilon = \frac{(1 - \rho_b) \times 100}{\rho_t}$$

Where, ρ_b and ρ_t are the bulk and true density, respectively in k/gm^3

3.2.1.6. Determination of viscosity

The viscosity of oat milk was measured by using Brookfield viscometer.

3.2.1.7. Determination of Bulk Density of powdered sample

The bulk density of powdered sample was measured by the method suggested by Goula and Adamopoulos. One gram of sample was weighed and filled in a 10 ml graduated cylinder. The cylinder was tapped by hand five times from a height of 10 cm and the bulk density was calculated as the ratio between the mass of powder contained in the cylinder and the volume occupied.

3.2.1.8. Determination of Wettability of powdered sample

The wettability of the powdered sample was measured by the time required for 1.0 g of sample deposited on the liquid surface (400 g of distilled water) to become completely submersed at 25°C (Vissotto *et.al.*, 2010)

3.2.1.9. Determination of Solubility Index (SI) of powdered sample

Solubility index of the powdered sample was determined by the method explained by Cortes Rojas and Oliveira. About 1 g of the powder was added to 100 ml of distilled water The mixture was stirred using a magnetic stirrer. After keeping the mixture at 37°C for 30 min, the mixture was centrifuged at 10,000 rpm for 15 min. The precipitate was transferred into a drying vessel and dried at 105°C until constant weight. The solubility index (%) was calculated as the ratio of the solid mass of supernatant to the amount of the original weight of sample (1g).

3.3 Proximate analysis

3.3.1. Determination of pH

The pH was determined by using a digital pH meter after standardizing it with buffers of pH 4.0 and 9.0.

3.3.2. Determination of moisture content

Moisture was estimated by accurately weighing 5g of the sample which was subjected to oven drying at 105°C for 4 hrs. It was again weighed after cooling in desiccator until constant weight was achieved. The resultant loss in weight was calculated as moisture content (AOAC, 2005).

$$\% \text{ Moisture } = \frac{\text{Initial weight} - \text{final weight}}{\text{Total weight of sample}} \text{ X } 100$$

3.3.3. Determination of Crude Fat

The method employed was that of solvent extraction using a Soxhlet extraction as described in method No. 30-10 of AACC (2000). 2 g of sample was taken in a thimble and placed in extraction tube of Soxhlet apparatus. About 250 ml of hexane was added in 500 ml bottom flask of the apparatus and connected to Soxhlet apparatus. The fat was extracted by running hexane over the sample at the rate of 3-4 drops per sec for about 5 hr. The solvent was recovered and the flask was kept in hot air oven for 10 min at 40-50ºC. The flask was cooled in desiccator and weighed. Fat percentage was calculated according to the following formula.

$$\% \text{ Crude Fat } = \frac{\text{Final weight of flask} - \text{Empty weight of flask}}{\text{Weight of sample}} \times 100$$

3.3.4. Determination of crude protein

The crude protein was determined by the Kjeldhal method as described in method No. 46-10 of (AACC, 2000). This is based on the fact that on digestion with concentrated sulphuric acid and catalysts, organic compounds are oxidized, and the nitrogen is converted to ammonium sulphate. Upon making the reaction mixture alkaline, ammonia is liberated, removed by the steam distillation, collected and titrated.

3.3.4.1. Digestion

Accurately weighed 200 mg of defatted ground sample and add a pinch of catalyst mixture K_2SO_4:$CuSO_4$:HgO red (91:8.2:0.8g) then transferred into the digestion flask, digestion was carried out with 5 ml concentrated H_2SO_4 for 2-3hr at 45ºC till the content becomes colorless.

3.3.4.2. Neutralization and Distillation

Digested sample was diluted to the 50 ml in volumetric flask and made final volume to 50ml with double glassed distilled water. Then the 5ml of aliquot was neutralized with 40% NaOH containing 5g of sodium thiosulphtate. Distillation was carried and liberated ammonia was absorbed in 2% boric acid solution containing methyl red as indicator.

3.3.4.3. Titration

The collected ammonia was titrated against 0.01N H_2SO_4. Titre reading was noted, % Nitrogen was calculated by using following formula and % protein was calculated by multiplying with 6.25 i.e. the conversion factor from % Nitrogen to % Protein. Simultaneously a blank sample was also carried out.

$$\% N = \frac{\text{CBR x Normality of } H_2SO_4 \text{ x 0.014 x D.F}}{\text{Wt. of sample (g)}} \text{ X } 100$$

Where, CBR= (Sample Burette Reading – Blank Burette Reading)

$$\% \text{ Protein} = \% \text{ Nitrogen} \times 6.25$$

3.3.5. Determination of Ash

The ash content was determined as a total inorganic matter by incineration of the samples at 550-600ºC according to method No. 08-01 of AACC (2000). Remaining inorganic materials are reduced to their most stable form, oxides or sulphates and are considered as “ash”.

Procedure

Oven dried 5 g sample was taken in a pre-weighed silica crucible and charred on the burner. Then it was ignited in the muffle furnace at 550-600ºC for 5-6 hours or till to get constant weight of greyish ash. The ash of sample was calculated with the following formula.

$$\% \text{ Ash} = \frac{\text{Weight of crucible with ash} - \text{Weight of empty crucible}}{\text{Total weight of sample}} \times 100$$

3.3.6. Determination of Crude Fiber

About 2 to 5 g of moisture and fat free samples were weighed into 500 ml beaker and 200 ml boiling 0.255 N (1.25 w/v) H_2SO_4 was added. The mixture was boiled for 30 minutes keeping the volume constant by addition of water at frequent intervals. At the end of this period, the mixture was filtered through a filter paper and the residue washed with hot water till free from acid. The material then transferred to the same beaker and 200 ml of boiling 0.313 N NaOH solution added. After boiling for 30 minutes the mixture was filtered through filter paper. The residue was washed with hot water till free from alkali followed with some alcohol. It was then transferred to crucible dried overnight at 80-100^{0}C and weighed. The crucible was heated in a muffle furnace at 550600^{0}C for 4 hours cooled and weighed again. The difference in the weights represented the weight of the crude fiber (AOAC, 2005).

$$\% \text{ Crude Fiber } = \frac{\text{Weight of residue} - \text{Weight of ash}}{\text{Weight of sample}} \times 100$$

3.3.7. Total Carbohydrates

Carbohydrates were estimated by phenol H_2SO_4 method (AOAC, 2000). 100 mg of sample was taken in conical flask added with 5 ml of 95% Sulfuric acid, stirred well for 5 min. in ice bath to avoid burning of sample. Then the volume of solution was made up to 25 ml with distilled water. The sample was heated in boiling water bath for 3 hr. using condenser. It was then filtered through Whatman No. 1 filter paper and filtrate was made up to 50 ml. from the filtrate 1 ml of sample was taken for color development. A total carbohydrate was detected by phenol-sulfuric method at 480 nm from standard calibration curve of D-glucose.

3.4. Estimation of minerals

Minerals like calcium, magnesium, potassium, phosphorous and iron were determined by using titration and spectrophotometric method (Atomic Absorption Spectrophotometer).

3.4.1. Preparation of mineral solution

The ash obtained was moistened with glass distilled water (0.5-1 ml) and concentrated HCl was added and evaporated to dryness on a boiling water bath. Again 5 ml concentrated HCl was added and evaporated to dryness as before. Lastly 4 ml of HCl and 5 ml of distilled water were added. This solution was warmed over a boiling water bath and filtered into the 100 ml volumetric flask using Whatman no.4 filter paper. After cooling the volume was made to 100 ml using distilled water and suitable aliqoute was used for the estimation of calcium and iron.

3.4.2. Determination of Calcium

Reagents

1) Ammonium oxalate
2) Conc. ammonia solution (25 % v/v)
3) Methyl red indicator
4) Sulphuric acid (2N)
5) N/100 $KMNO_4$ solution

25 ml mineral solution was diluted to 150ml with distilled water and neutralized with ammonia solution using methyl red as indicator till pink colour changes to yellow. Further the solution was boiled and 10 ml of 6 per cent ammonium oxalate was added. This mixture was boiled for few minutes and added with conc. glacial acetic acid (99.5 per cent) till the colour was distinctly pink. The mixture was kept aside in warm place (overnight) and when precipitate settled down, the supernatant was tested with a drop of ammonium oxalate to ensure the completion of precipitation. The contents were filtered through Whatman no.4 filter paper and given washings of warm distilled water. The precipitate was transferred to a beaker by making a hole in the centre of filter paper and by giving washings of sulphuric acid (2N, 5ml) twice. Then the solution was heated to 70ºC and titrated against N/100 $KMNO_4$. Simultaneously a blank was also run. The results were expressed as mg calcium/100g sample (Ranganna, 1986).

Formula: 1ml of 0.01N $KMnO_4$ = 0.2004mg Calcium

3.4.3. Determination of Iron

Iron content was determined by a-a, dipyridyl method described in AOAC (2005) exactly 10 ml of wet digested sample solution was pipetted into volumetric flask of 25ml capacity in triplicates. 1ml of hydroxylamine hydrochloride solution, 5 ml of acetate buffer solution and 2 ml of a-a, dipyridly solution were added into each volumetric flask. The volume was made up to 25 ml with glass distilled water and the content was mixed will. The intensity of the colour developed was read in spetronic 20 at 510 nm. Iron content of the digested sample solution was read from the standard curve of known concentration of iron.

Preparation of standard curve

Pipette 0.0, 0.5, 1.0, 1.5, 2.0, 3.0 and 4.0 ml of Fe standard solution into a series of 25 ml volumetric flasks and add to each of them exactly 0.2 ml of conc. HCl. Dilute each of them to exactly 10 ml with water, and then add reagents in the same way as for the sample, Plot the quantity of Fe (in mg) against the absorbance (I.C.M.R, 1990).

Iron content of sample (mg Fe / 100 g sample) =

$$= \frac{\text{Quantity of Fe in aliquot of ash solution (from calibration curve)}}{\text{Aliquot of ash solution taken for determination}} \times \frac{\text{Total volume of ash solution}}{\text{Wt. of the sample taken for ashing}} \times 100$$

3.4.4. Determination of Magnesium

Magnesium was estimated by colorimetric method. Measure 10 ml of ash solution into a 15 ml graduated centrifuge tube. Add 1 drop of methyl red indicator. Neutralize solution with NH_4OH and ammonium oxalate and make the solution to a volume of 13 ml. Mix and allow to stand overnight. Centrifuge for 10 min. and discard precipitate. Measure 1 ml of the supernatant liquid from above into a 15 ml centrifuge tube. Add 3 ml of water, 1 ml of ammonium phosphate and 2 ml of NH_4OH. Mix and allow standing overnight. Centrifuge for 7 min, discard the supernatant liquid, mix with 5 ml of dilute NH_4OH, centrifuge for 7 min and discard supernatant liquid. Dry the precipitate by placing the tube to container of hot water. Add 1 ml of dilute HCl and 5 ml of water to dissolve the precipitate. Add 1 ml of molybdic acid solution, 0.5 ml hydroquinone and 0.5 ml sodium sulphite solution. Mix and allow to stand for 30 min. Transfer the solution to colorimeter tube and read the absorbance in a colorimeter using a No. 66 red filter.

Set the instrument scale at zero with scale (Ranganna, 1986).

3.4.5. Determination of Potassium

It was determined by using flame photometric technique. The sample for potassium estimation was digested using $HClO_4$ and HNO_3. The 0.5 g sample was taken. In that, 5 ml HNO_3 was added and kept overnight. Next day, again 5 ml HNO_3 was added and sample digested by boiling on gas burner. Boiling continued till colour changes to colourless. The volume digested made 100 ml by adding distilled water and potassium was estimated by flame photometer (Ranganna, 1986).

$$\% \text{ Potassium} = \text{Reading (ppm)} \times \frac{\text{volume of digest}}{\text{weight of sample}} \times \frac{100}{1000000}$$

3.4.6 Estimation of phosphorous

It was done by spectrophotometric method. 5 ml of ash solution was obtained by dry ashing. 5 ml of molybdate reagent was added into ash solution and mix. Then 2 ml of aminonaphthol sulphonic acid solution was mixed and made volume up to 50 ml. A similar blank was prepared using water in place of sample. The solutions were allowed to stand for 10 minute and colour was measured at 650 nm by setting the blank at 100 per cent transmission.

$$\text{Phosphorous (mg/100g)} = \frac{\text{mg of phosphorous taken in for estimation} \times \text{total volume of ash solution} \times 100}{\text{ml of ash solution taken for estimation} \times \text{weight of sample}}$$

3.5. Methodology

3.5.1. Preparation of oat milk powder fortified with dates

For the preparation of oat milk powder, the rolled oats were soaked in water in the ratio of 1:2.7 and the oats were grinded to a slurry. Enzymatic hydrolysis with an enzyme concentration of 77.87mg/kg of oats, using α-amylase at the rate of was carried out to breakdown the complex starches into simpler sugars. The slurry was filtered and heated to deactivate the enzymes. The prepared oat milk was spray dryed at 120°C with the addition of maltodextrin at the rate of 15%. In the same way, the dates (after removing the seed and cap), were soaked in sufficient amount of hot water and ground. The mixture was screened and maltodextrin was added and spray dried. For further processing, the spray dried oat and dates powder were combined and standardized. For further processing, hot water was added for reconstitution to the prepared RTS powder and mixed thoroughly.

3.5.2. Preparation of oat milk powder with incorporation of different levels of dates powder

Spray dryed oat milk powder and dates powder were mixed in various ratios. Sensory analysis was carried out to determine the most acceptable milk powder. The addition of dates powder was done with the aim to impart a natural sweetness and also to increase the nutritional value of the oat milk. Vanilla flavor (2%) was added to mask the starchy flavour of the oats and to impart a fresh flavour. Additives like maltodextrin was added to provide bulk or body to the milk before spray drying and also to increase the yield of milk powder after drying.

(T_0) = Control without the addition of dates powder

(T_1) = With addition of 10% of dates powder

(T_2) = With addition of 20% of dates powder

(T_3) = With addition of 30% of dates powder

(T_4) = With addition of 40% of dates powder

(T_5) = With addition of 50% of dates powder

Table 1: Formulation of Oat milk powder fortified with dates mix

Sample	Oats powder (%)	Dates powder (%)
T_0	100	-
T_1	90	10
T_2	80	20
T_3	70	30
T_4	60	40
T_5	50	50

3.5.3. Methodology for preparation of oat milk powder fortified with dates

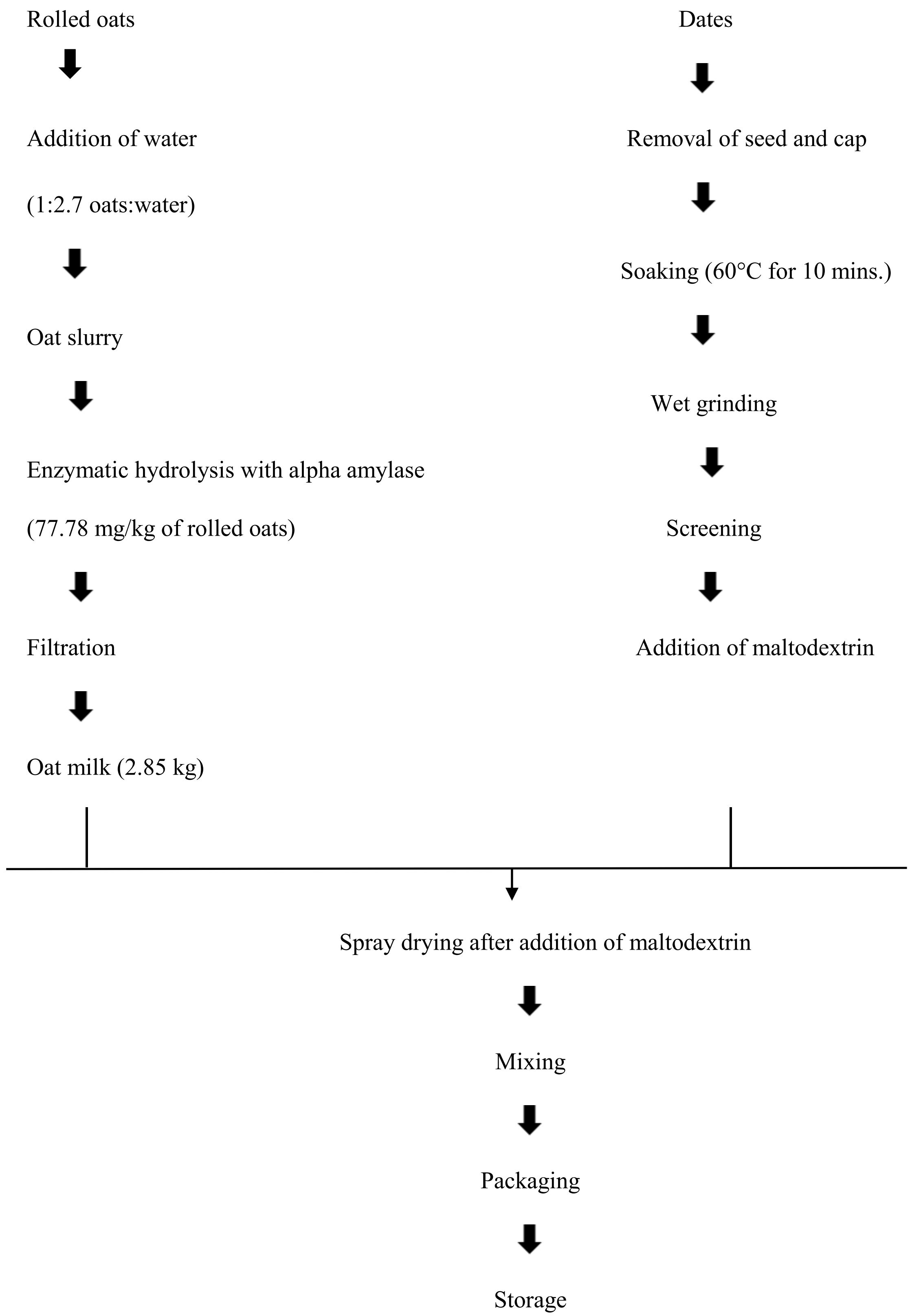

Fig 1: Process flowchart for preparation of oat milk powder mix

Plate no. 1: Spray dryer

3.5.4. Standardized recipe for the preparation of oat milk powder fortified with dates

Spray dryed oat milk powder and dates powder were mixed and standardized. The oat milk powder was considered as a control sample. The various ratios of oat milk powder and dates powder used as test samples were (90:10), (80:20), (70:30), (60:40) and (50:50). The levels of maltodextrin and other additives were kept constant for each ratio.

Table 2: Formulation of Oat milk powder fortified with dates mix

Sample	Oats powder (%)	Dates powder (%)	Maltodextrin (%)	Vanilla Flavour (%)
T_0	100	-	15	2
T_1	90	10	15	2
T_2	80	20	15	2
T_3	70	30	15	2
T_4	60	40	15	2
T_5	50	50	15	2

(T_0) = Control without the addition of dates powder

(T_1) = With addition of 10% of dates powder

(T_2) = With addition of 20% of dates powder

(T_3) = With addition of 30% of dates powder

(T_4) = With addition of 40% of dates powder

(T_5) = With addition of 50% of dates powder

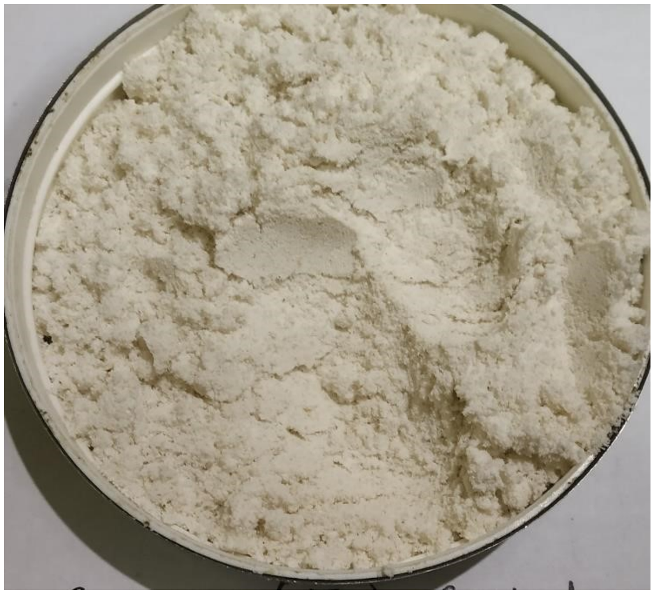

Plate no. 2: Final product

Plate no. 3: Prepared product after reconstitution of selected sample (T_4) in cold water

Table 3: Standardized recipe for RTS oat milk powder

Sr. No.	Ingredients	Quantity
1	Oats powder (g)	60
2	Dates powder (g)	40
3	Maltodextrin (g)	15
4	Vanilla flavour (ml)	2

3.5.5. Proximate analysis of milk powder

Chemical properties of oat milk powder (fortified with dates) such as moisture content, fat, protein, carbohydrate, ash content and mineral composition were determined as described earlier.

3.6. Sensory evaluation of prepared product

Sensory evaluation has been defined as a scientific method used to evoke, measure, analyzed and interpret those responses to products as perceived through the senses of sight, smell, touch, taste and hearing. Prepared milk powder was reconstituted in hot water and evaluated for sensory characteristics like color, flavor, taste, and overall acceptability by a panel of semi trained judges, comprised of doctoral students and academic staff members of College of Food Technology, V.N.M.K.V., Parbhani. Samples were scored based on a nine point hedonic scale. Judges were asked to rate the product on 9 point Hedonic scale with corresponding descriptive terms ranging from 9 to 1 as 'like extremely' to 'dislike extremely' (Meilgaard *et.al.,* 1999).

(Sensory score card is given in Appendix- I)

3.6.1. Microbial analysis of oat milk and milk powder with dates

Microbial analysis is the perfect quality assessment protocol performed in food products. These products pose the highest food safety risk and have the shortest shelf life because they are the most susceptible to microbiological deterioration and the possibility of the growth of pathogenic organisms. The results obtained for each count was recorded as colony forming unit per ml of sample i.e. cfu/ml. One gram of each sample was taken; in to this 9 ml of 0.5% saline was added and then further diluted to four folds. 1 ml of each appropriate solution was plated in required medium (Nutrient agar media and potato dextrose agar media) and then incubation was carried out. In each count after incubation, the average count of colonies present on petriplates were multiplied by dilution factor and expressed as cfu (colony forming unit)/g of sample.

3.6.1.1. Total Plate Count

Microbial analysis was done to determine total plate count (TPC) of the samples on the nutrient agar media for bacterial count by the method recommended by Ranganna (1986). Nutrient agar media was prepared and the samples were serially diluted up to 10-3 dilution factor. 1 g of the samples, suspended in saline solution, was transferred to the respective petri dishes of nutrient agar media. Three replicates were taken for each dilution. The inoculated petri dishes were incubated for 48 hours at 37+10°C and total colonies were calculated by the following formula.

$$\text{TPC (CFU/mL)} = \text{No. of colonies} \times \text{dilution factor}$$

3.6.1.2. Yeast and mold count

Microbial analysis was done to determine total yeast and mold count of the samples on the potato dextrose agar media for yeast and mold count by the method recommended by Ranganna (1986). Potato dextrose agar media was prepared and the samples were serially diluted up to 10-3 dilution factor. 1g of the samples, suspended in saline solution, was transferred to the respective petri dishes of potato dextrose agar media. Three replicates were taken for each dilution. The inoculated petri dishes were incubated in a incubator for 48 hours at 37+10°C for counting of yeast and mold.

3.6.1.3. Coliform count

The Coliform basically *E. coli* are the indicator microbes of water contamination by faeces and therefore it is mandatory to examine the contamination. The Coliform gives red pink colonies on Violet Red Bile (VRB) agar during analysis.

Using the pour-plate technique, appropriately 0.1 ml aliquots was taken in duplicate plates and tempered VRB agar was added. The agar was allowed to solidify and then overlay of about 5 ml of VRB agar was added. Allow agar to solidify. Plates were inverted and incubated at 35°C for 24 hours. Red colonies surrounded by a zone of precipitate and report as "presumptive coliforms cfu/ml.

3.7. Shelf life study of RTS milk powder mix

The prepared RTS milk powder mix was subjected to storage at ambient condition. Samples were taken at specific time interval for 30 days and up to 3 months to evaluate organoleptic and chemical characteristics at regular time intervals.

3.8. Measurement of theoretical energy value

Energy value is determined theoretically by using values of crude protein, crude fat and total carbohydrate content of sample and considering that 1 g of protein yields 4 Kcal energy, 1 g of fat yields 9 Kcal energy and 1 g carbohydrates yields 4 Kcal energy (Gopalan *et al.,* 2004). Total energy value in Kcal is calculated by adding above three energy values which gives energy value per 100 g of sample.

3.9. Assessment of techno-economical feasibility of prepared product

Assessment of techno-economical feasibility for production of RTS milk powder mix was carried out. The cost of production of most acceptable mix was calculated by considering the current prices of raw materials from local market including the processing and packaging cost. The processing cost was considered 20% of the raw material cost. The final cost was calculated per kg of final product.

3.10. Statistical analysis

All processing equipments and analysis of samples were run in triplicate. Analysis of variance was calculated using standard ANOVA procedure. The data obtained for various treatments was recorded and statistically analyzed by complete randomized design (CRD) to find out the level of significance as per the method proposed by Panse and Sukhatme, (1957). The analysis of variance revealed at significance at $P< 0.05$ level. The standard error (SE) and critical difference (CD) at 5 % level were mentioned where required.

RESULTS AND DISCUSSION

Chapter - IV

RESULTS AND DISCUSSION

The present investigation entitled "Studies on development of vegan milk powder (oat milk powder) fortified with dates" was carried out in the Department of Food Chemistry and Nutrition, with collaboration of Department of Food Process Technology, Department of Food Engineering and Department of Food Microbiology and Safety, Niche Laboratory, College of Food Technology, and Department of Soil Science, College of Agriculture, V.N.M.K.V., Parbhani during the academic year 2019-20. Sincere efforts were made to develop oat milk powder with fortification of dates powder.

The purpose behind this research project is to prepare a convenient functional food product, which is high in nutrition and to provide a milk souce which can be stored for a long time and deal with issues like lactose intolerance and hypercholoesterolemia at the same time. To become valueadded products, the value of oat milk powder must be increased through the addition of dates powder which makes it highly nutritious and more appealing to the consumers.

Physical, chemical and textural attributes were evaluated. The microbial quality of the product under ambient conditions was evaluated. The energy value and techno-economical feasibility of the developed milk powder mix was also studied. Results obtained during this investigation are presented and discussed with respect to experimental data obtained during course of study and relevant information available in scientific literature under subsequent following headings and sub headings.

4.1. Physical properties of oats

4.2. Physical properties of dates

4.3. Proximate composition of oats

4.4. Proximate composition of dates

4.5. Mineral composition of oats

4.6. Mineral composition of dates

4.7. Yield analysis of oat milk

4.8. Physical properties of oat milk powder

4.9. Proximate composition of oat milk powder

4.10. Physical properties of spray dryed dates powder

4.11. Proximate composition of spray dryed dates powder

4.12. Organoleptic evaluation of the oat milk powder fortified with dates mix

4.13. Physical properties of the prepared milk powder fortified with dates (T_4)

4.14. Proximate composition of the prepared milk powder fortified with dates (T_4)

4.15. Mineral composition of oat milk powder fortified with dates (T_4)

4.16. Microbial load of selected sample of the oat milk powder fortified with dates, (T_4) on storage at ambient temperature (30°C).

4.17. Shelf life study of oat milk powder mix with respect to packaging material

4.18. Total energy value of the oat milk powder (control) and the oat milk powder fortified with dates (T_4)

4.19. Techno economical feasibility of the prepared oat milk powder fortified with dates

4.1 Physical properties of oats

The physicochemical characteristics of raw material significantly affect the quality of finished product. And hence it is imperative to take these properties into consideration before processing into final product. During the present investigation, rolled oats were selected keeping the product yield and textural properties in mind. Rolled oats were also selected with respect to its physicochemical characteristics. The physical parameters of oats are depicted in Table 4.

Table 4: Physical properties of oats

Physical Parameters	Observations
1000 Kernel weight (gms)	30.89
Bulk density (gm/cm³)	0.419
True density (gm/cm³)	1.19
Porosity (%)	64.45
Angle of repose	46.4^{o}

*Each value represents the average of three determinations

Data from Table 4 showed that the average 1000 Kernel weight was 30.89 gms. Bulk density of oats was found to be 0.419 gm/cm³ whereas true density was 1.19 gm/cm³. Porosity of rolled oats was recorded to be 64.45 per cent, whereas Angle of repose was 46.4^{o} for oats. The results of physical properties of oats are in accordance with Sandhu K. *et.al*., (2015) and Solanki *et.al*., (2019).

4.2. Physical properties of dates

Physical characteristics of dates play a very important role in the spray drying technology for the development of the product.

Table 5: Physical properties of dates

Physical Parameters	Observations
Length (mm)	43.42
Width (mm)	18.47
Bulk density (gm/cm³)	0.56
True density (gm/cm³)	0.89
Length:Width ratio	2.35:1
Color	Dark brown
100 fruits weight (gms)	1303.4

*Each value represents the average of three determinations

The data from the Table 5 shows that the length of the date on an average, was 43.42mm, width of the date fruit was 18.47mm, its bulk density was 0.56 gm/cm³, true density was 0.89 gm/cm³, whereas the legth:width ratio was 2.35:1, they were dark brown in color and weighed 1303.4 gms for a 100 dates. The physical properties of the dates were calculated where the values were found to be similar with the results reported by Jahromi M. *et.al*., (2008) and Mahawar MK. *et.al.*, (2017).

4.3. Proximate composition of oats

The chemical composition of oats was carried out and the results obtained are tabulated in Table 6.

Table 6: Chemical composition of Oats

Chemical Parameters	Observations
Moisture (%)	4.20
Ash (%)	1.97
Total carbohydrates (%)	55.75
Crude Protein (%)	12.62
Crude Fat (%)	6.91
Total fiber (%)	13.65

The proximate composition of the oat grains were studied, which is the main ingredient in the preparation of oat milk powder. The grain has been analyzed for the various constituents like moisture, protein, carbohydrates, fat, ash etc. According to Table 6, the results of the analysis showed that the content of moisture in the oats was 4.206%, ash content was about 1.97 per cent, total carbohydrates ranged about 55.75 per cent, the crude protein content was 12.62 per cent, crude fat content was 6.91 per cent and the total fiber content was around 13.65 per cent. The results were compared and were found in agreement with Sterna V *et.al*., (2015) and Rasane P *et.al*., (2015).

4.4. Proximate composition of dates

The chemical composition of dates were analysed and are tabulated in Table 7.

Table 7: Chemical composition of Dates

Chemical Parameters	Observations
Moisture (%)	29.25
Ash (%)	1.77
Total carbohydrates (%)	63.68
Crude Protein (%)	2.03
Crude Fat (%)	0.48
Total fiber (%)	4.50

*Each value represents the average of three determinations.

Table 7 revealed the proximate composition of dates, which are used as partial replacement in the preparation of oat milk powder. Data in the table revealed that the moisture content of dates was found to be 29.25 per cent, whereas carbohydrate content found to be 63.68 per cent. It is noted that the protein content was moderate in concentration i.e. 2.03 per cent. Dates contains fat content of 0.48 per cent. Ash content of dates was further examined for the mineral analysis. Ash content in the dates was found to be 1.77 per cent whereas the fiber content was 4.50 per cent. Proximate composition of dates was analyzed and was found to be in accordance with Ahmed J *et.al*., (2014), Parvin S *et.al*., (2015), and Hussein AM *et.al*., (2017).

4.5. Mineral composition of oats

The mineral composition of oats were analysed. Minerals like calcium, magnesium, phosphorous, iron and potassium were evaluated and tabulated in Table 8.

Table 8: Mineral composition of Oats

Minerals	**Observations (mg/100g)**
Calcium	60.13
Phosphorus	474.06
Magnesium	115
Iron	9.23
Potassium	337

*Each value is an average of three determinations

The data regarding calcium, phosphorus, magnesium, iron and potassium content of oats is depicted in Table 8. The concentration of calcium was recorded to be 60.13mg/100g, oat was observed to have 474.06mg/100g of phosphorus, whereas 115mg/100g of magnesium and 9.23mg/100g of iron was found in oats. The potassium content was observed to be 337mg/100g. The results are similar to the findings quoted by Chavan and Kadam (1989), Hu *et.al*., (2014) and Youssef *et.al*., (2016).

4.6. Mineral composition of dates

The data pertaining to the essential mineral content of dates i.e. calcium, phosphorus, magnesium, iron and potassium is recorded in the Table 9.

Table 9: Mineral composition of Dates

Minerals	Observations (mg/100g)
Calcium	64.7
Phosphorus	55.6
Magnesium	55.2
Iron	1.18
Potassium	694

*Each value is an average of three determinations

The data regarding calcium, phosphorus, magnesium, iron and potassium content of dates is depicted in Table 9. The concentration of calcium was recorded to be 64.7mg/100g, dates was observed to have 55.6mg/100g of phosphorus, whereas 55.2mg/100g of magnesium and 1.18mg/100g of iron was found in dates. The potassium content was considerably high and was observed to be 694mg/100g. The results are more or less similar to the findings quoted by El-Sohaimy S.A. and Hafez E.E *et.al*., (2010), Ahmed J *et.al*., (2014) and Hussein AM *et.al*., (2017).

4.7. Yield analysis of oat milk

The selected oats, after preliminary operations of cleaning, sorting etc. was subjected to soaking for varied durations i.e. for 1 hour, 6 hours and 8 hours. After the soaking period, the oats were grinded and the slurry was filtered. The yield analysis of oat milk was carried out with respect to the amount of oat milk obtained after the soaking period and better results in terms of pH. The yield of oat milk by varying the soaking duration of the oats is tabulated in Table 10.

Table 10. Yield analysis of oat milk depending upon soaking duration

Sr. No.	Parameters	1 hour	6 hours	8 hours
1.	Yield (ml)	290.2	310.8	320.4
2.	pH	5.673	5.757	5.823

According to Table 10, it is observed that oats soaked for 1 hour had a yield of 290.2ml, pH of 5.673. Whereas, the oats soaked for 6 hours had a yield of 310.8ml, pH of 5.757. Soaking duration of 8 hours had an oat milk yield of 320.4ml, pH of 5.823.

4.8. Physical properties of spray dryed oat milk powder

After the spray drying of the oat milk, the physical parameters of the powder i.e. its yield, bulk density, wettability and solubility were measured. The results of the same are tabulated in Table 11.

Table 11: **Physical properties of oat milk powder**

Parameters	**Observations**
Yield	58.24%
Bulk density	0.53(gm/cm^3)
Solubility	69%
Wettability	89 secs

*Each value is an average of three determinations

The yield of the oat milk powder was about 58.24%. Whereas the bulk density of the same was 0.53(gm/cm^3), solubility index was 69 per cent, the wettability duration was 89 secs as given in Table 11.

4.9. Proximate composition of oat milk powder

The proximate composition of oat milk powder was evaluated and tabulated in Table 12.

Table 12: Chemical composition of oat milk powder

Parameters	Observations (%)
Moisture	2.27
Ash	3.74
Crude protein	8.12
Crude fat	4.23
Fiber	15.46
Carbohydrates	59.35

*Each value is an average of three determinations

As presented in Table 12; the chemical analysis of the oat milk powder shows that the moisture content of the milk powder was 2.2 per cent, ash content was 3.74 per cent, crude protein content was 8.12 per cent, crude fat content was 4.23 per cent, fiber content of the milk powder was around 15.46 per cent and the carbohydrates content was 59.35 per cent.

4.10. Physical properties of dryed dates powder

The spray dryed dates powder were evaluated for its physical properties like yield, solubility, wettability and bulk density, the results of which are given in Table 13.

Table 13: **Physical properties of dates powder**

Parameters	Observations
Yield	52.6%
Bulk density	0.462(gm/cm^3)
Solubility	83.5%
Wettability	108 secs

*Each value is an average of three determinations

The yield of the dates powder was about 52.6 per cent. Whereas the bulk density of the same was 0.462(gm/cm^3), solubility index was 83.5 per cent, the wettability duration was 108 secs as given in Table 13. The physical parameters of spray dryed powder were compared to Manickavasagan *et.al*., (2015) and were found to be more or less similar.

4.11. Proximate composition of dates powder

The proximate composition of spray dryed dates powder was evaluated and tabulated in Table 14.

Table 14: **Chemical composition of dates powder**

Parameters	Observations (%)
Moisture	2.37
Ash	5.53
Crude protein	4.75
Crude fat	0.64
Fiber	7.68
Carbohydrates	75.03

*Each value is an average of three determinations

As presented in Table 14; the chemical analysis of the spray dried dates powder shows that the moisture content of the dates powder was 2.37 per cent, ash content was 5.53 per cent, crude protein content was 4.75 per cent, crude fat content was 0.64 per cent, fiber content of the dates powder was around 7.68 per cent and the carbohydrates content was 75.03 per cent. The results are more or less similar to the findings quoted by Raza *et.al*., (2019).

4.12. Organoleptic evaluation of oat milk powder fortified with dates

The organoleptic evaluation of oat milk powder fortified with dates mix was carried out by ten members of semi trained panel and the scores were given by evaluating color and appearance, flavor, taste, consistency and overall acceptability which was compared with control sample and presented in Table 15.

Acceptance tests subjected to 9 point hedonic scale were performed for milk powder mix which were formulated by the addition of spray dryed oat milk powder and spray dryed dates powder in different proportions to know the acceptability of prepared milk powder mix. The acceptance scores were assigned for various sensory parameters- appearance, color, flavor, taste, consistency and overall acceptability. From the sensory mean score and the comments of the panelists, best combination was selected among prepared formulations.

Table 15: Organoleptic evaluation of RTS Powder mix (9 point hedonic scale)

Samples	Appearance	Color	Taste	Flavor	Consistenc y	Overall ceptabilit y
T_0	7.5	7.8	7.5	7.8	7.6	7.5
T_1	7.7	7.9	7.9	7.9	7.7	7.8
T_2	7.8	7.8	7.4	7.9	7.8	7.7
T_3	7.9	7.9	8.1	7.6	7.8	7.6
T_4	**8.1**	**8.0**	**8.4**	**8.1**	**8.1**	**8.2**

T_5	8	7.9	8.2	8	7.4	8
SE	0.577	0.0577	0.0577	0.0236	0.0527	0.527
CD at 5%	0.1778	0.1778	0.1778	0.0727	0.1624	0.1624

*Each value is an average of ten determinations

The result of Table 15 revealed that the study conducted to develop RTS powder mix, oat milk powder was mixed with spray dried dates powder in various proportions such as (90:10), (80:20), (70:30), (60:40) and (50:50) were used for T_1, T_2, T_3 ,T_4 and T_5 respectively and evaluated with reference oat milk powder alone (100:0) T_0.

Data obtained from the Table 12, it was recorded that the overall acceptability score awarded for sample T_4 was found higher than other samples (8.2) whereas T_0 received the lowest (7.5). The acceptance of samples depends on the ingredient variation. The sample T_4 was also reported as statistically at par with all the other samples.

The next parameter i.e. color and appearance serves as an important parameter for the acceptance of food samples. The highest score for color of milk powder mix (T_4) was obtained as (8.0). The lowest score found in the sample T_0 score (7.8). The sample T_4 was significantly superior to the other samples.

Flavor is one of the important sensory parameter. The flavor of oat milk powder mix was influenced by addition of dates powder which imparted a natural sweetness to the powder mix.

Sample T_4 obtained the maximum value (8.1) for flavor. The taste of T_4 sample of milk powder mix was significantly superior (8.4) than the other samples. T_4 sample was statistically at par with T_5 sample and significantly superior to T_3 sample.

T_4 was found to be the most preferred variant with respect to the sensory quality such as color, flavor, taste and overall acceptability. Oat milk powder with different proportion of dates powder improves the nutritional benefit of the product. Overall, it can be concluded that oat powder and dates powder in the proportion of (60:40) exhibits good sensory attributes. In comparison of different parameters by organoleptic evaluation, the formulation T_4 was selected because of its high acceptability than other samples.

4.13. Physical properties of the prepared milk powder fortified with dates (T_4)

After evaluating the organoleptic characteristics of the various oat and dates milk powder mix, the sample with the highest sensory scores (T_4), was selected for evaluating its physical, proximate and mineral composition. The physical properties of the selected sample are tabulated in Table 16.

Table 16**:** Physical properties of RTS (oat milk powder fortified with dates) powder

Parameters	Observations
Bulk density	0.52(gm/cm^3)
Solubility	78.2%
Wettability	95.4 secs

*Each value is an average of three determinations

The bulk density of the RTS powder mix was 0.52(gm/cm^3), solubility index was 78.2%, the wettability duration was 95.4 secs as given in Table 16.

4.14. Proximate composition of the prepared milk powder fortified with dates (T_4)

The proximate composition of the selected sample of oat milk powder mix i.e. (T_4) was carried out, the results of which are given in Table 17.

Table 17: **Chemical composition of oat milk powder fortified with dates powder**

Parameters	**Observations (%)**
Moisture	2.32
Ash	4.63
Crude protein	9.45
Crude fat	5.43
Fiber	11.57
Carbohydrates	65.19

*Each value is an average of three determinations

As presented in Table 17; the chemical analysis of the RTS powder shows that the moisture content of the powder was 2.32 per cent, ash content was 4.63 per cent, crude protein content was 9.45 per cent, crude fat content was 5.43 per cent, fiber content of the RTS powder was around 11.57 per cent and the carbohydrates content was 65.19 per cent.

4.15. Mineral composition of oat milk powder fortified with dates (T_4)

The data pertaining to the essential mineral content of dates i.e. calcium, phosphorus, magnesium, iron and potassium was evaluated. Given in Table 18, is the mineral composition of sample (T_4).

Table 18: **Mineral content of RTS milk powder mix**

Minerals	**Observations (mg/100g)**
Calcium	82.42

Phosphorus	464.06
Magnesium	105.2
Iron	10.41
Potassium	931

*Each value is an average of three determinations

The data regarding calcium, phosphorus, magnesium, iron and potassium of the final prepared product is depicted in Table 18. The concentration of these minerals was recorded to be 82.42, 464.06, 105.2, 10.41 and 931 (mg/100g) respectively.

4.16. Microbial load of selected sample of the oat milk powder fortified with dates, (T_4) on storage at ambient temperature (30°C).

Prepared oats and dates milk powder mix sample, on the basis of sensory quality, were stored at room temperature and analyzed at the interval of days for TPC and yeast and mold count. The data pertaining to microbial examination is shown in Table 19.

Table 19: Microbial analysis of milk powder mix

Storage period	Total plate count	Yeast and Mold
Sample (F1) (days)	**(cfu/ml) 10^3**	**count (cfu/ml)10^3**
Fresh	ND	ND
15	ND	ND
30	ND	ND
60	4.2×10^3	2×10^3
90	6.95×10^3	3.8×10^3

In the present investigation, the susceptibility of oats and dates milk powder mix to microbial growth by comparing the colony-forming units (CFU) on petri dishes inoculated with diluted milk powder mix suspensions. Selective growth media were used to differentiate between bacterial and mold/yeast growth. It was observed that the total plate count for the selected sample was found to be nil up to 30 days of storage at room temperature and yeast and mold count were not detected up to 30 days as well. But the total plate count had appeared to be 4.2×10^3 on 60th day, where the fungal count was 2×10^3 of storage and 6.95×10^3 till the 90th day, whereas the yeast and molds count were found on 90th day at range of 3.8×10^3. Selected sample was found to be low for susceptibility to microbial growth in terms of total plate count and yeast/mold count.

4.17. Shelf life study of oat milk powder mix with respect to packaging material

The organoleptic evaluation score of selected milk powder mix stored at ambient temperature is shown in Table 20.

Table 20: Organoleptic evaluation of selected sample (T_4) in different packaging material

Sensory attributes										
Parameters	**Appearance**		**Color**		**Taste**		**Flavor**		**Overall Acceptability**	
Days	**F_1**	**F_2**	**F_1**	**F_2**	**F_1**	**F_2**	**F_1**	**F_2**	**F_1**	**F_2**
Fresh	8.1	8.1	7.9	8.0	8.3	8.4	8.1	8.1	8.1	8.15
15	8.0	8.1	7.7	7.9	8.1	8.2	7.8	8.0	7.9	8.0
30	7.8	8.0	7.6	7.7	8.0	8.1	7.6	7.8	7.7	7.9
60	7.5	7.9	7.4	7.5	7.9	8.0	7.5	7.7	7.5	7.7
90	7.3	7.7	7.0	7.4	7.7	7.8	7.0	7.4	7.2	7.5

(F1- HDPE packed sample, F2- Aluminium coated pouch packed sample)

The data in the Table 20 revealed that there were certain changes occurred in sensory parameters during 90 days storage. The changes in organoleptic qualities were observed at 30 days interval. It was observed that milk powder mix packed in aluminate pouch scored highest (8.15) at initiation for overall acceptability as compared to sample packed in HDPE pouch.

Table 20 describes the sensory scoring comparison between HDPE packed sample (F_1) and Aluminate pouch packed milk powder mix sample (F_2). The color of the HDPE packed sample and F_2 are nearly similar however the F_2 sample was slightly superior to HDPE packed

(F_1) sample on fresh serving. Scoring on 15^{th} day depicts that there is significant difference between the two products as far as color and appearance is concerned however the progressing days suggested that there rise a slight difference between F_1 and F_2 sample where color perception for F_1 goes toward mere reducing direction. Till the 90^{th} day of evaluation F_1 sample (7.2) and F_2 (7.5) are found to be above acceptable level for its color property.

Organoleptic evaluation of oats and dates milk powder mix for taste attribute was analyzed for 90 days. The taste of the mix on fresh date was rated as 8.3 for F_1 sample and 8.4 for F_2 sample. After 30 days there was no change observed in the taste of milk powder mix. Taste after 60 days gave deprived results and rated as 7.9 and 8.0 for F_1 and F_2 sample respectively. The sample was further tasted after 90 days. As the product was stored at room temperature, the product gain moisture because of change in humidity, clump formation occurs thus change in taste was observed. Further observations after 90 days change the taste of the product and rated as 7.7 and 7.8 for F_1 and F_2 sample respectively.

Flavor is one of the important sensory parameter i.e., flavor is a combination of taste, smell and aroma. Similarly flavor too was observed for 90 days. On fresh date the product was rated as 8.1 and 8.1 for both the samples, however, with continuous storage of product, some volatile substances might have evaporated hence after 30 days there was slight change in flavor and thus was rated to be 7.6 and 7.8 for F_1 and F_2 samples respectively. Further observation for 60^{th}, and 90^{th} consecutive days was observed. The rating was given as 7.5 and 7.0, 7.4 and 7.5 for F_1 and F_2 samples respectively.

Overall acceptability of the product includes all the sensory attributes. The overall acceptability for F_1 and F_2 was rated as 8.1 and 8.15 respectively. After 30 days there was slight deprivation in the acceptability of F_1 sample and rated as 8.0 while there was no change in F_2. After 30 days there was slight changes observed and the product was acceptable. Further observation was done for 60 and 90 days. The rating was observed as 7.5 and 7.7 for F_1 and F_2 samples respectively after 60 days, while after 90 days the rating for F_1 and F_2 was given as 7.2 and 7.5 respectively.

It could be concluded that there was gradual decrease in overall sensory acceptability of milk powder mix with respect to storage period from 0 to 90 days.

From the Table 20 it can be seen that there was significant decrease in all the sensory parameters such as appearance, color, taste, flavor, texture and overall acceptability with successive increase in storage period.

4.18. Total energy value of the oat milk powder (control) and the oat milk powder fortified with dates (T_4)

Energy value is determined by using values of crude protein, crude fat and total carbohydrate content of sample and considering that 1g of protein yield 4 Kcal energy, 1g of fat yield 9 Kcal energy and 1g carbohydrates yield 4 Kcal energy. Total energy value is calculated by adding above three energy values which gives energy value per 100g of sample. The details of computing energy values of 100g of the selected variation of milk powder mix is summarized in Table 21.

Table 21: Theoretical energy value of oat and dates milk powder mix

Sample	Carbohydrates	Proteins	Fats	Total energy
Control	59.35	8.12	4.23	307.95
Selected (T_4)	65.19	9.45	5.43	347.43

Energy value of selected milk powder mix was found to be 347.43 Kcal/100g for T_4, and that of control sample was 307.95 kcal/100g.

Results obtained for energy value of milk powder mix showed that the higher energy value obtained for the selected sample than that of control due to the added benefit of dates powder. It can be decided from the Table 21 that 100 g of selected oat and date milk powder mix (T_4) provide 347.43 Kcal/100 g which was higher than that provided by control sample 307.95 Kcal/100 g. It is interesting to be mention that the energy value of prepared mix was found to be comparatively higher than commercial control sample and also gives an additional benefit due to the presence of dietary fiber content.

4.19. Techno economical feasibility of the prepared oat milk powder fortified with dates

The production cost of selected milk powder mix (T_4) was worked out on the basis of cost of raw materials, quantity of finished product and also including processing and packaging cost. Total cost of prepared milk powder mix was estimated on pilot plant trials and illustrated in Table 22.

Table 22: Techno-economic feasibility of milk powder mix (10 kg)

Ingredients	Rate (Rs)	Quantity (kg)	Cost (Rs)
Oats	120	6	720
Dates	500	4	2000
Maltodextrin	200	1.5	300
Vanilla essence	100	50 ml	100
Total raw material cost			3120
Processing cost @15% of the raw material cost			468
Packaging Cost / 10kg			80
Total production cost/10kg			3668/-
Production cost of porridge mix/kg			366.8/-

Table 22, depicted that the cost demanded for manufacturing of 10 kg of milk powder mix was found to be Rs. 3668. As 1kg packs were prepared from 10kg of total raw material so cost of unit pack was calculated Rs. 366.8. As compared to the cost of production of prepared oats and dates milk powder mix with the market samples, it was found that the market sample was more costly than developed mix which was with less nutritional qualities. So the product developed during the research studies has better nutritional qualities, organoleptic characteristics and calorie uptake as well as its less in cost than market sample. So this product can be suggested for commercial production.

SUMMARY AND CONCLUSION

CHAPTER-V

SUMMARY AND CONCLUSION

The present investigation entitled "**Studies on Development of Vegan Milk Powder (Oat Milk Powder) fortified with Dates**" was carried out to formulate innovative, convenient and nutritional rich product from oats and dates. With this intention, physicochemical analysis of oats and dates was carried out in the Department of Food Chemistry and Nutrition. Then their further processing was carried out at Department of Food Chemistry and Nutrition, College of Food Technology, V.N.M.K.V., Parbhani. After this, the spray dryed oat milk powder and spray dryed dates powder were studied for the yield, chemical and organoleptic quality of oats and date milk powder mix. The prepared milk powder mix was then formulated with different ratios for developing the oats and milk powder based beverage. The prepared beverage was assessed for its physicochemical analysis, microbial analysis and shelf life. And finally the techno-economical feasibility was investigated in terms of cost of production. The results obtained during present investigation are summarized and concluded as under:

The studies on liquid oat milk quality showed that the soaking of oats carried out for 8 hours had the maximum yield i.e. 320.4ml with a titratable aciidity of 0.45. The spray dryed oat milk powder was then studied for estimation of its chemical properties, physical properties which was then mixed with spray dryed dates powder and combined to determine its overall consumer acceptance. From this technical basis, organoleptic evaluation was carried out to determine which ratio had the highest hedonic score.

- To produce spray dryed oat milk powder and spray dryed dates powder, a pilot scale spray dryer was used, and their physicochemical qualities were determined. Both the powders were produced with the following processing conditions: maltodextrin (MD) was used as a carrier agent, inlet air temperature was kept at 120°C. The feedstock flow rate was kept at 25 ml/min (as provided as SOPs with the spray dryer). The carrier agent was added at the rate of 15% of the feed stock solution, and the feedstock to the spray dryer was prepared at 20% concentration.

- The spray dryed oat milk powder was fortified with spray dryed dates powder with different ratios (90:10, 80:20, 70:30, 60:40 and 50:50). Different methods employed for standardizing the formulation was further evaluated on the basis of organoleptic scores. T_4

  i.e. (60:40) was found to be the most preferred variant with respect to the sensory quality such as colour, flavour, taste and overall acceptability. Dates powder was added to increase the sweetness and overall acceptability and to provide an attractive flavour to the milk powder.

- The prepared milk powder mix was further analyzed for physical properties, chemical properties and mineral composition. It was revealed that the chemical parameters were increased in the T_4 sample as compared to the control sample which did not contain the dates powder.

- Microbiological analysis of freshly prepared oat and dates milk powder found that the sample was found to be nil up to 30 days of storage at room temperature and yeast and mold count were not detected up to 30 days as well. Further, the prepared beverage did not contain any cells of coli-form bacteria.

- The shelf life study of milk powder mix packed in aluminated packages was carried out by keeping the product at room temperature for 90 days storage period. It was observed that milk powder mix packed in aluminate pouch scored highest (8.15) at initiation for overall acceptability as compared to sample packed in HDPE pouch.

- The theoretical energy value was also analysed for the selected milk powder mix sample. Energy value of selected milk powder mix was found to be 347.43 Kcal/100g, and that of control sample was 307.95 kcal/100g. Results obtained for energy value of milk powder mix showed that the higher energy value obtained for the selected sample than that of control due to the added benefit of dates powder.

- As far as the technical feasibility is concerned, the milk powder mix reported total cost of production to be Rs. 366.8/kg.

CONCLUSION

The present investigation focuses on the development of oat and dtaes milk powder mix which imposes potential health benefits of oats and dates. The method of developing spray dryed powders of oat milk and dates was standardized and formulated. The process of preparation of milk powder mix using maltodextrin as a carrier agent at the rate of 15%, oats and dates powder ratio (90:10, 80:20, 70:30, 60:40 and 50:50), with 2% of flavoring was standardized. Thus, it can be concluded that T_4 sample with oat milk powder:dates powder ratio of 60:40 was found to be most appropriate to improve the quality of milk powder. Then, the prepared milk powder mix was analyzed for physical, chemical, mineral and microbiological characteristics. It was then stored at room temperature in aluminated stand pouches.

Therefore, the developed milk powder mix can be one of the upcoming health beverages. Expanding the trend of vegan lifestyles, the issues of lactose intolerance, and the demand for lowfat and low cholesterol foods have created a growing demand for non-dairy milk substitutes. Thus, oats and dates milk powder mix can easily be subjected to catch an eye of customers of every age group and can be made available for longer durations of time even in remote areas. Hence it is finally concluded that developed processing technology for preparation of oats and dates milk powder mix is technoeconomically viable and therefore can be commercially exploited. Moreover, it will be beneficial to the end user having nutritional and therapeutic value with added benefit of dates.

AREAS FOR FUTURE RESEARCH

- Further investigation is required for utilizing the technologies such as freeze drying, which can be an alternative to spray drying.
- Various other fortifying substances like nuts, added vitamins and minerals can also be added to increase the nutritional benefits of plant based milks.
- Future studies should be done to reduce costs of production of nut based milk.
- There is also scope for development of other flavoured plant milks.
- Studies should be done on developing milk from cheaper alternatives like amaranth, rice, peanut etc.

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APPENDICES

APPENDIX – I

VASANTRAO NAIK MARATHWADA KRISHI VIDYAPEETH
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Organoleptic Evaluation Score Card

Name of the evaluator: _ Date: _______

You are requested to evaluate the given samples and score the qualities of the given samples. Express the acceptability numerically given score point.

Score Point

9 - Like extremely 8 - Like very much 7

- Like moderately 6 - Like slightly

5 - Neither like nor dislike 4 - Dislike slightly

3 - Dislike moderately 2- Dislike very much

1 - Dislike extremely

Sample code	Appearance	Colour	Flavour	Taste	Consistency	Overall Acceptability
A.						
B.						
C.						

Sign of the evaluator

APPENDIX – II COMPOSITION OF VAROIUS MEDIA USED IN STUDY

1. Composition of Potato Dextrose Agar

Ingredients	Gms / Litre
Dextrose	20.00
Agar	15.00
Potato infusion from	200.00

2. Composition of Potato Dextrose Broth

Ingredients	Gms / Litre
Potato infusion from 200g	4
Dextrose	20
Final pH (at 25^{O} C)	7.2±0.2