

**EFFECT OF GOAT MILK BASED PROBIOTIC
PRODUCTS ON DIABETIC PATIENTS**

मधुमेह रोगियों पर बकरी के दूध आधारित प्रोबायोटिक
उत्पादों का प्रभाव

Ms. JASPREET KAUR

Thesis

Doctor of Philosophy (Home Science)

IN

(Food and Nutrition)



2023

**DEPARTMENT OF FOOD SCIENCE AND NUTRITION
COLLEGE OF COMMUNITY AND APPLIED SCIENCES, UDAIPUR
MAHARANA PRATAP UNIVERSITY OF AGRICULTURE AND
TECHNOLOGY, UDAIPUR-313001 (RAJASTHAN)**

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A

THESIS

SUBMITTED TO THE

MAHARANA PRATAP UNIVERSITY OF

AGRICULTURE AND TECHNOLOGY, UDAIPUR

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR

The Degree of

DOCTOR OF PHILOSOPHY IN HOME SCIENCE

(Food and Nutrition)

By

Ms. Jaspreet Kaur

2023

**MAHARANA PRATAP UNIVERSITY OF AGRICULTURE AND
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**CERTIFICATE-I
CERTIFICATE OF ORIGINALITY**

The research work embodied in this thesis titled: **“Effect of Goat Milk Based Probiotic Products on Diabetic Patients”** submitted for the award of degree of Ph.D. to Maharana Pratap University of Agriculture and Technology, Udaipur (Raj.) is original and bonafide record of research work carried out by me under the supervision of **Dr. Sarla Lakhawat**, Professor, Department of Food Science and Nutrition, College of Community and Applied Sciences, Udaipur. The contents of the thesis, either partially or fully, have not been submitted or will not be submitted to any other institute or University for the award of any degree or diploma.

The work embodied in the thesis represents my ideas in my own words and where others' ideas or words have been included, I have adequately cited and referenced the original sources. I also declare that I have adhered to all principles of academic honesty and integrity and have not misrepresented or fabricated or falsified any idea/data/fact/source in submission. I understand that any violation of the above will be cause for disciplinary action by the University and can also evoke penal action from the sources which have thus not been properly cited or from whom proper permission has not been taken when needed.

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CERTIFICATE – V

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This is to certify that **Ms. Jaspreet Kaur** (Ph.D. Scholar) has worked under me on “**Effect of Goat Milk Based Probiotic Products on Diabetic Patients**”.

1. I have monitored her research work.
2. Myself and the scholar were in contact with committee members and the research work was reviewed regularly.
3. The advisory committee members have gone through Ph.D. thesis critically and made the corrections as per the requirement.

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Major Advisor

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Date:

Place: Udaipur

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LIST OF ABBREVIATIONS

ADA	:	American Diabetes Association
ANOVA	:	Analysis of variance
BMI	:	Body Mass Index
BP	:	Blood Pressure
C.V.	:	Coefficient of variation
C.V.	:	Coefficient of variation
CBG	:	Capillary Blood Glucose
CHD	:	Coronary Heart Disease
CHO	:	Carbohydrate
CHOD/POD	:	Cholesterol Oxidase/Peroxidase
cm	:	Centimeter
cp	:	centipoise
CRP	:	C-Reactive Protein
CVD	:	Cardiovascular Disease
DM	:	Diabetes Mellitus
e.g.	:	Example
EDTA	:	Ethylenediaminetetraacetic acid
ELISA	:	Enzyme Linked Immunosorbent Assay
et al.,	:	Others
etc.	:	Extra
FAO	:	Food and Agriculture Organization
FBG	:	Fasting Blood Glucose
FFA	:	Free Fatty Acids
FPG	:	Fasting Plasma Glucose
g/gm	:	Gram
GDM	:	Gestational Diabetes Mellitus
h/hr	:	Hour
HbA1c	:	Glycated Haemoglobin
HDL	:	High Density Lipoprotein

i.e.	:	That is
IBS	:	Irritable Bowel Syndrome
ICMR	:	Indian Council of Medical Research
IDDM	:	Insulin Dependent Diabetes Mellitus
IDF	:	International Diabetes Federation
IDRF	:	Indian Diabetes Research Foundation
IR	:	Insulin Resistance
IS	:	Indian Standards
IU	:	International Units
Kcal	:	Kilo-calories
L.	:	Lactobacillus
l/	:	Liter
LA		Lactic Acid
LAB	:	Lactic Acid Bacteria
MPUAT	:	Maharana Pratap University of Agriculture and Technology
No.	:	Number
pH	:	Negative logarithm of hydrogen ion concentration
rpm	:	Round per minute
RT-PCR	:	Reverse Transcription-Polymerase Chain Reaction
S.	:	Streptococcus
S.Em.	:	Standard Error of Mean
SD	:	Standard Deviation
ssp.	:	Subspecies
TS	:	Total Solids
Type I DM	:	Type I Diabetes Mellitus
Type II DM	:	Type II Diabetes Mellitus
viz.	:	Vide licet (namely)
VRBA	:	Violet red bile agar

Vs.	:	Versus
W/W	:	Weight by Weight
WHO	:	World Health Organization
Wt.	:	Weight
%		Per cent
@		At the rate of
+ve		Positive
±		Plus or minus
μl	:	Microliter

INTRODUCTION

"Every time you eat or drink, you're either feeding disease or fighting it".

- Heather Morgan

Nowadays diabetes mellitus is one of the major global health issues because of its burgeoning prevalence and the extensive and diverse morbidity of its aetiology, impacting individuals, health systems and state financial system. Diabetes mellitus emerges when the body is incompetent in using glucose in its cells. Consequences of the unutilised glucose level in the bloodstream enhance its level. The major types of diabetes are Type I diabetes and Type II diabetes mellitus. Type I type is an autoimmune disease, meaning the body attacks itself. In this type, insulin-producing cells in the pancreas are damaged, so the affected person requires insulin every day, also called insulin-dependent diabetes. Type II diabetes is a common metabolic disorder where the pancreas is unable to make enough insulin or cells do not usually respond to the insulin, which is called insulin resistance. Insulin resistance (IR) induced by inflammation is a significant attribute of most patients with Type II diabetes mellitus. It is believed that there are multiple risk factors such as genetic liability, age, an unhealthy lifestyle and being overweight or obese for Type II diabetes mellitus. The burgeoning prevalence of diabetes mellitus is estimated to reach 366 million by 2030 (Azimi-Nezhad *et al.* 2008). Recent global estimation indicates adults have the diabetic condition, among those, 80 per cent reside in low- and middle-income countries (IDF, 2019). According to Figure 1.1 by 2040 estimated number of prevalence of diabetes mellitus is 642 million.

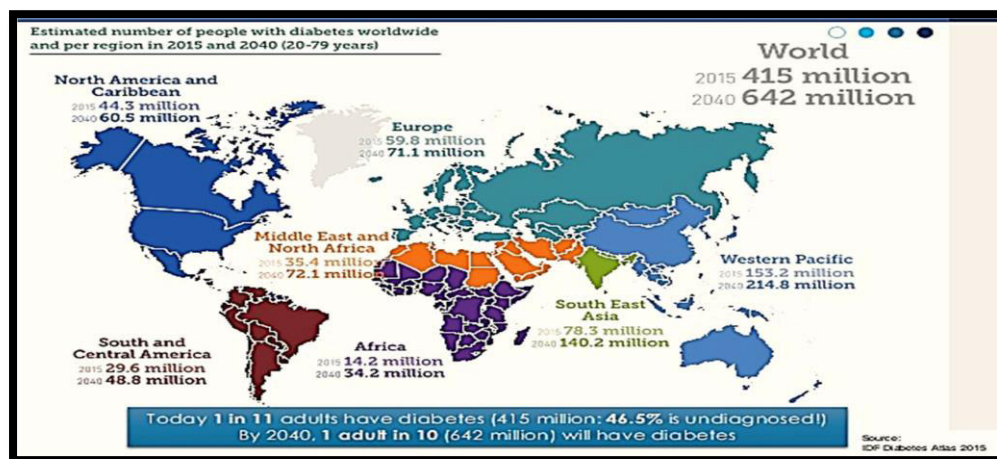


Figure 1.1 The emergent Diabetes Pandemic (IDF, 2016)

Primary symptoms of type-II diabetes mellitus are such as hyperglycemia, persistent thirst, polyuria, blurring of vision and weight loss (American Diabetes Association, 2014). Constant hyperglycemia is associated with reduced muscular strength, deprived exercise tolerance, and impaired nerve activity leading to neuropathy and poor physical functioning (Cioroiu and Brannagan 2014). Due to the progressive disabling of Type II diabetes mellitus may negatively impact the health-related quality of life due to the psychological feeling of being chronically ill, unpalatable dietary regulations, episodes of hyperglycaemia, unexpected hypoglycaemia, and perturbation of long-term consequences of ill-health (Dessie *et al.* 2020). Further, type II diabetes can be prevented through intensive lifestyle interventions directed at high-risk individuals or through population-wide changes to dietary quality, physical activity levels and levels of obesity (Haw *et al.* 2017 and Galaviz *et al.* 2018).

Probiotics have a wide range of therapeutic benefits and increased familiarity, due to which expansion of research and the market value of probiotics has increased sharply in the past few years. The food industry of India is one of the hot spots since the awareness about probiotics and their health benefits has grown tremendously among Indian denizens, the demand for probiotic foods has gained much popularity. Indian probiotic market was esteemed at \$2 million as per 2010 assessments. According to figures unleashed by US-based investigation firm Frost and Sullivan, in August 2012, probiotic products in India formulated remuneration of US \$310 million in 2011 (Raghuwanshi, 2015). The definition used presently was given by the Food and Agriculture Organization (FAO) of the United Nations World Health Organization (WHO), according to which probiotics are redefined as "live microorganisms which administered in sufficient amounts confer a health benefit on the host." In relation to food, the definition can be modified by emphasising that the beneficial effect is exerted by the microorganisms "when consumed in sufficient amounts as part of food". The most common probiotic bacteria are strains and species of *Lactobacilli*, *Bifidobacteria*, *Enterococci* and *Lactococci*. The species most thoroughly studied are *Lactobacillus acidophilus*, *Lactobacillus casei*, *Lactobacillus lactis*, *Lactobacillus helveticus*, *Bifidobacterium longum*, *Bifidobacterium lactis*, *Bifidobacterium animalis* ssp. *lactis* and *Bifidobacterium bifidum*, *Bifidobacterium longum*, and *Bifidobacterium bifidus* (Salminen *et al.* 2011). There are significant

species and strain differences between the probiotic microorganisms, and therefore, they do not all necessarily share the same characteristics (Baines, 2010). These health properties are culture-specific and are impacted by the various mechanisms. Probiotics have been used therapeutically to modulate immunity, lower serum cholesterol, treat atherosclerosis and arteriosclerosis, rheumatoid arthritis, cancer, *Helicobacter pylori* infections, improve lactose intolerance, and prevent or reduce the effects of atopic dermatitis, treat diarrhoea, and constipation, candidiasis and urinary tract infections etc. (Parvaneh *et al.* 2014 and Kumar *et al.* 2012). Probiotics have diverse mechanisms of action, whereas the exact way in which they exert their impacts is still not fully explicated. These mechanisms range from short-chain fatty acid production, reduction of gut pH, bacteriocin, immunomodulation and stimulation of mucosal barrier function. There is significant evidence that probiotics impacts various aspects of the acquired and innate immune system's response by prompting phagocytosis and IgA secretion, modifying T-cell responses, improving Th1 responses, and attenuating Th2 responses (Salminen *et al.* 1999; Guarner and Malagelada 2003 and McNaught and MacFie 2001). Probiotic products have health benefits like enhancement of intestinal health, improvement of the immune response, declined serum cholesterol levels, and cancer prevention. In fact, there are substantial studies to support probiotic use in the care of acute diarrhoeal diseases, prevention of antibiotic-related diarrhoea and improvement of lactose metabolism. Other bioactive components are produced by probiotic bacteria during the fermentation process, some of those nutrients and bioactive compounds are vitamins, proteins, bioactive peptides, fatty acids and oligosaccharides (Ebringer *et al.* 2008). Researches suggests that the risk of developing Type II DM may also involve factors from the gut microbiota. Type II DM patients had a moderate degree of gut microbial dysbiosis, and a decrease in intestinal *Roseburia* and *F. prausnitzii* was detected in their stool samples (Vazquez *et al.* 2007). The gut micro-flora has its importance in controlling and regulating different physiological processes. Once it is set, the gut micro-flora can be influenced by various internal or external factors, which can modify its ecological structure. Such factors are antibiotics, prebiotics and probiotics and thus the variety of processes in the human body can be altered. Considering this, the gut microbiota is a key factor in nutritional metabolism and possibly, its imbalance contributes to the development of obesity (Ley *et al.* 2006) and diabetes (Larsen *et al.* 2010 and

Andersson *et al.* 2010). These morbidnesses are associated with considerable changes in the constitution and the metabolic functions of the gut microbiome (Kootte *et al.* 2012). Interest in human micro-flora has only recently emerged and it has been shown that human micro-flora probably has a major role in maintaining the homeostasis of human metabolism and the application of probiotics is a useful approach for the modulation of the human microbiota.

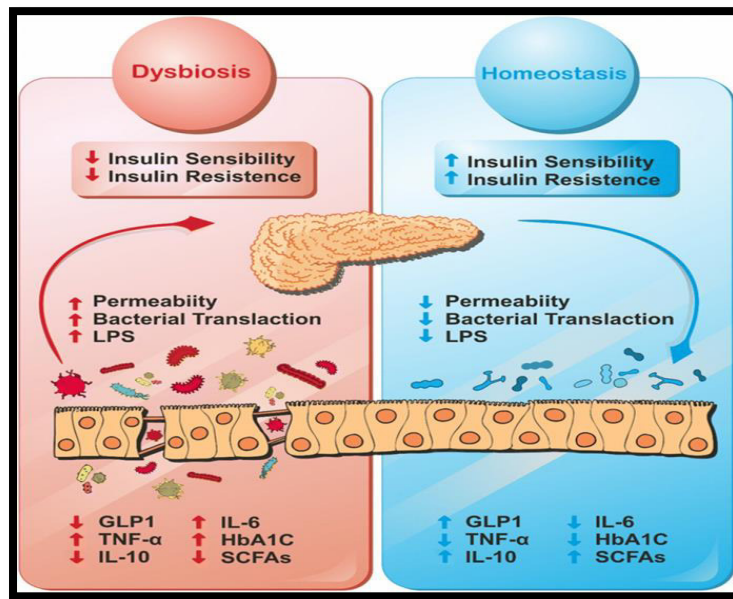


Figure 1.2 Role of probiotics to improve diabetes mellitus

Source: Yanming *et al.* 2020

Fermented dairy products are well-known as functional foods. It is nutritionally and therapeutically beneficial for human beings (Yadav *et al.* 2007). The fermented milk products are prepared by the action of microorganisms by adding a starter culture, which modifies the substrates and is thus generally palatable, safe and nutritious. (Campbell, 1994 and Anonymous, 2006). Fermented foods have great nutrition and health-enhancing properties, including preventing gastrointestinal infections, decreasing serum cholesterol levels and antimutagenic activity in the human body. Fermentation of milk is beneficial for ingestion by lactose-intolerant human beings and patients experiencing atherosclerosis (Shiby and Mishra 2013). A cultured milk beverage, a well-liked product in India, is called *Lassi*. It is not only refreshing, delicious and nutritious, but it also possesses thirst-quenching properties and a high therapeutic value. *Lassi* is a popular indigenous fermented milk beverage, which is usually prepared by mixing Dahi and water in the required proportions and

sometimes even spices or fruits are added. It is close to sweet stirred yoghurt (Fluid yoghurt).. It is served on a very large scale in cold drink shops and restaurants during summer in almost every state in India (Manuka *et al.* 2019). *Lassi* origin dates back to palaeolithic and neolithic times in India (Mathur, 1991). It is a ready-to-serve beverage that occupies a prominent place in the Indian diet). In the Middle East, including Iran and Lebanon, a similar salty yoghurt beverage named "Doogh" is popular ((Tiwari 1998; Kosikowski 1978; Tamime and Robinson 2007).

Yoghurt is a fermented dairy product having many health benefits and therefore, it is highly used by dairy researchers. The main strains used in yogurt starter cultures are *Streptococcus thermophilus* and *Lactobacillus bulgaricus* (Singh *et al.* 2012). One of the most important aspects of yogurt quality is texture. *Lactobacillus delbreuckeii bulgaricus* and *Streptococcus thermophilus* microorganisms both provide well-defined flavour and texture in yoghurt. Along with these properties, these two microorganisms are probiotic in nature and thereby exert maximum health benefits to consumers. The main processing parameters influencing the yogurt texture are fortification level and material(s) used, stabilisers type and usage levels, fat content and homogenisation conditions, milk heat treatment conditions, starter culture (type, rate of acid development and production of EPS), incubation temperature (influences growth of starter cultures, gel aggregation, bond strength), pH at breaking, cooling conditions, handling of product post manufacture (e.g. physical and temperature abuse) (Lucey and Sing 1997; Lucey 2004 and Sodini *et al.* 2004). Modern consumers are more concerned towards functional food, which is not only giving nutrition but also provides health benefits, viz., reduction of cholesterol, antidiabetic blood pressure in rhythm, etc. Yogurt has the potential to occupy a key position on the spectrum of food products that provide various health benefits to individuals through specific influences on their intestinal microbiota. In yogurt and fermented milk, proteins are partly degraded by the action of the bacterial proteolytic system that facilitates their digestion (FAO 2013 and Moreno *et al.* 2013). Yogurt contains different natural compounds with a high nutritional value, such as vitamins (mainly B₁₂, riboflavin and vitamin D); also, it contains proteins, peptides, and minerals (principally calcium, phosphorus, and potassium). Because of its nutritional value, yogurt is considered a foodstuff for daily consumption (Singh *et al.* 2013). This versatility, together with their acceptance as a healthy and nutritious

food, has led to their widespread popularity across all population subgroups (Mckinley 2005). It had become the lunch of choice for young women and a product for dieters. The use of yogurt as a calcium source has made it one of the fast-growing dairy products, but now it is more than just a calcium source. Yogurt and similar fermented dairy milk products are becoming major nutraceuticals directed at treating a variety of non-communicable disease conditions (Katz 2001).

Milk and dairy products have an important role in the human diet due to their many beneficial nutrients, such as proteins, sugars, minerals, and water-soluble vitamins (Barkallah *et al.* 2017). Goat husbandry popularity is increasing throughout the world, which has been a part of agriculture since almost the first use of domestic animals. This increase is reflected in a greater degree by the rise in the number of small herds maintained by individuals either as a source of revenue or as pursuance. Goats form a vital component of the livestock industry and an important factor in improving the socio-economic structure of the rural poor (Park and Haenlein 2006). A goat is universally called as "Poor man's cow" (Iqbal *et al.* 2008) and has enormous potential to be anticipated as the 'Future Animal' for rural and landless farmers under the changing agro-climatic conditions and diminishing resources in India. Developing countries rear 775 million of the 809 million goats in the world and amongst them, low-income nations possess 86 per cent, which has food insufficiency in their area. The worldwide goat population, 65 per cent is reared in the developing nations of Asia and 29 per cent in Africa. Currently, the global production of non-bovine milk exceeds 133 million tons per year, accounting for more than 17 per cent of all milk production. Of this, 13.5 per cent is goat milk output, which is considered one of the prime contributors to non-bovine milk production (Nune and de Renobales 2016, Ranadheera *et al.* 2018).

The dairy goat industry has congregated the desire of the consumer, in slow but firm growth, for products alternative to cow's milk. Innovative goat dairy products are therefore increasing, with ingredients that can satisfy the taste of today's consumer (da Silveira *et al.* 2015). Primarily of bovine origin, dairy products are the leading vehicles for the delivery of probiotic bacteria. Caprine (goat) milk also possesses the potential for the successful delivery of probiotics and despite its less pleasing flavour in some products, the utilisation of goat milk as a probiotic carrier has rapidly

increased over the last decade. The dairy industry has also beheld the chance to produce functional dairy products from goat milk, which may potentially benefit human health and contribute to attenuating issues related to CVD, obesity and diabetes (Albano *et al.* 2018).

Goat milk-based infant formulas are also attaining traction due to their good nutritional content, which prevents neurodevelopment disorders in children. In addition, the escalating requirement for goat milk in the personal care section for manufacturing creams, lotions and soaps is also catalysing the market. The demand for goat milk has also been augmented by the COVID-19 pandemic in early 2020. The pandemic has resulted in the rising consumption of goat milk due to its immunity-boosting benefits (<https://www.imarcgroup.com/goat-milk-market>).

Compositions of goat milk diverge with feeding, breed, season, management, environmental conditions, locality, stage of lactation, and health status of the udder, which also influence the taste of goat milk. Goat milk contains protein, lipids, carbohydrates, vitamins and minerals. Goat milk contains 3.8 fat, 3.4 protein, 4.1, lactose, 0.8, ash, 8.9 per cent, SNF (Park *et al.* 2007) and 87 water per cent respectively (Iqbal *et al.* 2008). Lipids are the most important components of milk in terms of cost, nutrition, and physical and sensory characteristics that impart to dairy products. The superior digestibility of goat milk, the proper composition of short and medium-chain fatty acids, and its content of bioactive compounds with an as lower level of α -s1-CN seem to give properties suitable for treating or preventing certain medical conditions (Park, 2007). The high proportion of short and medium-chain fatty acids in goat milk contributes to easier and faster digestion. Medium-chain triacylglycerols in goat milk are absorbed intact in the intestine and do not undergo degradation and reestrefication process. Micelle formation is not required for absorption since the molecules are taken up directly into the portal vein (Lopez-Aliaga *et al.* 2010). These attributes of goat milk that has contributed to its increasing interest by consumers (Almaas *et al.* 2006).

Excessive consumption of food containing high sugar and saturated fats is one of the key nutritional issues mankind has had to face recently. It has been linked to various serious health problems. (Sukhmani *et al.* 2018). Coconut sugar, Honey, table sugar, agave, blackstrap molasses, high fructose, maple syrup, corn syrup, and other

natural sweeteners contain glucose, fructose, and sucrose as their major constituents. However, sweeteners derived from natural sources contain a high caloric value, which leads to obesity, cardiovascular diseases and diabetes. There has been a persistent rise in the number of diabetic patients globally. Health awareness is improving nowadays and this has led to a remarkable rise in demand for low-calorie/fat products. Many low-calorie food products have been developed, adding dietary fibre or low-calorie sweeteners as consumers are striving to choose healthier food alternatives (Gandhi *et al.* 2018). Due to increasing health awareness, there has been an immense demand for sugar substitutes that provide fewer or no calories and contain better sweetening potency. There is a wide variety of artificial zero-calorie sweeteners on the market, likewise acesulfame potassium, cyclamates, saccharin, aspartame, etc. However, artificial sugar substitutes became associated with health complications, and the use of these artificial sugar substitutes has subsequently been restricted. Thus, there is a continuous search for high-intensity low, calorie or non-caloric sweeteners of natural origin that are safe for consumption. Stevia, which plays a vital role as a non-nutritive natural sweetener, emerged as a safe sugar substitute that does not pose any threat to human health (Savita *et al.* 2004).

Stevia rebaudiana (Bertoni) is a perennial shrub which belongs to the Asteraceae family, native to Paraguay. Out of 230, only two species – *rebaudiana* and *phlebophylla* - produce sweet steviol glycosides (Brandle and Telmer 2007). Indigenous people have used the leaves of this plant for centuries in medicines and to sweeten drinks such as maté, a green herbal tea (Brandle *et al.* 1998; Carakostas *et al.* 2008 and Lewis 1992). The stevia plant is now commercially cultivated in Argentina, Brazil, Columbia, Paraguay, China, Japan, Malaysia, South Korea, Vietnam, Israel, Australia, Kenya, and the United States. High-purity steviol glycosides are approved as sweeteners by all major regulatory authorities across the globe, and >150 countries have approved or adopted their use in foods and beverages. Reb-A was the first commercial steviol glycoside launched in the marketplace. All major global scientific and regulatory bodies have determined high-purity steviol glycosides to be safe for consumption by the general population. The majority of the regulatory approvals pertain to high-purity ($\geq 95\%$) steviol glycosides. Unpurified crude extracts of stevia have been reported to cause adverse effects on fertility in animals (Planas and Kuc 1968, Melis 1999). The process of purifying stevia leaf into

high-purity stevia leaf extract. This is similar to other plant-based ingredients, such as natural vanilla extract or cane sugar is made through various steps beginning from harvested, raw plant material through to the end product. The process begins by drying the leaves and then steeping them in hot water. Next, the liquid extract is filtered and purified with water or, in some cases, in combination with food-grade alcohol. If food-grade alcohol is used, it is later removed and no significant amount of alcohol remains in the end product (Prakash *et al.* 2008). Surveys have been utilised in various global jurisdictions to determine daily consumption estimates of high-purity steviol glycosides. The FAO/WHO Joint Expert Committee on Food Additives (JECFA) assessed international dietary exposure estimates with the use of a model that assumed steviol glycosides would replace all sweeteners used in or as food, based on the relative sweetness of steviol glycosides to sucrose. The committee estimated maximum intakes of 1.3–5 mg SEskg⁻¹ d⁻¹ worldwide. However, the committee acknowledged that these estimates were highly conservative and indicated that actual intakes were more likely to be 20–30 per cent of these values (JECFA, 2006). In India, the use of steviol glycosides was permitted by FSSAI in a notification issued in 2015 for a variety of products, including yoghurts, carbonated beverages, jams, fruit nectars, dairy-based desserts, ready-to-eat cereals, etc.

As described above, the health-promoting properties of goat milk have attracted consumers, which increased the consumption of goat milk. Increased demand for goat milk was driven by the small size of fat globules, less allergic, higher digestibility and fatty acids close to recommended ratio for the prevention of cardiovascular diseases and diabetes in humans. Probiotics and goat milk constitute plenty of health benefits for the body. Today, therapeutic regimens or nutritional interventions are among the primary recommendations for dealing with diabetes and maintaining health for a normal person also.

Hence, this research has been planned to develop probiotic products based on goat milk for diabetic people.

Objectives:

- ❖ To develop and standardize goat milk based probiotic products.
- ❖ To evaluate the physico-chemical and organoleptic properties of goat milk based probiotic products.
- ❖ To determine shelf life of the of goat milk based probiotic products.
- ❖ To study the effect of probiotic products on blood glucose levels of diabetic patients.

REVIEW OF LITERATURE

A comprehensive review of literature is an essential part of any scientific investigation. To collect the earlier findings review of literature is the simplest way to economizing and inquisition and almost essential before going to research endeavor. Thus this chapter attempts to present an overview of the previous research outcome which directly or indirectly related to present investigation entitled as “Effect of Goat Milk Based Probiotic Products on Diabetic Patients” under following heads:

2.1 Diabetes mellitus and it's status

2.2 Fermentation and it's history

2.3 Probiotics and it's role for health

2.4 Cultures: *Streptococcus thermophilus* and *Lactbacillus helveticus*

2.5 LASSI: it's physioco-chemical properties

2.6 Yogurt : it's physioco-chemical properties

2.7 Goat milk and it's products and health benefits

2.8 Stevia and its products and health benefits

2.1 DIABETES MELLITUS AND IT'S STATUS:

The World Health Organization (WHO) defines diabetes mellitus as a set of syndromes “*characterized by hyperglycaemia and disturbance of the carbohydrate metabolism that are associated with absolute or relative deficiencies in insulin action and- /or insulin secretion*”. (WHO) Several subgroups of diabetes mellitus have been identified. The most prevalent are type I, diabetes type II and gestational diabetes. Diabetes type II is twenty times more common than type 1 diabetes, with almost 90 per cent of patients having diabetes type II and approximately 10 per cent diabetes type I.

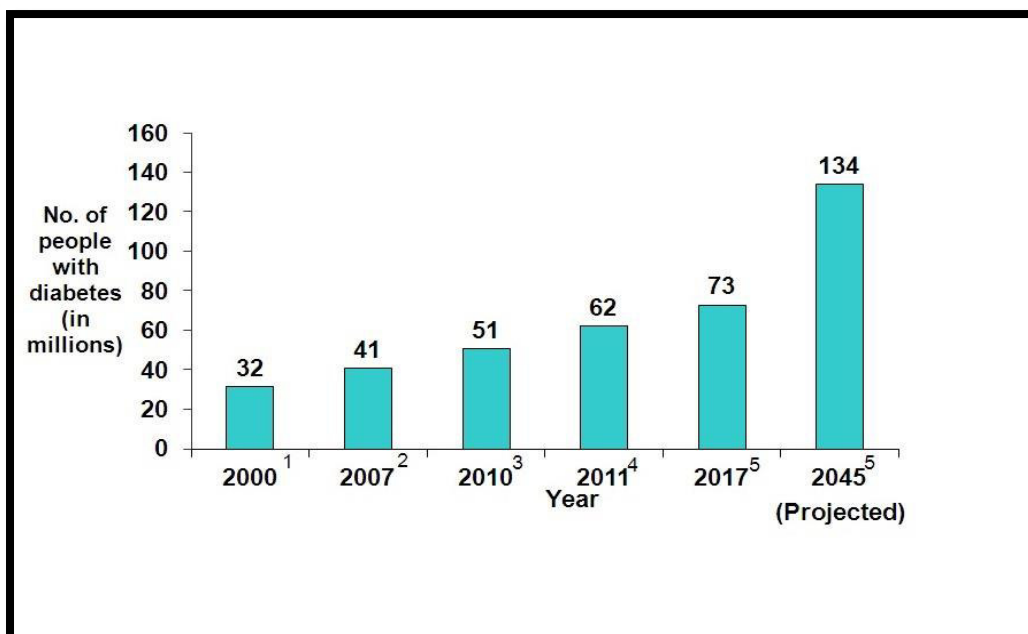


Figure 2.1 Prevalence of diabetes mellitus in India

Source: <https://www.indiascienceandtechnology.gov.in>

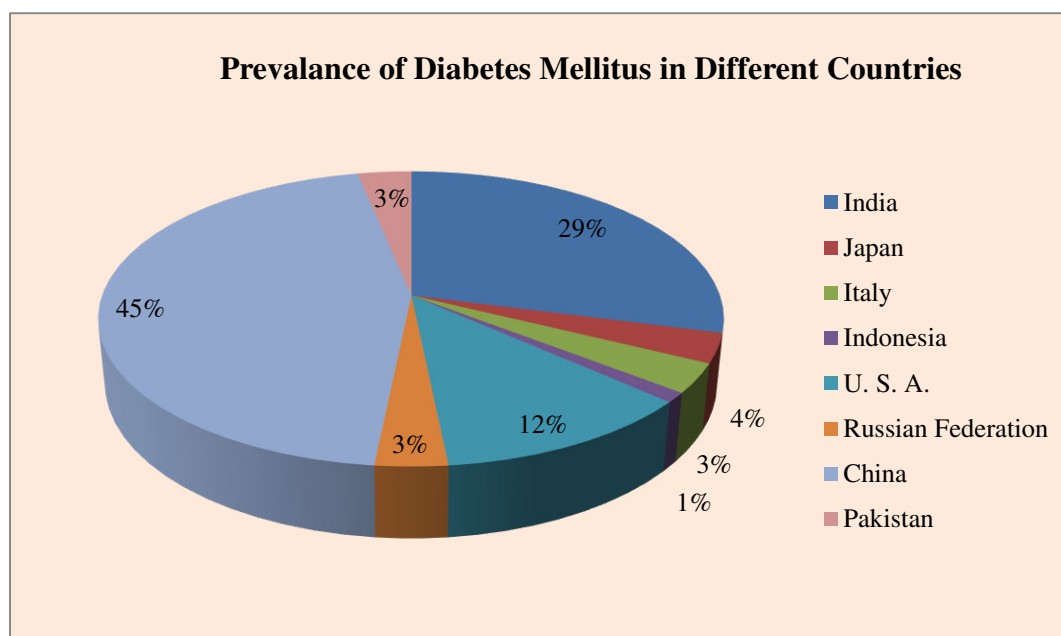


Figure 2.2 Prevalence of diabetes mellitus among top 8 countries

Source: <https://www.indiascienceandtechnology.gov.in>

Table 2.1 History of Diabetes mellitus

Year	Event
1889 ↓	The finding the role of the pancreas in diabetes mellitus was made by Joseph Von Mering and Oskar Minkowski. They found that upon absolute removal of the pancreas from dogs, the dogs exhibited all the signs and symptoms of diabetes and died shortly afterwards (Himsworth, 1936).
1910 ↓	Sir Edward Albert Sharpey-Schafer of Edinburgh in Scotland suggested that diabetics lacked a single chemical which was normally produced by the pancreas. Name of this chemical was later proposed to be insulin (Himsworth, 1936).
1921 ↓	Frederick Grant Banting and Charles Herbert Best repeated the work of Von Mering and Minkowski but went a step further and managed to show that they could reverse the induced diabetes in dogs by giving them an extract from the pancreatic islets of Langerhans of healthy dogs. This was a step forward in elucidation of the endocrine role of the pancreas in metabolism and existence of insulin (Banting, 1922).
1922 ↓	Scientists proceeded on to isolate insulin from bovine pancreases at the University of Toronto in Canada, thereby leading to the availability of an effective treatment of diabetes mellitus, with the first clinical patient being treated (Himsworth, 1936).
1935	The distinction between what is now known as type I and type II diabetes was made by Sir Harold Percival (Harry) (Himsworth, 1936).

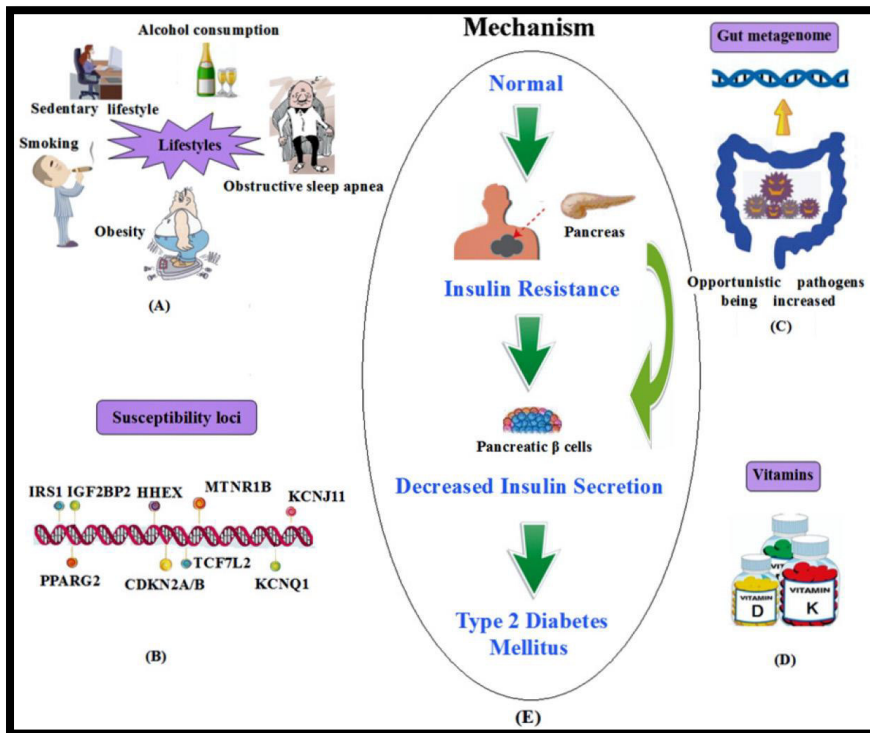


Figure 2.3 A review of the influencing factors and mechanism of Type II DM. (A) Lifestyles; (B) Susceptibility loci; (C) Gut metagenome association (D) Vitamins. (E) The mechanism of T2DM. (Yanling *et al.* 2014)

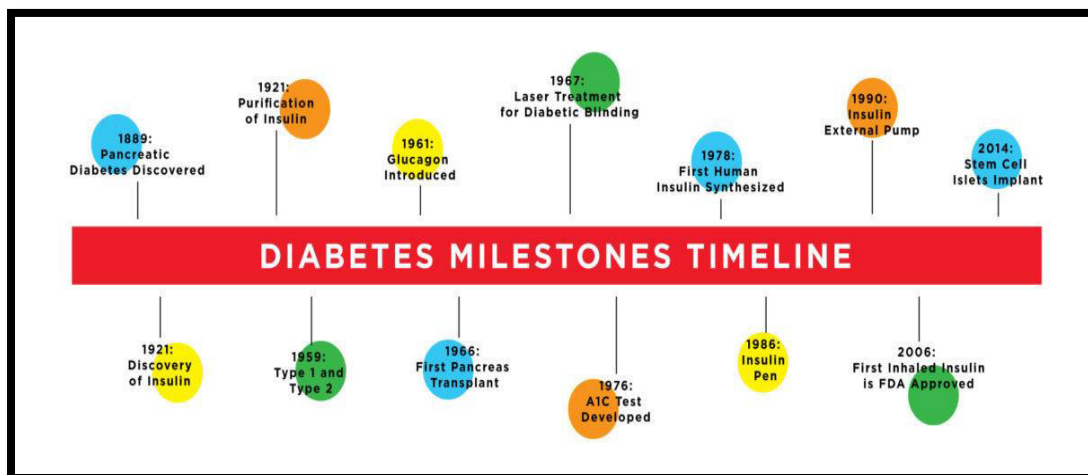


Figure 2.4 Diabetes milestone timeline (Kalberg, 2022)

Pal *et al.* (1994) reported that type II diabetes mellitus and all its related issues can be directly caused by oxidative stress, which primarily takes place intracellularly. Mitochondria are present in every cell in eukaryotic organisms that are oxygen

dependent and are a good source of oxidants. Some of the mitochondrial oxidants which are formed during respiration are hydrogen peroxide, hydroxyl radicals and superoxide (Paliyath, 2011).

Elton *et al.* (1996) and Kawakami *et al.* (1999) both reported that a history of a major depressive disorder increases the risk of type II diabetes mellitus. Studies showed that depression is accomplished by increased sympathoadrenal system activity as measured by dopamine, norepinephrine and adrenaline in cerebrospinal fluid, plasma and urine (Maes *et al.* 1991). Which are known to be related to impaired glucose tolerance (Surwit *et al.* 1992).

Kannel and McGee (1997) reported that type II diabetes mellitus is a global unresolved health challenge which is a risk factor for various other diseases such as hypertension, coronary heart disease, retinopathy and renal disorders (Papatheodorou *et al.* 2016).

Cerrillo (2000) reported that postprandial hyperglycemia leads to various complications. Atherosclerosis is a common disease caused by postprandial blood glucose, which leads to oxidative stress and redox imbalance. Oxidative stress occurs because of the metabolic effects of mitochondria.

Hajer *et al.* (2008) stated that insulin resistance is one of the major causes of type II diabetes mellitus along with dysfunction of pancreatic β -cells. A combination of these issues renders the body unable to regulate blood sugar. The typical response of the pancreas to the elevated blood glucose level and increased insulin resistance for more production of β -cells. Therefore more insulin secretion can compensate the insulin resistance. Increased production of β -cells and insulin causes additional stress on the pancreas, which leads to β -cells dysfunction. Insulin plays an important role in blood glucose homeostasis. Hyperglycemia can be defined as blood glucose of more than 100 mg/dL of blood (Wilson 2005).

Type II diabetes which accounts for 80-95 per cent of all diabetes, has an underlying asymptomatic phase of sub clinical stage which often remains undiagnosed for a number of years (IDF, 2013). Indian have a high risk of developing diabetes at a younger age compared with western people (Ramchandran and Snehlata 2010).

Sanghera *et al.* (2012) reported that diabetic neuropathy can be linked with foot ulcer, amputations, sexual dysfunction and non-healing skin wounds.

Dugas *et al.* (2018) studies obesity is a major trigger for type II diabetes development. Diabetes is a complex state that is elucidated by risk factors like total energy expenditure, physical activity level, dietary intake, heredity factors, and education qualification, along with unconventional causes, which are highly studied as microbiota associated with obesity.

Holman (2020) reported a high mortality rate in type I diabetes mellitus patients compared with a population without type I diabetes. Patients who have cardiovascular issues, obesity type I and type II issues were at high risk of severe COVID-19 or death.

2.2 FERMENTATION AND ITS HISTORY:

Chandan and Shahani (1995) stated that milk is the natural habitation of a number of lactic acid bacteria reason of the souring of milk held at bacterial growth temperature and appropriate time duration. Bacteria entered by various sources make soured milk which is undesirable in taste and can cause serious health hazards. However, in commercialization, starters containing specific genes and strains of culture are used to generate preferred flavoured, body and texture properties.

Tongnual and Fields (1997) reported that fermentation of various foods has been used for thousands of years for a diversity of causes, most vital for the preservation of food. The fermentation process of foods is also important for flavours and health benefits like remarkable increment in vitamin B content (Paliyath, 2011).

Hill *et al.* (2017) studied microbial fermentation has been used for a long period of time for preservation. Early dairy fermentation depended on the activity of indigenous microbiota present in the milk. However, nowadays, fermentation relies on specific cultures with specific properties like consistency.

2.3 PROBIOTICS AND IT'S ROLE FOR HEALTH:

Gilliland and Walker (1990) studied some strains *L. acidophilus* that can decrease cholesterol absorption by increasing the binding of cholesterol to the intestinal lumen. Other potential cholesterol-lowering properties of probiotics are

deconjugation of bile by salt hydrolysis, binding of cholesterol to the cellular surface and coprecipitation of cholesterol with deconjugate bile.

Holcomb and Frank (1991) studied that a low number of *L. bulgaricus* assist in improving the viability of bacteria due to reducing post-acidification (Shah, 1995).

Saarela *et al.* (2000) stated that probiotics' health benefits include enhancement of the immune system, decrease the level of serum cholesterol, cancer prevention, antihypertensive effects and improvement in lactose metabolism (Nagpal *et al.* 2012).

Shah (2000) reported that the viability of probiotic bacteria is an important factor for the therapeutic effects. There are various factors which can affect the viability of dairy products, like low pH and cold storage.

Moore *et al.* (2001) stated that expectant mothers getting intensive nutrition counselling and food supplements with *L. rhamnosus* GG (10¹⁰ CFU/ gm) and *B. lactis* BB12 (10¹⁰ CFU/ gm) improve glucose tolerance and decrease HbA1C level compared with the control group receiving only a control healthy diet.

Shah *et al.* (2001) recommend that the viable bacterial count of probiotic culture should be available for the therapeutic values varying from 10⁶ - 10⁹ CFU/gm in the food product, which was present in both samples as probiotic *LASSI* and symbiotic *LASSI* still storage at 4 ±1° C for 28 days.

Songisepp *et al.* (2005) reported the functional value that is antioxidant properties of probiotics in well being subjects and found significant enhancement the total antioxidant activity and total antioxidant status after consumption of probiotics.

Yadav *et al.* (2006) conducted a study on laboratory rats and presented that milk fermented by *L. acidophilus* and *L. casei* can have an anti-diabetic effect such that delays the inception of impaired glucose metabolism in mice which are fed with a high fructose diet. Intake of the product was reported to postpone the inception of glucose intolerance in the mice blood and reduce oxidative stress.

Yadav *et al.* (2007) studied the diet fortified with Dahi containing *L. acidophilus* and *L. casei* significantly postponed high fructose glucose intolerance, hyperglycemia and dyslipidemia in rats.

Elgert (2009) reported enteric bacteria changes the balance of pro-inflammatory and anti-inflammatory cytokine level of the intestine, become the predisposing factor for intestinal diseases. Th1 cells produce the pro-inflammatory cytokinase Th2 produced the anti-inflammatory cytokinase, which is important to maintain the homeostasis of immune system in the intestinal barrier.

Harisa *et al.* (2009) reported a significant reduction in MDA concentration after feeding diabetic rats with *L. acidophilus*.

Huey *et al.* (2009) reported the antihypertensive effects of probiotics via insulin resistance and sensitivity, treatment of lipid profile and alteration of rennin profile and conversion of bioactive phytoestrogen and estrogen.

Saccaro *et al.* (2009) reported that *Lactobacillus acidophilus* count 7.17 log CFU/ml at the day of storage. It was reduced 5.26 log CFU/ml at the fourteen days of storage, and the count has been fallen below five log CFU/ml on the 21st day of storage. The viability of probiotic bacteria in fermentation milk is declined due to damage because of acid formed (Shah, 2000).

Yun *et al.* (2009) reported a significant decline in fasting and postprandial glucose and a decline in HbA1c in probiotics (BNR 17) treated rats.

Abdul – Ghani *et al.* (2010) stated that adipose tissue and muscle resistance to insulin response in type II diabetes mellitus are triggered majorly by a complex combination of hereditary predilection, body composition, nutritional and environmental factors. Insulin receptors, post-receptor perturbations and glucose transporter are observed in type II diabetes mellitus. Eventually, peripheral tissues are exposed to chronic compensatory hyperinsulinemia and ultimately become resistant to insulin. Some other studies also showed that microbiota is related to the development of metabolic disorders, as obesity and diabetic patients present perturbations in the proportion of Firmicutes, Bacteroidetes and proteobacteria (Larsen *et al.* 2010).

Ooi and Liong (2010) analysed the few species of *Lactobacillus*, *Bifidobacterium*, enterococcus and streptococcus and represented the capacity to reduce the plasma cholesterol level. The major mechanism for reduction is that these probiotic bacterial cultures can decline the reabsorption of bile acid via enterohepatic circulation. This is because of the fact that conjugated bile acid, not free forms, is

incompletely absorbed and directed reverse to the liver. Bacteria cultures like Lactobacilli and bifidobacteria can hydrolyse the conjugated bile acid and excrete it faster and decline the extent of absorption.

Slacanac *et al.* (2010) stated that lactic acid bacteria are generally used as probiotic starter cultures and therapeutic effects from these are related to increased lactose intolerance, a balanced micro-biota, antimicrobial activity, stimulation to immunity and hypocholesterolemic and anti-oxidative properties.

Ejtahed *et al.* (2011) reported a significant decline in blood glucose levels and MDA levels in type II diabetes mellitus patients after being fed the probiotics yogurt.

Khor *et al.* (2011) reported ulcerative colitis and chron's disease are kinds of bowel diseases associated with the gut's microbial genetic predisposition and environment. Infringement of balance between the intestinal immunity and microbiome can lead to bowel diseases.

Chapman *et al.* (2011) reported that the therapeutic properties of probiotics consumption include as improvement in lactose intolerance, cancer management, reduce the level of serum cholesterol, enhanced gastrointestinal immunity, prevention of infection of the urinary tract and prevention of diabetes mellitus.

Stefanov *et al.* (2011) examine the pathogenesis of diabetes mellitus patients as multifaceted and among them one factor the oxidative stress. Food which contains probiotics is found to be a suppressor of oxidative stress. *L. acidophilus* and *L. casei* also attenuate oxidative stress and have hypoglycemic effects. It shows the *L. casei* reduced oxidative stress.

Honda *et al.* (2012) reported that in the management of diabetes, the specified strain of the lactic bacterium could be useful. Probiotics are live microorganisms that can exert an anti-diabetic impact and improve blood glucose homeostasis. Studies depicted that probiotic bacteria can help to maintain glycemia and dyslipidemia (Mahboobi *et al.* 2014).

Mazloom *et al.* (2013) studied diabetic patients aged from 25-65 years with fasting blood glucose ≥ 126 mg/dl who were diagnosed with diabetes mellitus for more than 15 years. Probiotics culture used *Lactobacillus acidophilus*, *Lactobacillus*

bulgaricus, *Lactobacillus bifidum* and *Lactobacillus casei*. Patients fed for six weeks and reported non-significance reducing trends in the level of IL-6, MDA, TG and insulin resistance.

Hussain *et al.* (2014) analysed the effect of probiotic and conventional yogurt on lipid profile in diabetes in type II and reported that probiotic yogurt feeding might be used as an alternative treatment approach.

Carl *et al.* (2015) analysed that fasting blood glucose level and glycosylated (HbA1c) haemoglobin level was declined in type II patient by nosh of probiotic yogurt, but probiotics also reduced diet-induced insulin resistance in healthy individuals; however not known.

Ostadrahimi *et al.* (2015) evaluated the consumption of probiotic fermented milk, which reduced more HbA1C and fasting blood glucose compared with conventional fermented milk.

Tonucci *et al.* (2015) studied in a double-blinded, randomised, placebo-controlled trial aged 35-60 years, 50 volunteers consumed daily 120 g/d of fermented milk for six weeks. Participants were divided into two groups; fermented milk consuming containing *Lactobacillus acidophilus* La-5 and *Bifidobacterium animalis* BB12 and the control group consumed conventional fermentation milk. Results depicted a significant reduction in fructoseamine level and HbA1c level reduced in the probiotic group and also found enhanced glycemic control, reduction in inflammatory cytokines and increase in acetic acid.

Zang (2016) stated after the meta-analysis that the consumption of probiotic yogurt plays a factor which can reduce the blood glucose level in diabetic patients.

Sabico *et al.* (2019) reported the impact of multi-strain probiotics consumption on type II diabetes mellitus in a randomized, double-blind, placebo-control trial for six months and concluded the benefits of probiotics for inflammation and promising adjuvant anti-diabetic therapy.

Tsalamandris *et al.* (2019) reported that diabetic patients developed constant inflammation with an elevated level of pro-inflammatory cytokinase, which have a negative impact on insulin. Lactic acid bacteria contain antioxidant properties which target the inflammatory status in diabetic patients. However, the effect is dependent

on the strain. *Lactobacillus rhamnosus* culture shows more impact compare with *Bifidobacterium* culture in the regulation of glycolipid metabolism and improving the gut micro-biota (Wang *et al.* 2020).

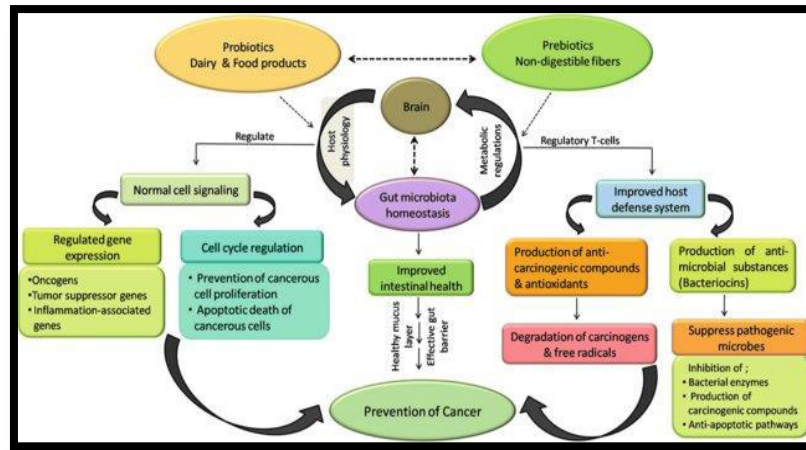


Figure 2.5 Role of probiotics in prevention of Cancer

Source: Sharma *et al.* (2021)

2.4 CULTURE: *Streptococcus thermophilus* and *Lactobacillus helveticus*:

2.4.1 *Streptococcus thermophilus*:

Aim *et al.* (1982) reported that during yogurt preparation, *S. thermophilus* grows optimally between 37-45 °C and hydrolyse lactose via β - galactosidase. This bacterium needs a complex of amino acids to grow and is nutritionally fastidious.

Pearce and Flint (1999) reported that *S. thermophilus* have a need for free amino acid for growth because it has inadequate proteolytic activity. These are cysteine, methionine, histidine, glutamic acid, valine, leucine, isoleucine, tryptophan, arginine and tyrosine. Though free amino acids naturally present in milk are not sufficient. The free amino acid supplements during heat treatment in milk or the absorption of short-chain peptide release by the breakdown of milk protein by *L. delbrueckii ssp bulgaricus*.

Beal *et al.* (1999) stated that started culture improves the nutritional properties and digestibility. It is also evident that *S. thermophilus* do not colonise the intestine, although consumption of viable cells can improve lactose digestion in lactose-intolerant persons. The possibility for this phenomenon is the viable cells of *S. thermophilus* survive in the stomach and are lysed in the GI tract. The intracellular β -

galactosidase is released and hydrolyses lactose. Lactose does not reach the large intestine and the symptoms of lactose intolerance do not occur.

Hui (2007) reported that *S. thermophilus* is a gram-positive bacteria that are gram catalage negative. It is small cocci in pair with long chains between 0.7-0.9 μm . It is a facultative anaerobe and its fermentation is homofermentative. Its optimum pH for growth is 6.5. This bacteria is identified among the lactic acid bacteria by its high-temperature tolerance. *S. thermophilus* can ferment in glucose, fructose, lactose and sucrose.

Delorme (2008) reported that *S. thermophilus* is of commercial use because it makes the texture in yogurt because of the secretion of exopolysaccharide (Broadbent *et al.* 2003).

Passephole *et al.* (2008) reported that *S. thermophilus* count in yogurt improved with inulin and aloe vera after a while. Microbial counts were stable on the first and seventh day, but there was a significant increase on the fourteenth and thirtieth day.

Srisuvor *et al.* (2013) studied that *S. thermophilus* increased in number over time. *L. bulgaricus* counts had a significant effect over time. *L. bulgaricus* counts had a significant effect over time. Cont was stable on the first day and seventh days and reduced significantly after that. The viability of *S. thermophilus* was higher than *L. bulgaricus* (Passephol *et al.* 2008).

Bintsis and Athanasoulas (2015) investigated the *S. thermophilus* and *L. Spp.* are both used as commercially significant starter cultures for various fermented dairy products, likewise yogurt, fermented milk, Mozzarella cheese and Feta.

2.4.2 *Lactobacillus helveticus*:

Khedkar *et al.* (1991) reported that *L. helveticus* MTCC 5463 strain was originally isolated from the vaginal tract of healthy adult females' in India at Anand Agricultural University.

Mishra *et al.* (2001) reported the positive impact on mucosal and tumoral immunity was directly associated with the proteolytic system of *L. helveticus* R389, Since milk containing the non-variant was not able to prompt immunity.

Ng and Griffiths (2002) also reported that *L. helveticus* R389 is proficient at hydrolysing milk protein to produce bioactive peptides that have positive effects on immunity. There is a need to work done to characterise the proteolytic system, which is responsible for the generation of these peptides and has a protective impact on Salmonella infection in vivo (Telleza *et al.* 2010, 2011).

Narva *et al.* (2004) conducted a study on male rats with osteoporosis and found that *L. helveticus* fermented milk helps to improve bone density and bone mass composition, which were analysed by dual-energy X-ray absorptiometry (DEXA).

Narva *et al.* (2004) observed the effect of *L. helveticus* fermented milk on acute changes in metabolic in postmenopausal females. The effects of probiotics on bone in 20 postmenopausal women with a mean age of 65 years and the BMI of 26 revealed the increment in the serum calcium level.

Songisepp *et al.* (2005) studied an RCT with hypertensive who consumed a fermented milk product containing bioactive peptide resulting from fermentation with *L. helveticus* strain LBK-16H compared with the placebo group. Subjects who were fed the fermented milk group reported a reduction in systolic and diastolic blood pressure compared with the placebo group.

Moreno and Le Blanc *et al.* (2005) conducted an in vivo study using a model of breast cancer in mice and concluded that milk fermented with *Lactobacillus helveticus* R389 or *Lactobacillus helveticus* L89 delayed tumour development.

Frece *et al.* (2009) reported that *L. helveticus* strains are generally isolated from milk products and intestinal microflora. *Lactobacillus helveticus* is present in fermented food products and also used as probiotic (Champagne *et al.* 2010, Kilpi *et al.* 2007 and Messaoudi *et al.* 2011).

Prajapati *et al.* (2011) reported that the indigenous strain *L. helveticus* MTCC 5463 has been proven to have probiotics potential with their specific beneficial role in the GI tract and it can also reduce the cholesterol level and stimulate immunity.

Moreno *et al.* (2013) analysed the impact of *L. helveticus* against diseases such as intestinal inflammation and cancer. Milk fermented with *L. helveticus* R389 deferred breast tumour growth by declining IL-6 and decreasing IL-10 in serum, mammary glands and tumor-infiltrating immune cells.

Parvaneh *et al.* (2014) reported that *L. helveticus* produces the proline containing peptides isoleucyl- propyl- proline and valyl – prolyl- proline, which may induce a greater availability of minerals.

Yamashita *et al.* (2014) studied the *L. helveticus* SBT2171, one of the lactobacillus bacteria used in Gouda cheeses preparation. An in-vitro study shows that *Lactobacillus helveticus* helps to prevent proliferation and cytokinase production in LPS- stimulated murine splenocytes. Oral administration of *L. helveticus* 2171 in mice who suffered from collagen-induced arthritis showed a decline in joint swelling and serum levels of anti-bovine type II collagen antibody (Yamashita *et al.* 2017).

Gohel *et al.* (2016) studied healthy volunteers with ages ranging from 64-74 years on 59 subjects who successfully completed a double-blind cross-over trial. The probiotic product (*LASSI*) is prepared by supplementing toned milk with honey and fermented with probiotic *L. helveticus* MTCC 5463 and *S. thermophilus* MTCC 5460. During the four weeks of the feeding period, 200 ml of *LASSI*, at least 10⁸ CFU/ml, was fed. Results show a significant improvement in the calcium level and no significant difference was found in haemoglobin and haematological parameters.

2.5 LASSI : Their Physical and Chemical Properties:

Speck (1984) reported that coliforms were absent in both samples in properties throughout 28 days of cold storage. The presence of coliform bacteria in dairy products is an indicator of unhygienic conditions or practices followed during storage.

Patidar and Prajapati (1998) reported that *L. acidophilus* and *S. thermophilus* counts in *LASSI* remained above 10⁷ throughout the refrigerated storage (7±1) for 15 days and consumers also reported that lactobacilli count remained at a satisfactory level even at the 27th day of storage.

Shah (2000) reported that the viable count of probiotic culture, which should be in the range of 10⁶ to 10⁹ in the product, was available in both the sample, *viz.*, probiotic *LASSI* and symbiotic *LASSI* samples after the refrigerated storage for 28 days.

Shuwu *et al.* (2011) studied the value-added honey-sweetened *LASSI* and fruit *LASSI* were developed. Five different levels of curd thinning were attempted and the sample at 3:2 curd-to-water ratio secured the highest score (8.32) for overall

acceptability. Among the four honey *LASSI* samples assessed by organoleptic viz., 27.5, 30, 32.5 and 35 per cent honey syrup, the sample at 30 per cent honey syrup secured the highest overall acceptability score of 8.20. Three diverse fruit pulp (mango, pineapple and banana) were used at 7 per cent (w/v) pulp level to develop fruit *LASSI*. Pineapple *LASSI* secured the highest overall acceptability score (8.33), followed by mango and, lastly the banana sample. The physico-chemical evaluation showed a significant increment in T.S. (18.93 and 18.65%), carbohydrates (17.74%) and a slight reduction in protein (2.1 and 1.98%) and ash content (0.45 and 0.46%) and a significant decrease in fat (2.0 and 1.5%) of both the honey and pineapple *LASSI* respectively against the control sample (17.78% T.S., 2.2% protein, 2.5% fat, 17.4% carbohydrates, 0.51% ash content). From the sensory quality and microbiological analysis, it was deduced that the products had a shelf life well above one week under refrigeration storage, considering no coliform counts and least yeast and moulds counts till the 10th day of storage as compared with the PFA standard limits for yoghurt.

Bhutkar *et al.* (2012) studied coconut milk blended *LASSI* made from different preparation of coconut milk. Physico-chemical properties of products were analysed. Four treatments were analysed as T₀, T₁, T₂ and T₃ and moisture content was 83.84, 81.81, 80.25 and 77.97 per cent, fat content was 2.94, 2.81, 2.76 and 2.74 per cent, protein content was 1.28, 1.74, 1.79 and 1.84 per cent, ash content 0.38, 0.42, 0.49 and 0.54 per cent, total solids 16.09, 18.19, 19.75 and 21.78 per cent and carbohydrates 11.55, 13.21, 14.70 and 16.92 per cent respectively. The Titratable acidity was analysed as 0.61, 0.59, 0.59 and 0.57 per cent, respectively.

Shukla *et al.* (2013) studied a probiotic beverage from whey and pineapple juice and reported a decrease in the titratable acidity in both whey and whey pineapple juice. Probiotic bacteria were reduced, but the viability count of probiotic bacteria was not reduced below 10⁶ CFU/ ml.

Nair *et al.* (2013) studied *LASSI*-like beverages made with different proportions of guar gum, CMC and inulin. The product was analysed organoleptically and physically. The organoleptic score for flavour, consistency, colour and appearance of *LASSI*-like beverage varied from 6.50 to 7.25, 6.16-7.12, 6.20-7.41 and 5.96-7.20, respectively.

Pardhi *et al.* (2014) reported that the level of finger millet flour increased and the total sugar content in *LASSI* also increased. It shows that the mean scores of sugar content range from 14.43, 14.83, 15.51, and 16.25 for the treatments T₀, T₁, T₂ and T₃, respectively. *LASSI* can be stored for up to 7 days at 5°C. There was no significant difference in the sensory qualities (Ranganadham *et al.* 2016).

Ghule *et al.* (2015) researched the strawberry-fortified *LASSI* and analysed its composition fat content ranges from 3.11-3.25, protein content 3.64-3.78, lactose content 3.73-3.82, total sugar content 13.92-16.29, ash content 0.73-0.80. pH level and acidity were 4.13 and 0.90-1.02 per cent, respectively. The fat content of *LASSI* samples declined with an increase in the level of strawberry pulp, while lactose content was enhanced with as increase in the level of strawberry pulp. The overall acceptability for the sensory score for *LASSI* made from 5 per cent strawberry pulp is (8.09 to 7.90). The mean lactobacilli count was recorded to be 4.33×10^7 CFU/ ml and *LASSI* samples under study did not show growth of any yeast and mould was also reported. The cost for the sensorial superior combination *LASSI* prepared with 8 per cent sugar and 5 per cent strawberry pulp could make a 200 ml serving at ₹ 6.80, which may be sufficiently lower with the value added and nutritionally enriched combination of strawberry pulp.

Kumar and Kumar (2016) studied the preparation of fruit-fortified probiotic buttermilk using *L. rhamnosus*. Probiotic *LASSI* was prepared by supplementing commercial *LASSI* (Verka, India) with probiotic *L. rhamnosus* (5%). Probiotic *LASSI* samples were fortified with antioxidants-rich fruit juice (10% v/v) of *Rubus ellipticus*, *Prunus domestica*, *Prunus armeniaca* and *Syzygium cumini*. The probiotic *LASSI* was prepared using free alginate (2%) and carrageenan (2%) encapsulated probiotic culture. The microencapsulated beads were characterised by the FTIR technique wherein the intensity of peak increased Apricot and raspberries fortified *LASSI* prepared using alginate encapsulated probiotic culture retained recommended probiotic values of 6.42 ± 0.03 and 6.55 ± 0.03 log CFU/ml respectively up to 5 days of storage. The antioxidant power of fruits fortified probiotic *LASSI* samples decreased successively during storage.

Ghule *et al.* (2016) reported the sensory characteristics of different brands sold in Kolhapur city as colour and appearance, flavour, consistency and overall acceptability. The sensory scores of *LASSI* brands were significantly different. The

highest score for colour and appearance was 8.44, scored by the KS₁ brand, followed by KS₄ (8.02). The mean score for body and texture ranged from 7.20 to 8.19, while the mean flavour score of the *LASSI* varied from 7.32 to 8.30. The lowest and highest flavour score for *LASSI* was scored for brands KS₃ and KS₁, respectively. The highest score (8.22) for consistency was allotted to the sample KS₁ *LASSI*, followed by sample KS₄ (8.21). *LASSI* of the KS₁ brand recorded the highest overall acceptability score (8.29), followed by KS₄ (8.09), KS₆ (8.06), KS₂ (7.73), KS₅ (7.56) and KS₃ (7.42). All the samples were acceptable and the rated in between liked moderately (Score 7) to liked extremely (score 9). Significant differences were also recorded in all the physico-chemical properties of *LASSI*. The average acidity (% L.A.) *LASSI* of brands KS₁, KS₂, KS₃, KS₄, KS₅, and KS₆ were 0.95, 0.78, 0.79, 0.92, 0.83 and 0.67, respectively, whereas the pH in *LASSI* sample was ranged from 3.83 to 4.3. The maximum viscosity was recorded for the *LASSI* from KS₄ brand (381.50 cP) followed by KS₆ (247.00 cP) and KS₁ brand (243.33 cP). A sample of *LASSI* with too low viscosity was rated with the minimum sensory score for body and texture and consistency by the panel members. The fat content in *LASSI* samples was 4.24, 3.00, 2.10, 3.62, 2.46 and 4.03, while, the mean protein content was 3.60, 3.15, 3.18, 2.79, 2.78 and 3.28 in KS₁, KS₂, KS₃, KS₄, KS₅ and KS₆, respectively. The lactose content (%) varied 3.84 to 4.45 and overall sucrose content ranged from 8.24 to 10.54 per cent in *LASSI* samples. The T.S. content in brands of *LASSI* samples in was in varied from 19.82 to 21.43 per cent. The mean T.S. content in *LASSI* of brand KS₁ was highest, followed by KS₂.

Hussain *et al.* (2016) studied also barbadensis Miller-supplemented probiotic *LASSI* (APL) and concluded that the reduction in pH during storage at 5°C. The probiotics cell count was also reduced during the study from 8.4 log CFU/ml on the day to 8.0 log CFU/ml on the 12th day.

Todkar *et al.* (2016) researched four per cent of standardised milk from the milk *LASSI* was prepared with different proportions of papaya pulp (0%, 4%, 8%, 12%) as in T₁, T₂, T₃ and T₄ by the weight of Dahi. Nutritional, physical and sensory properties were analysed. The sensory evaluation for (overall acceptability) showed that *LASSI* prepared by combination with 8 per cent papaya pulp (T₃) was most acceptable. Therefore, it is concluded that the best quality papaya *LASSI* can be

prepared by using 8 per cent papaya pulp. This *LASSI* contained 3.18 fat, 20.43 T.S. 0.92 per cent acidity and protein 2.61 per cent, respectively.

Todkar *et al.* (2016) reported the overall acceptability of *LASSI* on a 9-point Hedonic scale. The 8 per cent papaya pulp *LASSI* received the best rating (8.60 out of 9), while the plain *LASSI* (T₁) received the lowest rating (6.53 out of 9). As a result, it was shown that the amount of papaya pulp in *LASSI* boosted overall acceptability.

Todkar *et al.* (2016) reported that the maximum score (8.60 out of 9) was rated by the panel members to *LASSI* prepared from 8 per cent level of papaya pulp while the lowest score (6.53 out of 9) was secured by plain *LASSI*, i.e. T₁. Thus it was reported that overall acceptability was improved with the enhancement in the levels of papaya pulp in *LASSI*.

Dhumal *et al.* (2018) studied the *LASSI* prepared from buffalo milk with equal quality milk and optimised the level of pudina extract. Chemical analyses were done for the product. On average, the *pudina* extract used in *LASSI* for the treatment T₁, T₂, T₃ and T₄ contained moisture content observed was 88.45, 88.68, 88.75 and 88.84 per cent, fat 2.60, 2.53, 2.43 and 2.40 per cent, protein 2.22, 2.24, 2.27 and 2.30 per cent, ash 0.45, 0.45, 0.46 and 0.47 per cent, total solids 16.09, 18.19, 19.75 and 21.78 per cent and sugar 5.28, 5.18, 5.16 and 5.16 per cent, respectively. The study in respect of titratable acidity was 0.78, 0.76, 0.73 and 0.71 per cent and pH was reported as 4.19, 4.24, 4.25 and 4.27 per cent, respectively.

Dixit *et al.* (2019) studied moringa powder mixed at 0.5, 1.0, and 1.5 per cent of milk and whey. The addition of whey at 19, 29, and 39 per cent and pectin at 0.1 per cent and vanilla were to enhance the acceptability of the product of moringa *LASSI*. The chemical and microbial viability was analysed. The highest carbohydrate content was T₃ (16.69%), the highest protein content was T₃ (3.01%), the highest fat content was T₀ (2.47%), ash T₃ (0.82), Total solid T₃ (23.10), Lactic acidity T₀ (0.87) and pH T₀ (4.08). The highest mean value of Yeast and Mould count was the record in the Moringa *LASSI* of T₀ (7.80×10¹) and SPC of Moringa *LASSI* T₀ (19.00×10⁷). The mean value for viscosity (centipoise) of moringa *LASSI* of T₀, T₁, T₂, and T₃ was reported to be 243.22, 247.27, 252.07 and 258.14, respectively. The moringa *LASSI* prepared by blending with Dahi, whey and moringa powder, viscosity in samples of different treatments and control, the highest mean viscosity (centipoise) was recorded

in the sample of T₃ (258.14 cp) followed by T₂ (252.07 cp), T₁ (247.27 cp) and T₀ (243.22 cp). It specifies that increasing the amount of whey and moringa powder in the blend declined the viscosity content of moringa *LASSI*.

Kaur *et al.* (2019) studied *LASSI*, which was fortified with carrot pulp in an effort to increase the product's vitamin A content. The final product's physico-chemical profile was found to be 2.48 to 2.37 per cent fat, 0.42-0.51 per cent acidity (measured as lactic acid), 13.64 to 16.21 per cent total sugars, and 14.00 to 15.43 per cent TSS, respectively. While parameter including acidity, viscosity, fiber content, total sugars, and TSS increased with an increase in the percentage of carrot pulp, the fat content of the *LASSI* samples declined. The *LASSI* made with 10 per cent carrot pulp received the highest overall sensory score was "liked very much." It was discovered that the mean lactic acid count increased when the pulp was increased from 8.50 to 8.68 log CFU/ml. However, The *LASSI* samples were free from yeast and mould.

Krishna *et al.* (2019) researched on *LASSI* was made by inoculating heat-treated milk with several starter cultures, such as LS₁ - *Lactobacillus delbrukii ssp bulgaricus*, *Streptococcus thermophilus*, *Bifidobacterium bifidum* NCDC15; LS₂ - *L. delbrukii ssp bulgaricus*, *S. thermophilus*, *L. acidophilus* NCDC232; and LS₃ - By using probiotic Dahi and sugar syrup, *LASSI* was standardised to include 10 per cent total milk solids and 10 per cent sugar. The maximum sensory score was achieved by *LASSI* made from LS₃ - *S.thermophilus*, *B.bifidum* NCDC15, and *L.acidophilus* NCDC232. Additionally, the *LASSI's* shelf life was extended by six days when compared to the control *LASSI* made from *L.bulgaricus* and *S.thermophilus*. Excluding *L. bulgaricus* from the fermentation process may have reduced post-acidification during chilled storage, extending the shelf life and boosting the viability of probiotic cultures.

Sudheendra *et al.* (2018) reported that during storage, variations in the lactobacilli count were observed. In both the fresh products and throughout the refrigeration storage period, the lactobacilli count of both products was significantly higher than 8.5 log CFU/g (28 days). In simple probiotic *LASSI* (A) and synbiotic *LASSI* with carrot juice (B), the first counts were 33 and 43 x 10⁷ (8.52 and 8.64 log CFU/g), which are statistically non-significant. According to the statistical analysis,

there were highly significant changes in the storage of *LASSI* sample A up to the 14th day, at a seven-day interval, and from the 14th to the 21st day, changes were non-significant. Later, throughout day 28th, there were significant modifications. While in sample B, changes were highly significant up to day seven and then exhibited no change until day 28th. Until the seventh day, blend B's probiotic count was higher than blend A's, which may have been caused by the presence of prebiotics in carrot juice and the FOS added.

Pintu and Verma (2019) reported that optimized rabadi (sorghum based fermented milk beverages) and evaluated the effect of milk solid sources, namely, sour buttermilk and skim milk, were tried. The viscosity of the beverage prepared from sour buttermilk (62cp) was slightly higher than that of skim milk (55cp) based beverage.

Deshpande *et al.* (2020) studied the acceptable low-cost flavoured *LASSI* that can be prepared by using soymilk blended with buttermilk can be prepared with the addition of sugar (15%) and mango pulp (5%). Due to the low cost of blended-flavoured *LASSI*, it could have good market potential. The prepared product has been evaluated by trained judges based on 9- point Hedonic scale to prepare the flavoured *LASSI* with the blending of buttermilk and soymilk at different concentrations. The prepared product was evaluated for its colour and appearance, flavour, body and texture, taste and overall acceptability of the final product. The flavoured *LASSI* prepared with 70 per cent buttermilk and 30 per cent soymilk got the highest overall acceptability by the panel members than other treatments.

Jamdar *et al.* (2020) reported that an improved variety Parbhani Shakti which have been shown to increase protein, iron, zinc, and phytate content, which increases the bioavailability of better nutrients. In the proposed recipe, an attempt was made to produce *LASSI* blended with an extract of germinated bio-fortified sorghum (Parbhani Shakti) using skim milk and its compositional features were evaluated. Acidity, pH, fat, protein, total sugar, moisture, total solid, ash, and viscosity were observed to be normal. Ash levels in the *LASSI* sample were between 0.72 and 0.83 per cent. T₄ (0.83) had the highest value and T₁ had the lowest (0.72). For treatments T₁, T₂, T₃, and T₄, the average ash concentration was 0.72, 0.75, 0.81 and 0.83 per cent, respectively. From treatment T₁ to T₄, the recorded values were determined to be in

increasing order. This could be because the extract from germinated sorghum grains has more nutrients than buffalo milk. Due to the greater mineral content in sorghum, treatment T₁ significantly differed from treatments T₂, T₃, and T₄ (Kumar *et al.* 2018). The mean fat content for the developed *LASSI*'s was 0.44, 0.42, 0.40, and 0.32 per cent respectively for treatments T₁, T₂, T₃, and T₄. Due to the fat content of sorghum grains, the fat percentage in produced *LASSI* had slightly decreased. Treatment T₁ had the greatest value for acidity (0.82%), and Treatment T₂ had the lowest mean value for acidity (0.60%).

Jagdale *et al.* (2020) reported the physical-chemical characteristics of synbiotic buffalo *LASSI* were examined in the conducted study. Maltodextrin was used as a prebiotic for making the synbiotic *LASSI* and was added at T₁-1.0 per cent, T₂-1.5 per cent, T₃ 2.0 per cent, and T₀ control without maltodextrin. 1.5 per cent of all therapies contain the probiotic *Lactobacillus acidophilus*. It was determined that there was a significant variation in the total solids content of synbiotic *LASSI*. Additionally, it was shown that when the quantity of maltodextrin increases, the total solids content steadily decreases. The increasing incidence of microorganisms is the cause of the declining trends in total solids. Treatment T₀ had the highest total solids content (22.38%), whereas T₃ had the lowest (22.31%). Treatment changes were demonstrated to be highly significant. The synbiotic *LASSI* treatment T₀ without maltodextrin had the highest fat content (5.07%), whereas treatment T₃ had the lowest (5.043%). It was found that treatment differences are statistically significant at 1% significance level significance, indicating that the fat content in *LASSI* significantly decreased when maltodextrin levels rose. A higher rate of microbial proliferation was demonstrated to have a prebiotic level tendency. Thus, some fat can be reduced by microorganisms.

Kahandal *et al.* (2020) stated that the purpose of the current study, cow milk, was standardised to include 3.5 per cent fat in order to prepare *LASSI* using the starter strain of *Streptococcus lactis*. Diacetyllactis was added to standardised cow milk, which was then incubated for 8–12 hours at 22–25°C. 10 per cent water and 10 per cent sugar were mixed with curd to create the *LASSI*. Strawberry pulp was added at 0, 5, 10, 15, and 20 per cent. The findings of chemical analysis of the fat, protein, total solids, ash, acidity, and pH were obtained. The best treatment, T₄, included 15 per cent strawberry pulp and an overall acceptability score of 8.72. The acceptability

declined as the amount of strawberry pulp exceeded treatment T₄. The addition of strawberry pulp changed the *LASSI's* chemical makeup. From treatment T₁ to T₅, the *LASSI's* pH, ash content, and total solids increased, whereas its fat, protein, and titratable acidity levels reduced.

Saha *et al.* (2021) reported that yogurt-based beverage called *LASSI* originated on the Indian subcontinent. It is a great beverage to hydrate and build body digestion as well as an excellent food source. *LASSI* is a sound and expected response for stomach swelling because it is made from yoghurt or Dahi and is said to be very beneficial for our stomach-related framework. It is also mild on the stomach and contains lactobacilli, stable microbes that grease up the digestive organs and aid in smooth processing. Since *LASSI* is a probiotic drink, it aids in the growth of good microbes and inhibits the growth of harmful microbes in the stomach. *LASSI* can be overwhelmed with salt or sugar and helps prevent heat strokes since it is packed with probiotics. *LASSI* is highly recommended since it contains beneficial bacteria that aid in warding off harmful microbes and in preventing stomach swells. Excess water in the body causes swelling.

2.6 YOGURT AND IT'S PHYSIOCO-CHEMICAL PROPERTIES:

Kulpsch (1971) reported that to prolong the shelf life of milk, and there is a need for some specific culture and also specified that yogurt made from *Lactobacillus acidophilus* or *Bifidobacterium Bifidus* instead of *Lactobacillus delbruekii ssp bulgaricus* had better acceptability than regular yogurt after one month.

Tramer (1973) affirmed that the increase in acidity is what produces the characteristics coagulum of yogurt and thus, a reasonable development of acidity is needed to achieve the desired texture of the yogurt. As the acid accumulates in the milk and the acid level enhances, protein denature and takes an acidic taste. Acidity variation can change the texture. At a lower level of acidity, the yogurt will be sweeter and thinner; at high acidity, yogurt will be thicker and sourer (Goff 2009).

Aim (1982) reported that heat treatment, homogenisation and acidification are helpful in increasing the in vitro digestibility of culture dairy products. The protein of yogurt, acidophilus milk and Bifidus milk have been found to be more digestible compared with non fermented dairy products.

Gilliland (1985) stated that milk with cells of *Lactobacillus acidophilus* aids the digestion of lactose by such a person, has been reported lactose intolerant persons are better able to consume the fermented dairy product as yogurt. Individuals who consume fermented dairy products show less symptoms than those who consume the same amount of fermented milk. Yogurt was found to be accommodative in the digestion of lactose due to lactic acid bacteria used to make yogurt produce lactase and digest the lactose.

Tamime and Robinson (1985) reported that yogurt or yoghurt is a universally accepted product, which is taken from the Turkish name “Yogurt”. It is fermented milk with thermophilic lactic acid bacteria developed a preservation method for milk learnt by different communities.

Bibel (1988) reported that at the beginning of this century, Nobel laureate Elie Metchnikoff at the Pasteur institute associated health and longevity with the consumption of bacteria present in food such as yogurt, kefir and sour milk. The study hypothesised that these microflora control the infection because of enteric pathogens and regulate the natural toxemia, which plays a vital role in ageing and mortality.

Dannenberg and Kessler (1988) reported that the gel strength of yogurt is associated with the total effect of chemical interactions. During denaturation, β -lactoglobulin interacts with the binding of β -lactoglobulins with the K- casein on the casein micelles surface by disulfide bridging is responsible for improving gel and the viscosity of yogurt (Lucey *et al.* 1997).

Scrimshaw and Murray (1988) reported that after the fermentation, natural yogurt has the same or less lactose as milk, but in low-fat yogurt, the ranges can vary widely because of the elaboration process and the amalgamation of other ingredients during fat replacement.

Beal *et al.* (1999) observed higher viscosity in the stirred yogurt, which was incubated at lower temperatures compared to gels incubated at high temperatures (Martin *et al.* 1999; Sodini *et al.* 2004 and Lee and Lucey 2006). As the incubation temperature increased, there was a reduction in the organoleptic properties, such as mouth coating and smoothness of stirred yogurts (Cho-Ah Ying *et al.* 1990).

Shah and Jelen (1990) reported the changes in the Titratable acidity, pH and hydrogen peroxide value in yogurt stored in plastic cups at 4°C and 10°C and in glass bottles stored at 4°C. The increment in the Titratable acidity was more in the samples stored at 10°C. This indicated the continued residual fermentation at this temperature. However, the reduction in the pH was not enough to reduce the pH of yogurt <4.0, which is harmful to probiotic bacteria survival (Laroia and Martin 1991, Shah *et al.* 1995 and Lankaputhra *et al.* 1996).

Tamime and Robinson (1999) reported that the desired pH for yogurt is 4.6, which is partially cooled before adding fruit and flavours. Yogurt products are often blast chilled to <10°C in cold storage to decrease further acid production. Many commercial yogurts have solid milk content from 14-15 per cent.

Danone Vitapole (2000) reported that milk and milk products are considered an ideal food medium for delivering probiotics and other functional ingredients which are not found in regular curd as they are a good source of protein, calcium and a variety of vitamins, minerals and some bioactive compounds (Danone 2000).

Iniguez *et al.* (2001) reported that probiotic yogurt shows little organoleptic properties and a significant variation between experimental yogurt and probiotic yogurt in Cuba with 70 per cent of buffalo milk and 30 per cent cow milk stored at 4°C in half litre plastic container.

Lourens and Viljoen (2001) observed that fermented dairy products, i.e. yogurt are the most accepted food which contains live probiotics bacteria delivery to human GIT. Despite the importance of the viability of the probiotic bacteria, surveys have shown the poor viability of bifidobacteria in yogurt culture (Akalin *et al.* 2004).

Trachoo (2002) reported that yogurt contains a titratable acidity of not less than 0.90 per cent expressed as lactic acid. The addition of non-fat dry milk increases the calorific value of yogurt and acid production; however, about 50 per cent of non-fat dry milk is lactose (Kalab *et al.* 1983).

Lee and Lucey (2003) stated that the incubation temperature impacts the physical and microstructure of yogurt. The application of high incubation temperature resulted in a reduction in gelation time and 'G' value at 4.6 pH and an increase in

LTmax, B and whey separation compared with yogurt gels incubated at low temperature (Lee and Lucey 2004).

Songisepp *et al.* (2005) analyse the functional efficiency of anti-oxidative characteristics of probiotics in well-being subjects and found significant enhancement in the total antioxidant activity.

Güven *et al.* (2005) reported that the curd stability of yogurt is one of the most important physical characteristics. There are different factors which have influenced the stability of curd. These can be total solids, protein and acidity of yogurt, homogenisation, activity of starter culture and temperature of storage.

Chandan *et al.* (2006) stated that yogurt is produced by lactic acid fermentation of lactose by lactic acid bacteria, for instance, *L. bulgaricus* and *S. thermophilus*. The synergistic response of these two bacteria provides a specific texture, composition and organoleptic properties of yogurt. Fresh yogurt can be processed further into concentrated yogurt by partial removal of whey. Whey can be removed by traditional cloth bag or centrifugal separator method (Sumarmono *et al.* 2014). Concentrated yogurt is in semi-solid and with a creamy consistency, acid flavour and total solid content between 23-25 g per cent (Nsabimana 2005).

İsleten and Karagül-Yüceer (2006) reported that the pH value significantly reduced during the storage period for all the yogurt types from day seven onwards (Mohan *et al.* 2020). This can be ascribed to post-acidification, which is because of the continued production of organic acids in yogurts during the food product shelf-life by the starter culture bacteria.

Varga (2006) reported that the enrichment by the honey of yogurt as organoleptic properties. In the study, the 1 per cent honey-added yogurt sample was weak in flavour. On the contrary, the flavour of yogurt sample with 5 per cent honey was strong and was yogurt was very sweet. The honey added at the concentration of 3 per cent improves the sensory properties of the yogurt without the inhibitor effect on the starter bacteria.

In the study, the taste and consistency on mouth and appearance of yogurt improved with the increase of the honey content added (0%, 2% and 4 %) in the

yogurt on the 7th day of storage while the odor score of yogurt increased to 2 per cent honey ratio and decreased at higher honey rates.

Tojo Sierra *et al.* (2006) reported that yogurt had reduced fat content than milk because of dairy components used in preparation. Fatty derivatives are also produced by bacteria that contribute to the aroma of yogurt. Dairy products have a high level of conjugated linoleic acid (Fernandez *et al.* 2016).

Varga (2006) stated that plain dairy products have a sour taste and are hence not preferred by some consumers. Their flavours may need to be improved with sweeteners (Sert *et al.* 2011, Paucean *et al.* 2011). Various sweeteners can be added to yogurt, so today, most yogurts are sweetened using sugar (Aryana and Olson 2017).

Donkor *et al.* (2007) stated that a reduction in the apparent viscosity of symbiotic yogurt supplemented with resistant starch during refrigerated storage and the phenomenon was significantly affected by both types of probiotic bacteria and the probiotics under consideration. A steady decrease in apparent viscosity was also reported by Aryana and McGrew (2007) in probiotics *L. casei* yogurts with differing chain length oligofructose and inulin. This is due to the activity of bacterial enzymes on the casein micelles structure.

Hayaloglu *et al.* (2007) reported that the storage temperature significantly affected storage time. Yogurt stored at 25-30°C stayed for one day, at 7°C for five days and at 4°C for ten days. A minor difference with this study was the addition of fruits lengthens the storage time. The longer storage time in the yogurt was due to the increased acidity that prevents the proliferation of pathogen bacteria. An enhancement in the acidity level due to the digestion of milk sugar by bacteria which release the lactic acid as a metabolic product. Therefore, increasing milk acidity and causing coagulation, known as the denaturation process. Longer storage period of yogurt added with fruit due to higher acidity as well as metabolic bacteria or the fruit acidity (Yildiz 2010).

Tamime *et al.* (2007) reported that yogurt derived from milk other than bovine tends to diverge in various sensory and physiochemical properties because of differences in the milk composition. For example, yogurt derived from milk with a high-fat content (e.g. buffalo, sheep) has a more creamy texture compared to that

derived from milk with less fat content. While, the milk-producing animal species significantly influence the yoghurt's characteristics.

Aryana and Olson (2008) stated that the pH of yogurt was reduced during storage in cold storage between 3.8 and 4.5. The reduction in pH is because of the degradation of lactose in lactic acid (Hassan and Amjad 2010).

Korbekandi *et al.* (2008) reported that the probiotic count remained above 10^7 throughout the cold storage period ($5\pm 1^\circ\text{C}$) of 21 days in the probiotic yogurt.

Harisa *et al.* (2009) reported a significant reduction in MDA concentration after feeding diabetic rats with *L. acidophilus*.

Allgeyer *et al.* (2010) reported that when consumers have knowledge about the health benefits of probiotics/probiotics yogurt, the choice to consume this kind of food production increases. Approximately 37 per cent of consumers stated consuming yogurts with probiotics several times a month, whereas 42 per cent reported consuming probiotic yogurt.

Attita Allah *et al.* (2011) reported that the milk utilised for the preparation of set yogurt in Khartoum State, Sudan consist of whole milk powder or a mixture of recombined whole and skim milk powder. This study was carried out to evaluate the microbial count of yogurt during storage. High total bacterial counts were observed both in mixed (52.55×10^5) and whole milk powder (12.16×10^5) indicating the poor quality of milk. The coliform count was also highly significant in the yogurt up to 7 days of storage.

Bravo *et al.* (2011) reported that changes in gut microbiota in adult rodents with probiotics could modulate pain, brain biochemistry and behaviour. Another study found a similar pattern in humans.

Ejtahed *et al.* (2011) reported a significant reduction in the blood glucose and MDA level in type II diabetes mellitus after feeding probiotic yogurt.

Ekram *et al.* (2011) reported that yogurt is usually made from recombined whole and skim milk powder; however, some recent dairy industries commenced to use fresh milk instead of powder milk. Physico-chemical and microbial viability was analysed during the storage period of plain set yogurt. The mean value for the physic-

chemical properties as total solids, solid non-fat, fat, protein, ash content and titratable acidity 14.02, 10.95, 3.06, 3.86, 0.66 and 1.31 per cent, respectively. However, the mean value for the fresh milk yogurt were reported as 15.04, 11.51, 3.53, 4.42, 0.82 and 1.33 per cent, respectively. The chemical contents of yoghurt samples were significantly different due to variations in the preparation and type of milk used, while non-significant differences were found for titratable acidity due to variations in the type of milk used. Similarly, the storage period significantly affected Total Solids (TS), Solids Not Fat (SNF) and protein contents and titratable acidity.

Grenham *et al.* (2011) reported that the gut-brain axis is a bidirectional interface between the gastrointestinal and the brain. It is synchronised at hormonal, immunological and neural levels to maintain homeostasis and dysfunction of the axis leads to pathophysiological consequences. The repeated co-occurrence of stress-related psychiatric disorders, such as GI disorders and anxiety, also focuses on the significance of this gut-brain axis (Cryan and Dinan 2012 and Matsumoto *et al.* 2013).

Senaka *et al.* (2012) studied goat milk-based plain yogurt and stirred fruit yogurt by *L. acidophilus* LA-5, *Bifidobacterium animalis subsp. Lactis* BB-12 and *Propioibacterium jensenii* 702. Yogurts were stored at 4°C for four weeks and analysis was done weekly. *P. jensenii* shows the highest viability. Blending of fruit supports the viability of Lactobacilli. The blending of fruits significantly increases the syneresis and decreases the viscosity and water-holding capacity and enhances the organoleptic properties.

Cruz *et al.* (2013) reported the highly viable strains as *L. acidophilus* and *B. Longum* to cause slight changes in the physico-chemical and sensory parameters of yogurt during cold storage.

Mazloom *et al.* (2013) studied diabetic patients aged 25-65 years with fasting blood glucose ≥ 126 mg/dl which were diagnosed as having type II diabetes for less than 15 years and were eligible to study for the effect of probiotics. Probiotic strains used in the study were *L. acidophilus*, *L. bulgaricus*, *L. bifidum* and *L. casei* fed for six weeks. Results show non-significant declining trends in the level of triglyceride, IL-6 and insulin resistance.

Loveday *et al.* (2013) stated that yogurt could be stirred or set. Stirred yogurt is the consequence of a mechanical effect, such as shearing after fermentation. The set yogurt usually is fermentation in a pack, achieved a firmer structure compared with the stirred yogurt. Other kinds of yogurt are plain yogurt, fruit-flavoured yogurt, whipped yogurt, granola-topped yogurt, frozen yogurt, drinkable yogurt and Greek yogurt. These yogurts can have different levels of fat content (Cassel 2014).

Mehdia *et al.* (2013) analysed that two types of milk powder were used to prepare from skimmed milk powder at 15 per cent and whole milk powder at 13.7 per cent and ground linseed. Results show that the addition of ground linseed decreased the pH levels and elevated the titratable acidity during fermentation and cold storage. The organoleptic score shows that the supplemented yogurt had better texture, taste and odour scores compared with the control. The microbial profile of yogurt with linseed supplements significantly increases the lactic acid bacteria count during storage.

Mukdsi *et al.* (2013) Consumption of goat milk cheese containing *L. Fermentus* CRL1446, which increased the intestinal feruloyl esterase function, which resulted in increased ferulic acid bioavailability, improved oxidative status and defence against stress-related disorders. Patients who consumed probiotic goat milk yogurt 300 gm per day for six weeks showed that blood glucose levels and antioxidant levels increased.

Tillisch *et al.* (2013) show the effect of the consumption of fermented milk on humans containing probiotic mixture as *B. animalis* subsp *Lactis*, *L. bulgaricus*, *Streptococcus thermophilus* and *Lactococcus lactis* Subsp *lactis* on gut-brain interactions. Results show brain activity, which plays a vital role in controlling emotions and sensations after feeding fermented milk to healthy women. The study clearly shows the association between the consumption of probiotics and inflexion of brain activity and also gives evidence of the modulatory impact of probiotics on gut-brain interaction.

Vera and Rodriguez (2013) stated that the traditional yogurt preparation process is basically based on the microbial acidification of milk because of lactic acid bacteria, which use the milk lactose and generates lactic acid. This reduces the pH of

milk to 4.5 and reaches the isoelectric point of protein which coagulate with typical lactic flavour and with a specific aroma yogurt obtained (Andrade *et al.* 2010).

Hussain (2014) reported that the shelf life of acidified milk was ten days and for orange-flavoured yogurt was eleven days. During storage, primary attributes are affected as flavour, odour and the development of fungal growth. The viability of bacteria in yogurt and probiotics was assessed for twenty-one days.

Mani-Lopez *et al.* (2014) reported that the food industry wants to expand the range of probiotics yogurts, but each probiotic bacteria offer different and specific health benefits. Little information exists on the influence of probiotic cultures on the physico-chemical characteristics of yogurts and fermented milk. Six samples of yogurt with one control have been analysed for physiochemical and microbial properties. Starter cultures were used as *L. delbrueckii* *S. bulgaricus* and *S. thermophilus* and probiotics *L. acidophilus*, *L. casei* and *L. reuteri* were used. Reduction in pH (0.17 to 0.50 units) and enhancement in titratable acidity (0.09 to 0.29%) were reported during storage. The yogurt with *S. thermophilus* and *L. delbrueckii ssp bulgaricus* and *L. reuteri* was different in firmness. Probiotic cultures also lost viability during a storage period of three weeks.

Marion *et al.* (2015) studied to evaluate the possibility of the addition of Cupuassu pulp probiotic culture *L. acidophilus* LA-5 and probiotic inulin to improve the texture of goat milk yogurt in the six treatments. Those six treatments were as: natural (N), prebiotic (Pre), probiotic (Pro), cupuassu (C), synbiotic (S), and probiotic with cupuassu (PC). The microbial viability of probiotic in yogurts of Pro, S, and PC was analysed. Along with these, some instrumental analyses were also done as pH, colour, apparent viscosity and texture, to measure the effect of these different ingredients on goat milk yogurt. These probiotic bacteria remained viable ≥ 7 log CFU /ml throughout the 28 days of storage. Apparent viscosity and firmness are reduced in PC yogurt. The consistency was highest in the yogurt with added probiotics compared with other yogurts during the storage period. The cohesiveness remained constant in all the yogurt samples. The study concluded that cupuassu pulp amassing improves the texture of goat milk yogurts. Therefore, this pulp could be an imperative technological strategy for the dairy goat industry.

Zahra *et al.* (2015) studied innovative functional food like low-fat yogurt enriched with phytosterol. Phytoestrol dispersion was prepared by using oil/water emulsion. The emulsion was added to yogurt milk. Various physico-chemical properties like pH, Titratable acidity, syneresis, firmness and apparent viscosity of enriched yogurt were analysed during the storage period. Results show that the enriched yogurt, apparent viscosity and syneresis were lowered and firmness was higher compared with the control. The addition of phytosterol to the yogurt had a significant effect on acidity. The distribution of phytosterol in different parts of one sample was not uniform. An organoleptic result shows that there was no significant difference between control and enriched yogurt samples in appearance, flavour, texture and overall acceptability.

Aziri- Anper, (2016) stated that pH is an indicator of the presence of lactic acid and organic acid. This is a key factor in ensuring the quality of yogurt. Recant research found the pH declined during the shelf life of yogurt due to the activity of lactic acid bacteria, producing lactic acid from lactose. Also, other organic acids are formed from the residual enzyme formed by starters during fermentation (Damin *et al.* 2009, Christopher *et al.* 2009). Titratable acidity values are because of lactose fermentation by lactic acid bacteria (Paseephol *et al.* 2008).

Fernandez *et al.* (2016) reported that lactose is the main carbohydrate present in dairy products which give ascends to lactic acid after fermentation. Based on the type of product and industrial additives, this disaccharide can reach up to 98 per cent of the total carbohydrates in natural yogurt.

Mokoonlall *et al.* (2016) reported mainly two types of yogurt (I) Firm/Set yogurt is obtained directly after fermentation (II) Stirred yogurt is obtained with an addition post-fermentation process, in which yogurt is mixed and smoothed. The viscosity values of yogurt are affected by several factors starter cultures, the composition of milk and heat treatment (Velez-Ruiz *et al.* 2012, Mohan *et al.* 2020). Likewise, as the total solids enhance, the viscosity and firmness of the yogurt improve (Tarrega *et al.* 2016).

Wulansari and Andri (2016) evaluated the composition (total solids, water, fat and protein content), qualitative characteristics (Aroma, Colour and texture) and quantitative properties (Free fatty acid and lactic acid) of cow milk yogurt with the

addition of different fruit for 15 days storage period. The results showed that the addition of fruit significantly affects the composition and properties, whereas storage time affects the quantitative characteristics.

Ibsen *et al.* (2017) studied the relationship between the subgroup of dairy products and the risk of type II diabetes mellitus. Data used from the Danish diet comprise 54 men and 277 women aged 50-64 years. Whole-fat yogurt products in place of low-fat milk, whole-fat milk or buttermilk were related to a lower rate of type II diabetes. The findings of study reported that intake of whole-fat yogurt in place of low-fat yogurt products is related to a lower rate of type II diabetes mellitus.

Janny (2018) stated that the reduction in the pH of yogurt could have influenced the viscosity of yogurt. As pH decreases, the distance from the isoelectric point of casein increases, reducing the stability of this protein and reducing curd stability (Bylund, 1995).

Kishor *et al.* (2018) stated that the moisture content of T₀ (100 % goat milk) ranged from 86.03 -87.70 per cent, T₁ goat and cow milk (60 + 40) ranged from 85.60-86.74 per cent, T₂ goat and cow milk (70 +30) 86.30-85.26 per cent T₃ goat and cow milk (80+20) ranged from 85.22-86.46 per cent. The results showed that treatment T₁ was best in moisture per cent. The difference in the score for moisture of yogurt was significant.

2.7 GOAT MILK AND ITS HEALTH BENEFITS:

Time Magazine (1931) reported that selenium, a trace mineral essential for a healthy immune system and has antioxidant qualities, is prominent in goat milk. According to legend, Mahatma Gandhi kept himself healthy by consuming raw goat milk following protracted fasts.

Macy (1949) reported that goat milk had a lower freezing point and refractive index compared with bovine milk. Goat milk contains higher acidity in fresh as well as in thermally processed compared with bovine milk.

Parkash and Jenness (1968) stated that goat milk's density is comparable to that of cow milk's, however it has lower refractive index and freezing point and higher specific gravity, viscosity, and titratable acidity than cow milk (Haenlein and

Wendorff, 2006). Goat milk has a freezing point of roughly -0.580°C , a viscosity of 13.4 mP at 27°C , and a titratable acidity as lactic acid that ranges from 0.11 to 0.18 per cent (Roy and Vadodaria, 2006).

According to Zeman (1982) total of 40 per cent of people who are allergic to cow's milk proteins can tolerate goat's milk protein. Goat milk also has a higher mineral concentration, and these features inhibit bone demineralization. In comparison to cow and human milk, goat milk is better at being digested, has a larger buffer capacity, is more alkaline, and has medicinal benefits. Compared to cow milk, goat milk fat exhibits higher surface tension, viscosity, and specific gravity levels (Park *et al.* 2007).

Maraval and Vignon (1982) observed significant changes in the major mineral concentrations of goat milk throughout the first seven weeks of lactation. Overall, goat milk has less Na and S and more Ca, P, K, Mg, and Cl than cow milk (Haenlein and Caccese, 1984, Park and Chukwu, 1988, Chandan *et al.* 1992).

Park and Chukwu, (1988) reported that milk from goats and sheep has far more minerals than milk from humans. Human milk only contains a quarter to a sixth of these essential minerals, compared to goat milk's 134 mg Ca and 121 mg P/100 g. Although macro-mineral concentrations may not change considerably, they depend on the breed, nutrition, individual animal, lactation stage, and condition of the udders.

Prasad *et al.* (1990) reported that the method of manufacturing paneer from goat milk has been studied and standardised at Central Institute for Research on Goats. Fresh goat milk is heated to $87\text{--}88^{\circ}\text{C}$, and then citric acid granules (0.15% W/W) are added to coagulate the milk. According to reports, the composition of the milk from which paneer was made had an influence on the yield (13.31-19.34%), moisture (42.22-51.80%), protein (17.86-21.88%), fat (24.50-29.50%), and ash content (1.53-2.23%). Freshly made paneer didn't taste salty or have a goaty smell. The paneer's body had a smooth, semi-hard consistency and no air bubbles. The product enjoyed wide acceptance and high consumer demand (Pal *et al.* 1994, 1997; Agnihotri and Pal, 1996, Agnihotri and Pal, 1997).

Redmond *et al.* (1998) In addition to the central nervous system, taurine is important for the production of bile salts, osmoregulation, antioxidation, and calcium transport. Compared to cow milk, goat milk has 20-40 times more taurine.

Clark and Sherbonb (2000) stated that human milk does not include α 1-CN, goat milk has a casein profile that is more similar to that of human milk than that of cow milk (Silanikove *et al.* 2010). Milk from goats with modest alleles has the potential to make milk for people with allergies (Roncada *et al.* 2002). Albenzio *et al.* (2009) proposed that such milk might be utilised for feeding CMPA subjects due to the high prevalence of weak and null alleles for the α 1-CN locus and the high frequency of the A0 genotype at the α 2-CN locus in the Garganica breed population.

Bozanic *et al.* (2001) reported that fermented beverages were made from goat milk by two per cent of inoculums of yogurt culture DVS-YC 180 (*Streptococcus thermophilus* and *L. delbrueckii subsp. Bulgaricus*).

Guo *et al.* (2001) reported that goat milk's chemical composition is mainly seasonal in its variation. The primary components of goat milk are high in the early lactation stage, decline quickly, stay low for an arbitrary amount of time, and then rise again toward the conclusion of lactation.

Drakoularakou *et al.* (2003) studied the better development of *L. acidophilus* in sheep milk ($> 10^9$ CFU/ ml) compared to goat milk or cow milk ($> 10^8$ CFU/ ml) after 12 hours of incubation at 37 °C, representing the impact of variation in the constituent between a different kind of milk on probiotic viability. The higher acidity development in goat milk compared with cow milk and sheep milk.

Tziboula-Clarke (2003) reported that the distinct "goaty" flavour of goat milk, which is a crucial trait, is attributed to the goat milk's different fat composition and its comparatively more considerable amount of short- and medium-chain fatty acids. Additionally, some of the few technological issues with processing goat milk into value-added products include salty flavour, poor scum formation due to tiny fat globules, a lack of agglutinating euglobulines, and difficulty in fat separation (Agnihotri and Prasad, 1993).

According to Lara-Villoslada *et al.* (2004) reported in an in-vitro investigation, goat milk protein does not possess the same ability to attach to IgE and IgG as cow milk protein has. Cow milk has been demonstrated to increase lymphocyte proliferation, IL-4 production, histamine secretion and IgG production (Sanz Ceballos *et al.* 2009; Lara-Villoslada *et al.* 2004) which are all factors indicating more extended allergic inflammation.

Rathinam, (2005) stated that the demand for organic and healthier dairy products is rising globally, especially in India. The estimated 14180\$ million market value of these products in developed nations is growing substantially, on average, by 10-12 per cent yearly. The predicted elevation in some nations will exceed 30 per cent. India is the world's top producer of goat milk and will gain more from this trend of customer interest in milk free of harmful residues because natural pasture grazing of goats is the norm. Goat milk can be utilised as a major raw material for manufacturing gourmet cheeses in India, which is developing its cheese industry significantly and following France's example. France produced 68,000 tonnes of goat milk cheese in 1999, of which more than 30% was exported and the remaining portion was consumed domestically (Tziboula-Clarke, 2003). Density of goat milk is higher than goat milk, but is lesser than in sheep milk, whereas both have more specific gravity, viscosity, titratable acidity, but lesser freezing point and refractive index cow milk (Parkash and Jenness, 1968, Haenlein and Wendorff, 2006).

El-Agamy (2007) found that in terms of protein composition, goat milk is comparable to human milk. Beta casein, a key casein protein, is different from casein in cow milk and is present in goat and human milk. Additionally, goat and human milk have entirely different alpha- and beta-lactoglobulin peptide mappings from cow milk.

Park *et al.* (2007) reported that goat milk's casein curd, which is softer and smaller than that produced by cow milk, which provides it with its high level of digestibility. Because of this, the human digestive system can absorb and provide its more digestibilities. Since the fat globules effects dispersion and a more homogeneous mixture of fat, their size also influences digestion. The globules' enormous overall surface area makes it simpler for enzymes to access and interact with the lipids (Attaie and Richter, 2000).

The sensory qualities of the fermented goat milk products made in an experiment by Ranadheera *et al.* (2008) containing *Lactobacillus acidophilus* LA-5, *Bifidobacterium animalis* subsp. *lactis* BB-12, and *Propionibacterium jensenii* 702 were not negatively impacted. Probiotics in fermented goat milk beverages have been successfully preserved by adding collagen hydrolysate, cheese whey, and acai pulp (*Euterpe oleracea*) (Rigoto *et al.* 2019).

Conesa *et al.* (2008) reported that goat milk contains higher vitamin A content than cow milk because goats convert all β -carotene from foods into vitamin A in the milk. Goat milk is always whiter than cow milk because of the same reason. Low quantities of vitamin B₆ and vitamin D, both of which are crucial during infancy, are present in both goat and cow milk (Juarez *et al.* 2011).

A mixed starter culture of *Streptococcus thermophilus* CR12, *Lactobacillus helveticus* PR4, and *Lactobacillus plantarum* 1288 was used to create fermented goat milk by Minervini *et al.* (2009). These cultures caused the production of GABA, a signalling substance that inhibits the central nervous system, as well as an in vitro ACE-inhibitory activity, which reduces high blood pressure.

Park (2009) reported that goat milk contains bioactive lipids that have an impact on the immune system. First, the health benefits and bioactive properties of conjugated linoleic acid (CLA) in humans are diverse. Linoleic acid has several isomers, the two most prevalent of which are cis-9, trans-11 (c9, t11) and trans10, cis-12 (t10, c12), collectively referred to as CLA. CLA is a crucial bioactive component of goat milk and is naturally present in ruminant milk and meat. In terms of triggering an immunological response, CLA has been discovered to alter immune mediators such as cytokines, eicosanoids, prostaglandins and immunoglobulins. Additionally, CLA has the capacity to lower allergy-related immunoglobulin IgE in individuals, indicating the lipid's potential to be anti-allergic. Moreover, it has been demonstrated that CLA has anti-inflammatory properties by reducing the body's production of pro-inflammatory cytokines linked to cancer, atherosclerosis, irritable bowel syndrome and other immunopathologies.

Christensen (2010) reported the properties of goat milk, like alkalinity, buffering capacity and digestibility, diverge from bovine and human milk. It also contains various health benefits which make it useful in human medicine and diet.

Despite of the beneficial effects, consumer dislikes goat milk and its products because of its goaty flavour and odour. Fermentation makes it acidified which affects its organoleptic profile. Goat milk contains an average smaller casein level compared with bovine milk. Along with this, goat milk also contains a lesser concentration of s1- casein and a higher degree of casein micelles dispersion which impact its product's rheological properties.

Nitric oxide (NO) and cytokine release in human blood cells were evaluated by Jirillo *et al.* in the 2010 study. The results demonstrated that goat milk might trigger cytokine and IL-6 release along with NO release from blood cells. In addition to having cardiac protective benefits for milk consumers, the NO release can also reveal antimicrobial activity, which helps to avoid infections. It is a pro-inflammatory cytokine, IL-6 is an acute phase TNF- reactant and development element for B cells and IL-10 is an anti-inflammatory cytokine. The mix of released cytokine has been proposed to sustain immune homeostasis by goat milk.

Lopez-Aliaga *et al.* (2010) reported that goat milk's high concentration of short- and medium-chain fatty acids makes it easier and more rapidly to digest. Goat milk contains medium chain triacylglycerols, which are not degraded or refined and are instead absorbed intact in the colon. Since the molecules are taken up directly into the portal vein, the production of micelles is not required for absorption.

Plevina *et al.* (2010) reported that goat milk contains a good quantity of protein, fat, vitamin and minerals compared with cow milk. Goat milk contains approximately more than 25 per cent vitamin B6 and, 13 per cent calcium, 47 per cent vitamin A. It also contains three times more niacin, one and half times more potassium and four times more copper. Goat milk has better antirahiticheskin properties because of the high content of calcium, phosphorus, cobalt, copper, selenium, magnesium, magnesium, iron manganese, and sialic acid, which play a vital role in maintaining immunity.

Riberio and Ribeiro (2010) reported that goat milk is an excellent source which provides benefits to maintain health, physiological functions and helps to improve the nutritional level in children and elderly people. It can be recommended to those people who are allergic to cow milk.

Slacanac *et al.* (2010) reported that goat milk has a more digestible, mineral bioavailability and fat and protein profile compared to cow milk and the consumption of probiotic fermented goat milk has been attributed as therapeutic food (Minervini *et al.* 2009).

Tamime *et al.* (2011) evaluated the landscape of goat-fermented milk, where the majority of goods are historically created locally in various Middle Eastern and other countries, while some are produced on an industrial trial scale. The name of the fermented milk and yoghurt differ from country to country and location, and the local strains of LAB (traditional and adjunct, including probiotic organisms), yeasts, and moulds (or combinations of these) used locally have an impact on the quality. They mentioned dry fermented milk such as Kurut, Keş, and Kiş, as well as Italian Gioddu, Chinese Tarag, Caprine probiotic yoghurt, kefir, concentrated yoghurt Labneh, salted yoghurt Tuzlu, and Shankleesh.

Senaka *et al.* (2012) reported that yogurt made from bovine milk is usually consumed, but there is a yearning to find an alternative to cow milk due to some gastrointestinal issues and demand for novel dairy products. Goat milk contains more digestibility and less allergic properties compared to cow milk.

Senaka *et al.* (2012) reported that goat milk is more easily digested than cow's milk, making it suitable for lactose intolerant. Compared to cow's milk, goat's milk is easier to absorb, leaving less undigested material in the colon to literally ferment and result in the painful symptoms of lactose intolerance.

By treating milk with the enzyme -galactosidase, Pruksasri and Supee (2013) assessed the viability of creating goat milk that contains galactooligosaccharides (GOS). The findings showed that at pH 4.5 and 40°C, the greatest GOS produced was roughly 13.9 per cent of total sugars. When compared to conventional goat milk, goat milk with a greater GOS concentration tasted significantly better and was more widely accepted (control). Additionally, goat milk with GOS demonstrated good stability in an acidic environment. After being subjected to high heat treatment and shelf life conditions, GOS in goat milk remained stable.

Prebiotics and probiotic bacteria combined with each other, according to Noelia and Enzo (2014), enhance the quality of food preparation. The features of goat

milk and symbiotic yoghurt were investigated in this research. The skimming procedure was examined and improved using raw goat milk. Inulin, gelatin, sugar, *Streptococcus salivarius subsp. thermophilus*, *Lactobacillus delbrueckii subsp. bulgaricus*, and *Lactobacillus casei subsp. rhamnosus* were used to create possible non-fat symbiotic yoghurt. The lactic acid bacteria and probiotic culture's chemical properties, acceptability, and viability were evaluated. The raw milk had a protein and fat level of 2.90 and 3.56 g/100 ml, respectively. The ideal skimming procedure was accomplished for 15 minutes at 4°C and 9,800 rpm. The created product had a 4.04 to 0.04 g/100 mL protein and fat content, acceptable sensory qualities, and a 95 per cent acceptance rate. The probiotic culture count was higher than 1×10^6 CFU ml⁻¹ and the lactic bacteria count was higher than 9×10^7 CFU ml⁻¹, which ensures their effectiveness and ability to propagate in the colon. Throughout the 21 days of storage, the yoghurt remained steady. As a result, this study demonstrates that goat milk yoghurt is a reliable source of the probiotic *L. casei* culture and inulin.

Costa *et al.* (2014) Although goat milk yoghurt is a fantastic source of fatty acids, protein, and minerals, and many consumers do not like the flavour that these milk and dairy products' caprylic, capric, and caproic acids provide it.

Nurliyani (2015) reported the effects of kefir mixed with soy milk and goat milk on plasma glucose levels, glutathione peroxides and lipid profile and the improvement in the pancreatic β -cells in diabetic rats. Outcomes of the study revealed that diabetic rats fed with goat milk and soy milk kefir had reduced the triglyceride than the rats fed kefir with the combination. Reducing of plasma in diabetic rats fed goat milk kefir was reduced more than rat fed kefir mix.

Nurliyan *et al.* (2015) studied the viability of the microorganisms was not negatively impacted by replacing goat milk with black rice extract in the kefir fermentation, but the pH may have decreased. Whenever black rice extract or inulin is used in place of goat milk, the antioxidant activity of kefir may be increased. The alcohol and total phenolic contents of the kefir increased during the seven days of storage at low temperatures, whereas the pH declined. The research demonstrates that the effects of a minimum 2.0 ml dose of kefir made from goat milk and black rice extract for the treatment of diabetics will be comparable to those of the anti-diabetic drug glibenclamide.

Kumar and Sharma (2016) studied that goat milk has the highest content of L-glutamine of any other milk product and due to that, the blood's pH is raised. L-glutamine is an alkalizing amino acid. Because goat's milk is a more effective acid buffer than cow's milk, it has long been used and advised as a supplement in the treatment of ulcers. The digestive system is soothed by goat's milk. Goat milk has been found to improve children's sleep patterns and help them feel fuller between meals.

Streptococcus thermophilus and *Lactobacillus* were combined by Sanna *et al.* (2005) to produce yoghurt with a significant amount of folate and positive sensory characteristics. Goat milk, cow milk, and a combination of the two were used to make concentrated yoghurt (Labneh). When compared to Labneh, made using cow's milk, made from goat's milk had higher moisture, ash, and fat contents but lower pH, total solids, protein, and lactose contents. When compared to Labneh made simply with cow's milk, Labneh made with goat's milk contained higher levels of short- and medium-chain fatty acids and lower levels of long-chain FAs. The sensory panel's best options were samples made with 40 per cent goat's milk and 60 per cent cow's milk (Serhan *et al.* 2016).

Bhat *et al.* (2016) studied a distinctive goat milk bar that was designed using two distinct kinds of natural sweeteners and rose flower extracts as an added component (Addition levels: 0, 5, 10, 15 and 20%). (Cane sugar and palm sugar). The nutrition bars' near composition, textural characteristics, and sensory attributes were assessed. The results were positive, and the novel nutritional bar recipe made with goat milk and rose extracts opens the possibility for potential future commercial use of the item. Goat milk's high nutritional content and rose extracts' dual roles as a natural antioxidant and antibacterial agent can help this unique product persist longer at room or refrigerated temperatures, which will attract more customers.

International Dairy Meet (2016) Brazil's goat industry is developing and has had a remarkable expansion in recent years, particularly in the northeast and southeast. Only 26 million of the 35 million litres of goat milk that Brazil now generates each year are industrialised. Due to its superior digestibility, hypoallergenic, nutritional value, and therapeutic and dietary qualities, known in the nutrition of children and the elderly, goat milk is significant as a functional food.

Matias *et al.* (2016) studied goat milk cheese which contains *L. fermentum* CRL 1446 resulted in increased intestinal feruloyl esterase (FE) function, which resulted in augmented ferulic acid bioavailability, and improve oxidative stress. Goat milk products were also found to be beneficial for intestinal micro-biota. Consumption of fermented goat milk (yogurt 300 ml) contained *L. ferementum* ME-3 for 21 days results found anti-atherogenic symptoms and improved gut micro-biota and anti-diabetic effects.

Popovic-Vranjes *et al.* (2017) studied customers who like goat milk and items manufactured from it because they have positive health effects. Customers desire wholesome, safe for consumption, aesthetically pleasing, and nutritious (Linares *et al.* 2017). Consumer demands are met by products made from goat's milk, such as fermented goat's milk (Mituniewicz-Malek *et al.* 2019), goat's milk yoghurt (Beltran *et al.* 2018), and goat's cheese (Barlowska *et al.* 2018).

Aryana *et al.* (2017) reported a significant decline in insulin growth factor and fasting blood glucose due to feeding kefir for ten weeks.

Kumar and Sharma (2017) reported that fresh goat and fresh cow milk seem precisely the same after being chilled in a glass overnight, but the cow milk separates into two phases, with cream on top and skim milk at the bottom. This is a typical occurrence brought on by the substance agglutinin. In order to maintain the homogeneity of cream and skim milk, cow milk is mechanically homogenised to break down the cell wall of the fat globules.

Kumar and Sharma (2017) reported that fresh goat and fresh cow milk seem precisely the same after being chilled in a glass overnight, but the cow milk separates into two phases, with cream on top and skim milk at the bottom. This is a typical occurrence brought on by the substance agglutinin. In order to maintain the homogeneity of cream and skim milk, cow milk is mechanically homogenised to break down the cell wall of the fat globules. Superoxide is released by this mechanical homogenisation (free radicals). The body may experience a variety of issues as a result of these free radicals, including mutation. Goat milk is naturally homogeneous due to its smaller fat globules and lack of agglutinin, which eliminates the problems brought on by mechanical homogenisation techniques.

Albano *et al.* (2018) reported the one approach is connected to the cholesterol-lowering effects of certain LAB. Seven LAB strains have been identified to have the ability to lower cholesterol, first in broth and later in cheeses during ripening when utilised in the production of goat yoghurt and cheese from raw milk. With the exception of *L. plantarum* VS166 and VS513, it was observed that the cholesterol-lowering property was more significant in cheeses ripened for 60 days than for 30 days. Additional research on autochthonous LAB and their strains may result in better traditional goods or novel functional dairy goat products.

Kishore *et al.* (2018) conducted a study to develop yogurt from cow milk and goat milk. Control was prepared with (100 %) goat milk, T₁, T₂ and T₃ was prepared with 60:40, 70:30 and 80:20 per cent, respectively. T₃ sample of yogurt found the best and different parameters as protein (3.40 %), fat (4.82 %), lactose (4.54 %), moisture (85.72 %), ash (0.71%), total solids (14.28 %) and acidity (0.76 %) was respectively.

KSH, (2018) reported that in Hungary, milk is the primary product of the goat industry. An estimated 3-5 million litres of goat milk are produced annually. However, only 0.6 million litres are produced in milk factories. Goat milk items are made in factories and milk is the major source of income for farmers who raise goats for milk. It is typical for goat breeders to sell their goat milk and goat milk-based goods directly to consumers (Hungarian Chamber of Agriculture, 2017). In 2017, Hungary produced 3.5 million litres of goat milk.

Mazroea *et al.* (2018) studied the novel foods are among the products manufactured with goat's milk. Goat's milk is regarded as a valuable dairy product. The purpose of this research is to compare the composition of cow's milk and goat's milk formulas to breast milk in order to highlight the value of goat milk formula, which has higher levels of calcium, magnesium, and phosphorus than cow's milk formula but lower amounts of vitamin D and B₁₂ than cow's milk formula. Although there is a non-significant difference among the protein, fat and lactose contents of cow's and goat's milk, the proportion of protein and fat in goat milk formula is lower than that in cow milk formula.

Ranadheera *et al.* (2018) shown that goat milk is an effective probiotic LAB carrier in cheese, yoghurt, fermented milk, milk beverage, ice cream, and other foodstuffs because these dairy products can promote bacterial viability and high

survival levels during storage. The functional attributes of probiotics, including gastrointestinal tolerance and adherence to the intestinal epithelium, can be enhanced by dairy goat products. However, some LAB strains can produce volatile aromatic compounds that might enhance the sensory profile of dairy products, making them occasionally too "goaty" for consumers. As a result, these LAB strains can pose difficulties with the sensory profile of dairy products.

2.8 STEVIA AND ITS PRODUCTS AND HEALTH BENEFITS:

Parpinello *et al.* (2001) reported that using a combination of stevia (160 mg/L) and sugar (56 g/L) in the formulation of peach juice, resulted in a 25 per cent reduction in calories without impairing the product's sensory qualities compared to the control sample, which contained 9 per cent sucrose.

Rubio *et al.* (2007) reported that stevia was chosen in consideration of the strategy to avoid the weight gain that results from diets with low energy density (Fernandez *et al.* 2012). It is a plant with a sweetening power nearly 300 times greater than cane sugar (Lemus *et al.* 2012). It's been used for several things, especially in managing diabetes, because it lowers plasma glucose and insulin levels, suggesting that stevia may help regulate glucose (Lemus 2012). Since this stevioside does not stimulate appetite and has little to no acute toxicity, using it as a supplement is safe (Geuns 2004) and there is no risk of gaining weight (Anton 2010).

This study aimed to make and assess a functional yoghurt cake made with stevia leaves and sweetened with hot water extract. The typical yoghurt cake's ingredients were swapped out for healthier alternatives that also served a purpose. When substituting sweeteners, we used hot water extract of stevia in place of sucrose, butter was substituted by olive oil, full cream milk was substituted by skimmed milk, the whole egg was substituted by egg white and 72 per cent extraction wheat flour was substituted by whole wheat flour. Orange peels and lemon rind were also mixed with the formulated yoghurt with stevia extract. Organoleptic evaluation for the appearance, colour, odour, flavour, texture, and sweetness of the regular yoghurt cake and formulated yoghurt cake for both products were conducted.

According to Panpatil and Polasa (2008) stevioside are stable throughout a wide pH and temperature range. When dissolved for more than two hours at 60°C,

they remained stable and showed no signs of degradation under pH ranges of 1 to 10, but a very slight loss of up to 5 per cent has been recorded when heated to an elevated temperature of 80°C with pH ranges of 2 to 10. However, when exposed for two hours to an extremely acidic environment with a pH of 1 and a temperature of 80°C, the steviosides completely decomposed (Abou-Arab *et al.* 2010).

Abdel-Salam *et al.* (2009) studied which aimed to make and assess a functional yoghurt cake made with stevia leaves and sweetened with hot water extract. The typical yoghurt cake's ingredients were swapped out for healthier alternatives that also served a purpose. When substituting sweeteners, we used hot water extract of stevia in place of sucrose, butter was substituted by olive oil, full cream milk was substituted by skimmed milk, the whole egg was substituted by egg white and 72 per cent extraction wheat flour was substituted by whole wheat flour. Orange peels and lemon rind were also mixed with the formulated yoghurt with stevia extract. Organoleptic evaluation for the appearance, colour, odour, flavour, texture, and sweetness of the standard yoghurt cake and the diabetic-friendly variation for both products were shown. The obtained results showed that the regular yoghurt cake and formulated yoghurt cake for diabetics hold a good score. The biological analysis of rat blood parameters of control and yoghurt cake for diabetics groups showed that bilirubin, high-density lipoprotein (HDL) cholesterol, triglycerides, cholesterol, creatinine, alkaline phosphatase, blood glucose and gamma-glutamyltranspeptidase (γ -GT) values (means) in yoghurt cake for diabetics group were similar to results obtained in regular yoghurt cake group. There was a slight decline in urea and aspartate aminotransaminase (AST) (37.05 mg/dl and 86.82 U/L, respectively) values in yogurt cake for the diabetics' group compared with the control (43.26 mg/dl and 97.00 U/L, respectively). It was concluded that a prepared functional yoghurt cake possessing hot water extract of stevia, whole wheat flour, egg white, skimmed milk, yoghurt, orange peel lemon rind and olive oil would be a fabulous meal for diabetics.

Gardana *et al.* (2010) studied that stevia has the ability to regulate the heartbeat for cardiopulmonary signals and to control blood pressure. Using hot water to extract stevia leaves can control a person's blood pressure. Numerous investigations have shown that stevia and its constituents have hypotensive and diuretic properties. Stevia functions at the membrane level in a manner similar to blood pressure-lowering medications, which are known for their ability to lower blood pressure by widening

artery walls. Numerous research findings indicated that stevia could reduce blood pressure by extending the arteries.

Giri *et al.* (2012) researched kulfi which prepared with the addition of stevia indicated the highest overall acceptability, whereas no significant effect was observed on the body, texture, and flavour of the kulfi if the level of replacement of sugar with stevia was less than 50 per cent. However, a negative impact on the textural effects and flavour was regarded when the level of sugar replacement with stevia exceeded 50 per cent.

Gupta *et al.* (2013) reported that stevia has been found to increase the insulin action on cell membranes, increase insulin production, stabilise glucagon secretion, lower post-prandial blood sugar levels, and improve glucose tolerance to taken carbs in humans and animals. Stevia offers a wide range of processes that affect type II diabetes and its ultimate problems. As a result, sugars can be replaced with SGs or Stevioside from the Stevia leaf to facilitate optimal glucose management.

Mehrotra *et al.* (2014) evaluated the impact of sugar replacement on Shrikhand's chemical composition. The mixture with 30% sugar and 20% stevia extract was considered the most acceptable. The sample and the control were preserved in refrigeration for up to 21 days. Samples containing 30 per cent stevia leaves and 100 per cent sugar TPC count were measured during storage conditions every seven days. The mixture containing 100 per cent sugar and 30 per cent stevia leaves initially displays identical TPC values. With longer storage times, an increase in TPC count was seen. The control Shrikhand sample mean TPC ranges from 1.34 to 2.67 CFU g⁻¹ and from 1.00 to 2.34 CFU g⁻¹ for control Shrikhand. The Shrikhand samples can be kept in a refrigerator for three weeks on the conditions of microbial characteristics.

Miller and Perez (2014) reported that obesity, a nutritional disorder in which excess body fat accumulates in various body areas, is the most common nutritional problem worldwide. Clinical definitions of obesity also include cases where the excess body weight exceeds 20 per cent of the body's optimum weight. Overeating, bad food choices and eating habits, and physical inactivity have all contributed to the disturbing rise in obesity. Obesity is inversely correlated with caloric intake; the more calorically rich the diet, the more common obesity becomes. In this situation, it is

important to establish an effective weight-management strategy that reduces the likelihood of obesity. Low or zero calorie sweeteners are the greatest choices for people who have high fondness towards desserts.

Pradhan (2016) reported that since stevia has no calories and does not cause blood sugar levels to surge, observations indicate that one g of its crude extract is 100–150 times sweeter than sucrose.

Tondare *et al.* (2019) reported that the consumers in Gujarat prefer the sweet form of amrakhand, while those in Maharashtra prefer the tangy/sour variety. Amrakhand is a variation of shrikhand mixed with pureed mango and is popular in the western portion of the southern peninsula of India. Amrakhand's high sugar content increases the total amount of calories consumed; carbohydrates are the food group that dieters and diabetics are most limited from eating. There is a strong likelihood that stevia, a natural sweetener with no calories and beneficial effects on human health, may be used in the food sector. The study's major aim was to examine the sensory quality of Amrakhand, which was made using powdered stevia leaf extract at different dosages. Based on the amount of chakka, six alternative ratios of sucrose and stevia were used: 30:70 (T₁), 25:75 (T₂), 20:80 (T₃), 15:85 (T₄), and 10:90 (T₅). The 100:0 sucrose and stevia leaf extract powder concentration was used to create the control sample (T₀). The recommended order of the samples was as follows: T₁>T₂>T₀>T₃>T₄>T₃>T₄>T₅. Sensory qualities (colour and appearance, flavour, mouthfeel, body and texture, taste, and overall acceptability) were assessed by a panel using a 9-point hedonic scale. The judging panel found Amrakhand with treatment T₁ (30:70) to be more acceptable.

Marjan *et al.* (2020) reported in the present research that ingesting two per cent stevia-sweetened tea (once or twice daily for two months) significantly altered the fasting blood glucose and HbA1c levels in diabetes patients with no statistically significant differences with compare to the control group (sucralose group). Red blood cells (RBCs) have a life span of around 120 days. Therefore, a 50 per cent turnover in two months can be evaluated; routine evaluation is frequently done every three months.

MATERIALS AND METHODS

Search for knowledge is usually called research. The study designs and methodology explains the sense behind the methods used by the researcher in the research study. It explains the relevance of a particular technique and method adapted for the research purpose of drawing a general conclusion.

This chapter elicits the procedure adapted for conducting the present research study. Dairy foods play a central role in most dietary recommendations. They provide nutrients and bioactive compounds essential for health that are difficult to obtain in diets with no or limited dairy products. *Lassi* and yogurt are popular dairy products that were introduced hundred years ago. Fermented food products, especially those incorporated with probiotics consumption, have been related to several health advantages, including improved immune and digestive systems. *Lassi* and yogurt consumption and marketing have grown in popularity worldwide as people become more conscious of their health benefits. So the study has been conducted and the details of the materials used, experimental methods followed and techniques adopted during the course of the investigation are under the following sub-headings.

3.1 Experimental Site:

The study was carried out at the College of Community and Applied Sciences, College of Dairy and Food Technology, MPUAT, Udaipur and Hanumangarh Jn. (District Hanumangarh) (Rajasthan). The experiment was conducted in three phases, as presented under following heads (Figure 3.1).

3.2 PHASE I: DEVELOPMENT AND PHYSICO-CHEMICAL ASSESSMENT OF PROBIOTIC PRODUCTS

3.2.1 Procurement of cultures

3.2.2 Propagation of cultures

3.2.3 Procurement of raw material

3.2.4 Preparation of probiotic products

3.2.5 Sensory profile evaluation of probiotic products (Fresh)

3.2.6 Gross composition and Physico-chemical evaluation of probiotic products (Fresh)

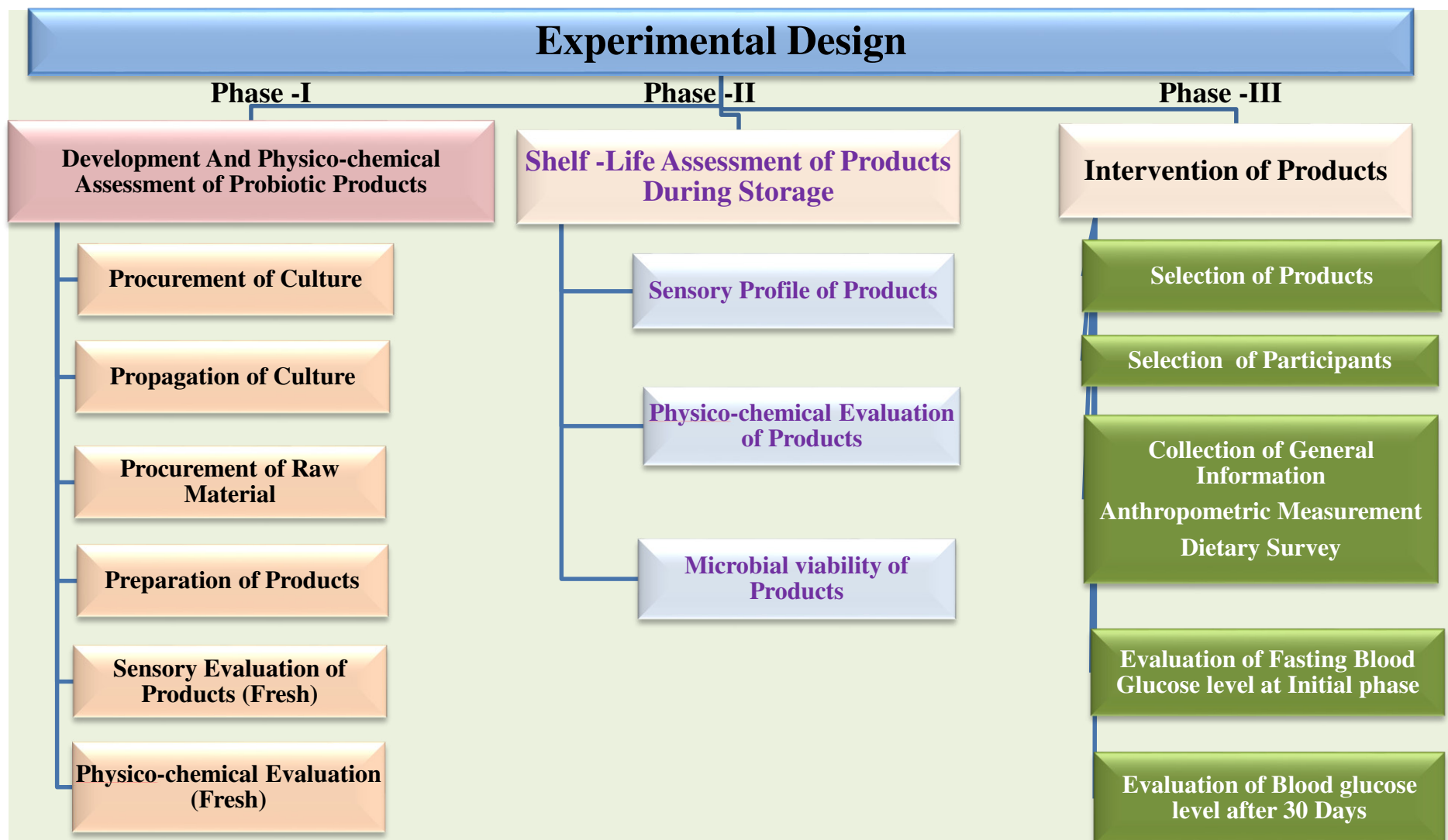


Figure 3.1 Experimental Design

3.3 PHASE II: SHELF LIFE ASSESSMENT OF PROBIOTIC PRODUCTS

3.3.1 Physico-chemical Evaluation of developed probiotic products during storage

3.3.2 Sensory profile of probiotic products during storage

3.3.3 Microbial viability evaluation of probiotic products during storage

3.4 PHASE III: INTERVENTION OF PRODUCT

3.4.1 Selection of Product

3.4.2 Development of questionnaire

3.4.3 Selection of diabetic patients

3.4.4 General Data Collection

3.4.5 Anthropometric measurement

3.4.5 Dietary Survey

3.4.6 Estimation of Fasting Blood Glucose Levels

3.5 ETHICAL APPROVAL

3.6 STATISTICAL ANALYSIS

3.2 PHASE I: DEVELOPMENT AND PHYSICO-CHEMICAL ASSESSMENT OF PROBIOTIC PRODUCTS

3.2.1 Procurement of Cultures

The *Lactobacillus helveticus* MTCC 5463 pure strain (probiotic culture) (earlier known as *Lactobacillus acidophilus* V3) and starter culture *Streptococcus thermophilus* MD2 was adopted for the proposed experiment. The Dairy Microbiology Department of the SMC College of Dairy Science, Anand Agricultural University, Anand, provided the culture.

3.2.2 Propagation of cultures:

It was propagated by incubation at 37°C for 8–12 hours and storage at 4–2°C in sterilised reconstituted skim milk (11 per cent T.S.). Before use, three repeated transfers of culture were provided to maintain activity during the research.

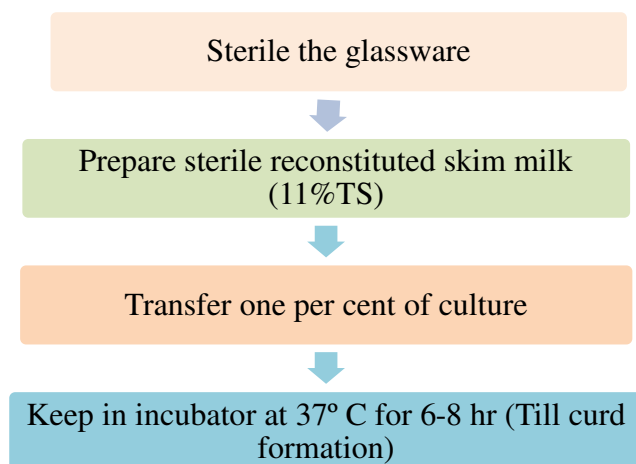


Figure 3.2 Flow Chart of propagation of Culture

3.2.3 Procurement of raw material:

3.2.3.1 Goat Milk: Goat milk was procured from the Bijanwas and Amberi Villages nearby Udaipur city.

3.2.3.2 Stevia: Stevia extract and Powder procured from the local market of Udaipur city.

3.2.3.2 Flavours: Different flavours were purchased from the local market of Udaipur City.

3.2.4 Preparation of products:

3.2.4.1 Composition of Products:

Sample No.	Table 3.1 Composition of <i>Lassi</i>			
	Curd (%)	Flavour	Water (%)	Stevia
LS ₀ (Control)	90	Without flavour	10	Without Stevia
LS ₁	90	Pineapple	10	Stevia Extract (90µl)
LS ₂	90	Kesar Elachi	10	Stevia Extract (90µl)
LS ₃	90	Rose	10	Stevia Extract (90µl)
LS ₄	90	Pineapple	10	Stevia Powder (0.35g)
LS ₅	90	Kesar Elachi	10	Stevia Powder (0.35g)
LS ₆	90	Rose	10	Stevia Powder (0.35g)

Sample No.	Table 3.2 Composition of yogurt		
	Curd (%)	Flavour	Stevia
YS ₀ (Control)	100	Without flavour	Without Stevia
YS ₁	100	Kevada	Stevia Extract (90µl)
YS ₂	100	Kesar Pista	Stevia Extract (90µl)
YS ₃	100	Raspberry	Stevia Extract (90µl)
YS ₄	100	Kevada	Stevia Powder (0.40g)
YS ₅	100	Kesar Pista	Stevia Powder (0.40g)
YS ₆	100	Raspberry	Stevia Powder (0.40g)

3.2.4.2 Method for Preparation of probiotic *Lassi* and probiotic yogurt:

Goat milk was used to prepare the curd with starter culture and probiotic culture. Fresh milk was preheated to 90°C for 5 min, filtered through muslin cloth, and then cooled to the temperature (37°C) of inoculation. Then mix both culture (1% of each culture (*Streptococcus thermophilus* MD2 and *Lactobacillus helveticus* MTCC 5463)) and allowed to settle the curd than cool it and mix the desired flavour and sweetening agent. Stevia extract and stevia powder were used for sweetening the probiotic *Lassi* and yogurt. Ten per cent water was used to maintain the consistency of *Lassi*. Control was prepared without stevia (Extract and Powder) and flavours and experimental samples were prepared with different flavours. The flow diagram for the preparation of probiotic *Lassi* and probiotic yogurt is shown in Figure 3.3 and 3.4 (Plate-1).

3.2.4.2.1 Method for Curd Formation:

First, all utensils, equipment and containers were cleaned and sanitized before use. Filtration of milk was done for the removal of debris and homogenization of milk has been done. Then processed milk was pasteurized at 90-95°C for 5-10 min. After the heat treatment, the milk was cooled to 43°C, inoculated with the starter and probiotic culture, and incubated at 37°C for 6-7 hours.

3.2.4.2.2 Cooling:

When the desired acidity (0.80-0.90%) was achieved, the set curd was immediately transferred to the refrigerator to detain further activity of the culture.

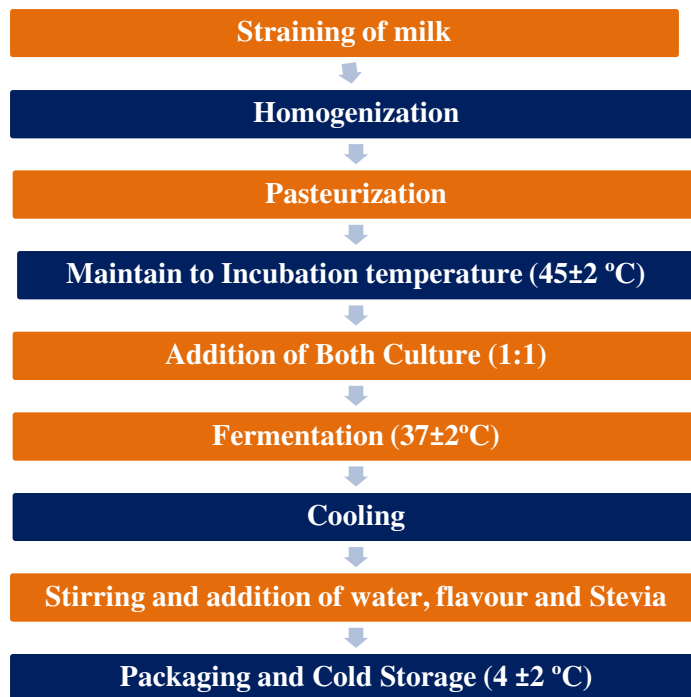


Figure 3.3 Flow chart for preparation of *Lassi*

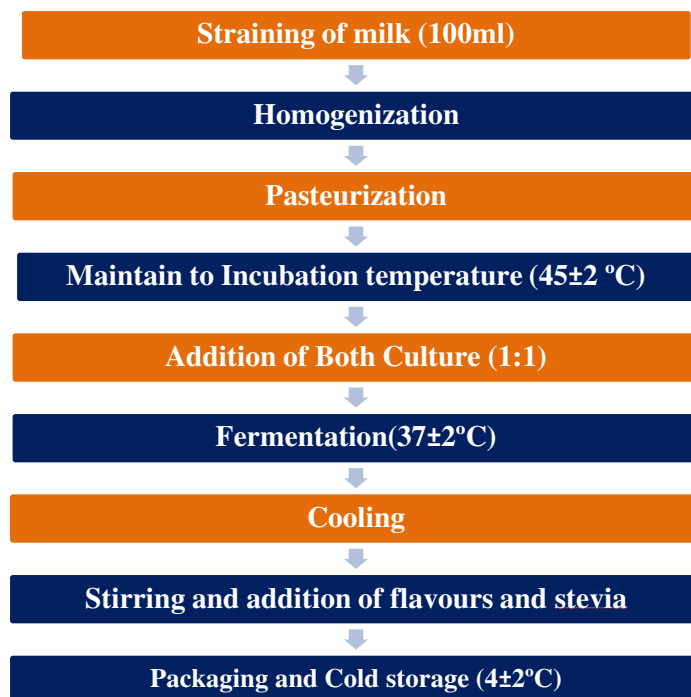


Figure 3.4 Flow chart of preparation of yogurt modified from Weerathilake (2014), Lee and Lucey. (2010).

3.2.4.2.3 Breaking of Curd and addition of flavour and sweetening agent:

After allowing sufficient cooling time (5-6 h), the curd was broken by a mechanical stirrer. Then control and experimental samples were prepared. Control samples were without flavour and stevia (Extract or Powder) and the experimental samples were prepared with different flavour and stevia (Extract or Powder). This was labelled as probiotic *Lassi* and probiotic yogurt.

3.2.4.2.4 Packaging:

The probiotic *Lassi* was then filled into glass bottles (200 ml) and probiotic yogurt was packed in the glass jars (150 ml) and stored at refrigeration temperature ($4\pm 2^{\circ}\text{C}$) for shelf-life studies.

3.2.5 Sensory profile evaluation of probiotic products (Fresh):

3.2.5.1 Score card:

A score card (Nine-point hedonic rating scale) was applied to evaluate the delectability and acceptability of food items based on certain significant aspects (Appendix-I). The following qualities were taken into consideration: colour and appearance, flavour, mouth feel, consistency and overall acceptability. Yeh *et al.* (1998) stated that the nine-point hedonic rating scale is one of the most admired ways to evaluate consumer acceptability. The sensory qualities of each developed product were rated on a nine-point hedonic scale in the current research. The aim was for the subject to select the score that best reflected their opinion of the product. The evaluations were given numerical values for computations, ranging from 9 (Liked extremely) to 1 (disliked extremely).

3.2.5.2 Selection of Panel Members:

To assess the acceptability of developed products produced during the experiment, a panel of fifteen judges comprised of staff and students from the College of Community and Applied Sciences, Department of Food Science and Nutrition at MPUAT, Udaipur, was chosen. The experts' collective experience, knowledge, willingness, and sincerity were also considered.

3.2.5.3 Evaluation of products:

The products, *viz.* probiotic *Lassi* and probiotic yogurt were evaluated for sensory acceptability. The best flavour of *Lassi* and yogurt was selected on the basis

of sensory scores of products prepared and findings used for further research (Plate-3).

3.2.6 Gross composition and Physico-chemical evaluation of probiotic products (Fresh):

The representative samples of probiotic *Lassi* and probiotic yogurt used in the present study were analysed for gross composition and physico-chemical parameters as described under section (Plate-2).

3.2.6.1 Determination of Total Solids (TS):

Total solids (TS) were determined according to the procedure described in ISO 6731:2010. In a dry and clean previously weighed petri dish, 5 ml sample was poured and the weight of sample was recorded. After that, the dish was transferred to the oven maintained at $100 \pm 2^\circ\text{C}$. After 3 h, the dish was immediately transferred to a desiccator, cooled and weighed. The above procedure of heating at $100 \pm 2^\circ\text{C}$ for 30 min, cooling and weighing was repeated, until difference weight between successive weighing did not exceed 0.5 mg.

$$\text{Total solids (\%)} = \frac{100 \times W1 \text{ (g)}}{W2 \text{ (g)}} \times 100$$

Where,

W1 = weight in g of residue after drying

W2 = weight in g of sample

3.2.6.2 Determination of Solid Non Fat (SNF):

Determination of solids-non-fat (SNF) was determined by taking the difference between total solids (%) and fat (%) content as described in section 3.2.6.1 and 3.2.6.5 (Igbabul *et al.* 2014).

$$\text{Solids-Non-Fat (\%)} = \text{total solids (\%)} - \text{fat content (\%)}$$

3.2.6.3 Determination of pH:

The primary working mechanism of the pH meter is based on the exchange of ions from the sample solution to the inner solution (pH seven buffer) of the glass electrode through the glass membrane that generates electric voltage. Therefore, the result of the pH meter is based on the combined relation between electric voltage, ion concentration, and pH reading.

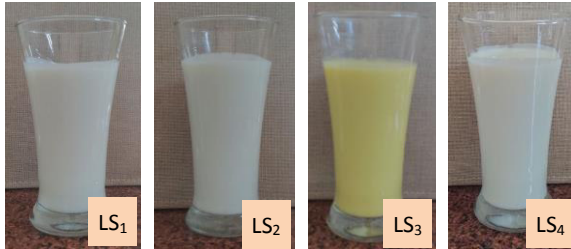


Plate 1 Preparation of product



Plate 2 Physico-chemical evaluation of product

The pH of probiotic *Lassi* and yogurt was determined using a Digital pH meter (Hana pH meter No. 211). The pH meter was standardised using pH 4.0 and pH 7.0 buffer solutions. The yogurt samples were stirred with a small amount of distilled water before pH measurement. pH was measured further for shelf life evaluation.

3.2.6.4 Titratable Acidity (TA):

Titrateable acidity of the samples (*Lassi* and Yogurt) was measured as per the ISO 6091:2010. Ten milliliters of thoroughly mixed sample was taken in a porcelain dish, and then 1 to 2 drops of phenolphthalein indicator was added to the sample. The contents of dish were titrated against 0.1 N NaOH till the appearance of light pink tinge, which persisted for 30 s in the solution. Titratable acidity was calculated using the following for:

$$\text{Acidity (\%)} = \frac{9 \times V \times N}{W} \times 100$$

Where,

V = Volume in ml of 0.1N NaOH required for titration

N = Normality of NaOH solution

W = Volume in ml of milk taken for the titration

3.2.6.5 Determination of Viscosity:

Viscosity is the measure of the internal friction of a fluid. This friction becomes apparent when a layer of fluid is made to move in relation to another layer. The greater the friction, the greater the force required to cause its movement, called "shear". Shearing occurs when the fluid is physically moved, pouring, mixing, etc. The measure of the speed at which these layers move with respect to each other is called the "shear rate." The viscosity of probiotic *Lassi* and yogurt samples was determined by using Brookfield viscometer.

Procedure:

The viscosity of probiotic *Lassi* and yogurt was assessed by using Brookfield viscometer.

- ❖ Turn on the viscometer and allow standing, must be auto zero, after the few seconds the screen appears which indicates 2 digits.
- ❖ Now press the key, the screen displays to remove spindle, after removing the spindle and pressing the key the instrument begins, it is auto zeroed.
- ❖ After approximately 15 see, the screen displays the instruction to replace spindle
- ❖ Attach the spindle to viscometer by screwing then on the lower shaft using left hand thread.
- ❖ Press the spindle key and up and down arrow keys. When the desired code is displayed release the arrow key.
- ❖ To select spindle, first press either up and down key which cause the area to show current speed, press the set speed key for adjusting the speed (60 RPM).
- ❖ Insert center of this spindle in the *Lassi* and Yogurt samples (100ml) until the fluid level is at the immersion groove on the spindle shaft. Tilt the spindle slightly while immersing to avoid air entrapment.
- ❖ Allow time for the individual reading to stabilize, record the value.

3.2.6.6 Determination of Fat Content:

The fat content of developed products was estimated using a milk butyrometer, which is opened at both ends. After closing the stem side opening with a good quality acid resistant silicon stopper, 10 ml of Gerber sulphuric acid, 10.75 ml of *Lassi* and yogurt sample and one ml of iso-amyl alcohol were added in succession into the butyrometer. After closing the neck side with a lock stopper, the contents were properly mixed and centrifuged at 1200 rpm for 5 min to get a clear fat column (ISO 488:2008).

3.2.6.7 Determination of Ash :

The ash was estimated by the official method described in A.O.A.C. (1984). When a substance is heated at 550°C for a long period in the presence of air, all organic as well as nitrogenous matter gets burnt. So that only inorganic matter in the form of simple compounds such as oxides is left behind.

Procedure

Known quantity (5 ml) of samples of probiotic *Lassi* and probiotic yogurt were taken in a previously weighed crucible. These crucibles were kept in the muffle furnace which was set at 550°C and were kept the crucible until a white or light grey ash was obtained. The crucible was then cooled at room temperature by keeping it in desiccators and instantly weighed for the ash content. Thus, the ash obtained was calculated using the following formula.

Calculation

$$\text{Ash (\%)} = \frac{(W_3 - W_1)g}{(W_2 - W_1)g} \times 100$$

Where,

- W₁ = Empty crucible weight (g),
- W₂ = Crucible + Sample weight (g) before ashing, and
- W₃ = Crucible + Ash weight (g) after complete ashing.

3.2.6.8 Determination of Protein content:

Total protein was determined as per Micro-Kjeldhal method described in (ISO 8968: 1).

Principle

Protein content is estimated by the Kjeldhal method, which is based on the determination of nitrogen present in the sample as ammonium sulphate by boiling with concentrated H₂SO₄ with a catalyst such as hydrogen peroxide. The ammonia released from ammonium sulphate is then estimated.

Procedure:

Known quantity (100 mg) of sample of *Lassi* and yogurt was placed into a Micro-Kjeldhal digestion flask. 10 ml of concentrated H₂SO₄ was added and heated for 10 minutes at 100°C. Afterwards 10 ml of hydrogen peroxide was added to it and again digested at 370°C until clear digest was obtained. After cooling, 2 ml of distilled water was added and the digested mixture was transferred to the distillation apparatus along with washings of digestion flask. Then, 40 ml of 40 per cent NaOH was added prior to steam distillation. 100 ml conical flask containing 20 ml of 4 per

cent boric acid solution and 4 drops of mixed indicator was placed under condenser so as to have condenser tip dipped in acid solution. The distillation continued until 50 ml distillate was collected. The distillate collected was titrated against 0.01 N H₂SO₄. A blank was also run without the sample. The nitrogen content of the sample was converted into protein by using a factor of 6.25.

Calculation

Volume of 0.01 N H₂SO₄ used for the test sample = x ml.

Volume of 0.01 N H₂SO₄ used for the blank = y ml.

Volume of 0.01 N H₂SO₄ used for the protein sample = x – y ml = z ml.

$$\text{Nitrogen (\%)} = Z \times \frac{\text{Normality of H}_2\text{SO}_4 (0.01)}{\text{Weight of the sample taken (mg)}} \times 14.007 \times \frac{100}{1000}$$

Where,

$$Z = \text{Volume of H}_2\text{SO}_4 \text{ used for protein sample (Sample titre - blank titre)}$$

Conversion of nitrogen into protein

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

3.2.6.9 Determination of Carbohydrate:

The Carbohydrate content of samples was calculated by difference method (NIN 2003). Carbohydrate content was determined by subtracting the sum of the values (per 100 g) of moisture, crude protein, fat and ash from 100. Formula for determining total carbohydrates is given below:

$$\text{Carbohydrate content (\%)} = 100 - (\text{moisture} + \text{protein} + \text{fat} + \text{ash})$$

3.2.6.10 Determination of Energy:

The energy value of sample was calculated by using physiological fuel value per gram of protein, fat and carbohydrate NIN (2003).

$$\text{Energy content (Kcal/100g)} = (\% \text{ protein} \times 4) + (\% \text{ carbohydrate} \times 4) + (\% \text{ fat} \times 9)$$

3.2.6.11 Determination of Mineral Profile:

Mineral solutions of selected products were prepared by wet ashing. The sample was digested with a mixture of acids to form a clear white precipitate which

was then dissolved in water and made up to a definite volume. An aliquot from this was used for determination of selected minerals.

Preparation of mineral solution:

Sample was digested by the wet digestion method. One gram sample was taken and dissolved in 5ml of concentrated nitric acid, added 5 ml of 70 per cent perchloric acid and digested till a colourless or a slightly greenish solution was obtained. The digested sample was transferred into a 100 ml volumetric flask and made to the volume with distilled water (Duhan *et al.* 2002). These samples were then loaded to Atomic Absorption Spectrophotometer and concentration of elements was estimated.

In the present investigation, mineral elements *i.e.* calcium, iron, magnesium, copper, zinc and potassium contents were analyzed using atomic absorption spectrophotometer (ECIL, model AAS 4141). Diluted sample was drawn up in the atomizer burner assembly through a capillary and converted by means of stream of compressed air to a fine spray which after condensation of large droplets was mixed with acetylene and burnt in a long flame at the burner light coming from the hollow cathode lamp, after transversing the flame entered a monochromatic wave set at 324 nm and fell on photomultiplier tube (photocell). The tube converted the light radiation into electrical energy which was measured by galvanometer. Calcium, iron, magnesium, copper, zinc and potassium was estimated at wavelengths 422.7 nm, 249.1 nm, 285.5nm, 324.9nm, 214.2nm and 234.5 nm, respectively.

Phosphorus: Phosphorus content of the samples were analyzed by using method given by USDA, (1954) ammonium molybdate reacts with inorganic phosphorus or phosphomolybdate which is yellow in colour. The intensity of the colour is estimated calorimetrically.

Procedure: One ml of test solution (mineral solution) was taken in a volumetric flask. 5ml of H₂SO₄ solution, 5ml of ammonium vanadate, and 5ml of ammonium molybdate solution was added to the volumetric flask was kept one by one. Make up the volume up to 50ml mark with the distilled water, keep the flask for 30 minutes and read the OD at 400nm. OD of the sample was compared with that of the standard graph obtained using potassium dihydrogen phosphate.

3.3 PHASE II: SHELF LIFE ASSESSMENT OF PROBIOTIC PRODUCTS:

The keeping quality of the food is related to changes the lipid moiety and moisture content in the food undergoes during the period of storage. Assessing the storage stability of the developed product is essential to find out the maximum duration of storage during which the food would be safe for consumption without any significant loss in its quality. Therefore, keeping qualities and antioxidant activity of the selected flour and products were assessed by estimating sensory quality, Physico-chemical properties and microbial viability.

The samples of probiotic *Lassi* and probiotic yogurt was packed in glass bottles and Jars stored at $4\pm 2^{\circ}\text{C}$ were evaluated for their sensory and physico-chemical characteristics as well for microbiological changes (Plate-4) on day of preparation to 11 days of storage. However the study was intended for one month of storage period but due to poor acceptability at 13th day of storage, evaluation was conducted for 11th day of storage.

3.3.1 Physico-chemical evaluation of developed probiotic products during storage:

Physico- chemical evaluation of developed probiotic products during storage as per the method described in section 3.2.

3.3.2 Sensory profile of probiotic products during storage:

The product was subjected to the sensory evaluation by an expert panel of fifteen judges for colour and appearance, flavour, mouthfeel, consistency and overall acceptability criteria. Fresh product at 0 days and the stored products (3rd, 5th, 7th, 9th and 11th days storage at $4\pm 2^{\circ}\text{C}$) were brought to 10°C before giving for judging. The score given by them on 9 point hedonic scale (Appendix-I) were taken to determine the acceptability level of product.

3.3.3 Microbiological Analysis of probiotic products during storage:

Methods followed for analysis of changes in microbial viability are discussed below (Plate-5).

3.3.3.1 Preparation of samples for microbial analysis:

Eleven ml of sample was pipetted out (aseptically) and pour it to 99 ml phosphate buffer dilution blank to acquire 1:10 dilution. Subsequently 1 ml of above dilution was used to prepare further dilutions in 9 ml phosphate buffer tubes. Suitable dilutions were made and poured in a set of sterile Petri dishes in triplicates. The time duration that elapsed between preparations and pouring of the appropriate dilutions in sterile petri plates did not exceed 15 min.

3.3.3.2 Lactobacilli Count:

The Lactobacilli count was carried out as per the procedure described in the IDF standards (117 A: 1989). Serial dilutions of the samples were prepared as described in section 3.3.3.1 and 1 ml from selected dilutions was poured into triplicate plates and mixed with tempered MRS medium. After setting of the agar, another layer of the same medium (5-7ml) was poured. The plates were then incubated at $37\pm 2^{\circ}\text{C}$ for 72 h. After incubation, plates were removed to count the colonies. The lactobacilli count was expressed as CFU/ml.

3.3.3.3 Streptococci Count:

Streptococci count of products was determined as per the method described for milk by the Bureau of Indian standards (IS: 1479, Part III, 1962). Serial dilutions of the samples were prepared as described in section 3.3.3.1 and 1 ml from selected dilutions was poured into triplicate plates and mixed with tempered M17 medium.

After setting the agar, another layer of the same medium (5-7ml) was poured. The plates were then incubated at $37\pm 2^{\circ}\text{C}$ for 48 h. After incubation, plates were removed to count the colonies. The Streptococci count was expressed as CFU/ml.

3.3.3.4 Yeast and Mould Count:

The procedure followed was according to Indian Standards, IS: 5403 (1969). Serial dilutions of the samples were prepared as described in section 3.3.3.1 and suitable dilution was selected for evaluation. The petri plates containing the diluted samples were poured with melted and cooled potato dextrose agar medium at around 45°C (adjusted to 3.5 pH using 10% sterile tartaric acid solution within 30 min of addition to the sample), mixed properly, and allowed to solidify.



Plate 3 Sensory evaluation of product

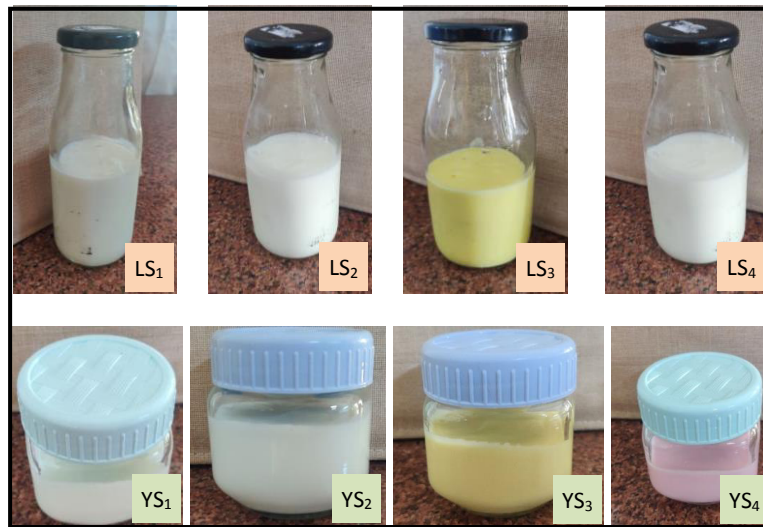


Plate 4 Storage of products

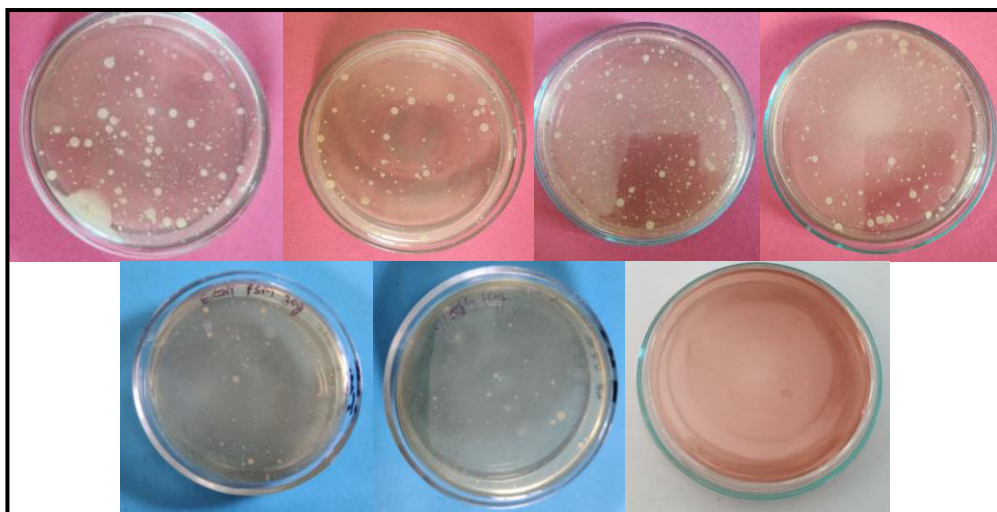


Plate 5 Evaluation of microbial viability

The plates were incubated at 22-25°C for 3-5 days before counts were recorded. A comparable procedure is also given for the enumeration of yeasts and moulds from food sources by Marshall (1992).

3.3.3.5 Coliform Count:

The Coliform count was estimated by the procedure laid down in ISI Hand Book (1981). Serial dilutions of the samples were prepared as described in section 3.3.3.1 and 1 ml from selected dilutions was poured into triplicate plates and mixed with tempered VRBA medium (Appendix-II). After setting the agar, another layer of the same medium (5-7ml) was poured. The plates were then incubated at 37±2°C for 24 h. After incubation, plates were removed to count the colonies. The coliform count was expressed as CFU/ml.

3.4 PHASE III: INTERVENTION OF PRODUCT:

3.4.1 Selection of Product:

Product was selected as per the sensory evaluation. Between both of product which scored highest was selected for the intervention. *Lassi* with Rose flavour received the highest score which was selected for administrated to experiment group.

3.4.2 Intervention of Product:

Selected product (Probiotic Lassi with rose flavour) was delivered (300ml) to experiment group. The product was provided fresh to the experiment group and ensure the consumption of product through phone call and daily feedback.

3.4.3 Development of questionnaire:

To collect the information in person, the pre-structures interview schedule was used. The interview schedule (Appendix-III) developed by the investigator consisted of four segment i.e. general information, anthropometric measurement, dietary survey and estimation of fasting blood glucose level. Further, booklet was developed and distributed to participants (Appendix-VII).

3.4.4 Selection of Diabetic Patients:

60 Diabetic patients Type II Diabetes mellitus age from 29-45 years (male) and divided in two groups (According to their consent to administration the product)

: control group and experimental group were selected for the experiment. Outdoor patient from Hisariya Hospital, Hanumangarh and Chawala Nursing Home, Hanumangarh were selected. Both the groups were prior informed about the details of study and patients were selected for the study with their consent (Appendix-VI) and administration of product done under the observation of Dr. Suresh Hisaria (Appendix-V).

3.4.5 Data Collection:

The data were collected through personal interview of the participants (Plate 8). The respondents were interviewed either at their residence or at their farm. At the time of interview, all possible efforts were made to develop a good rapport with the respondents.

3.4.5.1 General information of participants:

It included the details of family members with regards to age, education, family size, occupation, family type, marital status and sleep time.

3.4.6 Anthropometric measurement:

Anthropometry is used to assess and predict performance, health and survival of individuals and reflect the economic and social well-being of populations. Anthropometry is a widely used, inexpensive and non-invasive measure of the general nutritional status of an individual or a population group. The physical examination emphasized the measurement of height, weight and BMI (Plate-6).

This part of the interview schedule consisted of records of height (cm), weight (kg) and BMI of subjects, as the growth and physical status of body are profoundly influenced by diet and nutrition. Changes in body dimensions reflect the overall physical condition and wellbeing of individuals and populations.

3.4.6.1 Weight:

Weight is the simplest measurement of the growth and the nutritional status. It was measured in kilograms using a standard weighing machine.



Plate 6 Data collection from participants



Plate 7 Evaluation of Fasting Blood Glucose level



Plate 8 Distribution of Booklet

3.4.2.2 Height:

The height of an individual is influenced both by genetic and environmental factors. Height is affected only by long term nutritional deprivation; it is considered an index of chronic or long duration malnutrition. Height was also measured in centimeter using standard scale.

3.4.6.3 Body Mass Index:

Body mass index is defined as the individual's body weight divided by the square of their height in meter. The formulas universally use in medicine produce a unit of measure of kg/m².

3.4.7 Dietary Survey:

Diet is a vital determinant of health and nutritional status of people. The dietary habits of individuals/families/communities vary according to socio-economic factors, regional customs and traditions. Comparison of nutrient intake of individual with the RDA is carried out which provides a measure of adequacy or inadequacy of the nutrients. Quantitative methods of dietary enquiry were used to collect data regarding the food intake of participants via oral questionnaire (24 hrs. Recall Method).

The dietary intake data of the participants were collected for three consecutive days in a week (inclusion of 1 holiday, Sunday), using 24 hr. Recall Method. According to Dwyer (1994), a three-day dietary recall avoids the chances of over or under-estimation of the intakes and thus improves the accuracy of dietary assessment. A set of standardised utensils was used to ask about the total amount of cooked food prepared for the family and the amount of raw ingredients used in the food item. The nutrient content of the consumed food was calculated using values of "Nutritive value of Indian Food" (Gopalan *et al.* 2010) and with the help of software diet soft. The comparison of the dietary and nutrient intake of participants was done with the RDA 2020 (Reference Man) given by ICMR.

3.4.8 Estimation of Fasting Blood Glucose Levels:

Fasting Blood Glucose levels was estimated under the observation of Dr. Suresh Hisaria (Appendix-V and Plate 7).

3.4.8.1 Sample Collection:

5 ml of fasting blood was collected in clean, dry and sterilized vials (10 ml) and allow standing it at room temperature for 15 minutes. The supernatant (serum) was then separated and stored at 2-8°C and used for blood glucose level estimation.

3.4.8.2 Fasting blood glucose level assessment:

Fasting blood glucose level of control group and experiment group was estimated GOD/POD enzymatic method based on end point colorimetry Trindes (1969) and Tietz (1976) using enzymatic kit of live life Laboratories on fully autoanalyser SELECTRA E (Merck Company) at the Laboratory of Hisaria Hospital, Hanumangarh.

3.4.8.3 Specimen Preservation:

Serum was separated from the blood cells at the earliest after drawing of the blood, as the rate of glycolysis is approximately 7 mg per cent per hours at room temperature. Blood glucose was estimated within 36 hours of serum collection, as glucose in serum is stable for 72 hours at the temperature range of 2-8°C.

Principle: Glucose is oxidized by the enzyme GOD to give D- gluconic acid hydrogen peroxide. Hydrogen peroxide in presence of the enzyme POD oxidizes phenol which combine with 4- amino antipyrine to produced a red colour quinoneline dye. The intensity of colour produced is proportional to glucose concentration in the sample, which was measured at 505 nm.

Manual Assay:

- ❖ Blank solution was prepared by pipetting out 1000µl of glucose reagent into the clean, sterilized and dry vial.
- ❖ To prepare standardised solution, 1000µl of glucose reagent was pipette out into the vial into the vial and to that 10 µl of sample was added and mixed well. Similarly, all the sample solution were prepared.
- ❖ All the prepared solutions were incubated at 37°C for 10 minutes.
- ❖ Thereafter the absorbance of blank, standard and samples was measured at 505 nm within 60 minutes.

❖ Blood glucose was then calculated using the formula given underneath.

Calculation:

$$\text{Blood Glucose Level (mg/dl)} = \frac{\text{Absorbance of Sample (O.D.)}}{\text{Weight of the sample taken (mg)}} \times 100$$

3.5 ETHICAL APPROVAL:

The Human Research Ethical Committee of Maharana Pratap University of Agricultural and Technology approved the research work (Ethics approval number: MPUAT/DR/20/7428) (Appendix-IV).

3.6 STATISTICAL ANALYSIS:

The data obtained during the present investigation were analyzed by completely randomized design (CRD) (Steel and Torrie, 1980). The following statistical model was used for analysis,

Statistical model

$$Y_{ij} = \mu + T_j + E_{ij}$$

Where,

Y_{ij} = Response due to j^{th} number of treatments in i^{th} replication,

$i = 1, 2, \dots, r, (r = 5),$

$j = 1, 2, \dots, t, (t = 4),$

μ = General mean,

T_j = Effect due to j^{th} number of treatments, and

E_{ij} = Uncontrolled variation or random error due to j^{th} number of treatments in i^{th} replication.

Analysis of variance (ANOVA):

Sources of variation	Degree of freedom	Sum of square	Mean square	'F' calculated	'F' Table (At 5 %)
Treatments (Samples)	(t-1)	SST	MST	$\frac{MST}{MSE}$	-
Error	(n-t)	SSE	MSE	-	-
Total	(n-1)	SSTO	-	-	-

The standard error of mean (S.Em.), Critical Difference (C.D. at 5 per cent) and Co-efficient of Variation (C.V. %) were calculated as under.

$$\text{S.Em. for Treatments} = (\text{MSE}/r)^{1/2}$$

$$\text{C.D. at 5 \% for Treatments} = \text{S.Em.} \times \sqrt{2} \mu' \text{ table } t_{0.05} \text{ at error d.f.}$$

$$\text{C.V. \%} = \frac{(\text{MSE})^{1/2}}{\mu'} \times 100$$

Where,

- S.Em. = Standard error of mean,
- C.D. = Critical difference,
- R = Number of replications,
- μ' = General mean of sample data,
- MSE = Mean square error, and
- C.V. % = Co-efficient of variation.

RESULTS AND DISCUSSION

Fermented foods have immense importance since they supply a significant amount of nutrients with a broad range of aroma, flavour and texture, which improve the human diet. Goat milk has the potential to deliver probiotics and has some specific characteristics which bestow it from cow milk, like fat globules size, which is smaller than cow milk, provides a smoother texture in derived products, lesser amount of alpha-casein, consequential in softer gel products. Goat milk has a more typical flavour, which might be due to the liberation of short-chain fatty acids, which gives a goatee smell (Fontecha, *et al.* 2000 and Fontecha, *et al.* 2005).

With expanding health awareness, there has been a massive demand for sugar substitutes that provide fewer or no calories and possess better sweetening potency. Stevia, which plays a vital role as a non-nutritive natural sweetener, emerged as a safe sugar substitute that does not pose any threat to human health. So the combination of goat milk, probiotic culture and stevia to make the flavored probiotic products may have benefits for diabetes and obesity.

This chapter deals with the results and discussion of the present investigation and inferences drawn after subjecting the data to statistical analysis. The study was undertaken at the Department of Food Science and Nutrition. Based on different aspects carried out, inferences were drawn after subjecting the data to statistical analysis. Hence, required repetitions for each parameter of samples were measured to ascertain these aspects. The results have also been depicted graphically, wherever found necessary. The results obtained for the study are discussed in the chapter under the following heads:

4.1 Gross composition and physico-chemical evaluation of probiotic *Lassi* at the day of preparation

4.2 Gross composition and physico-chemical evaluation of probiotic yogurt at the day of preparation

4.3 Sensory profile of probiotic *Lassi* at the day of preparation

4.4 Sensory profile of probiotic yogurt at the day of preparation

- 4.5 Sensory profile of probiotic *Lassi* during storage**
- 4.6 Physio-chemical properties of probiotic *Lassi* during storage**
- 4.7 Microbial viability of probiotic *Lassi* during storage**
- 4.8 Sensory profile of probiotic yogurt during storage**
- 4.9 Physio-chemical properties of probiotic yogurt during storage**
- 4.10 Microbial viability of probiotic yogurt during storage**
- 4.11 General information of participants**
- 4.12 Anthropometric measurement of participants**
- 4.13 Diabetes mellitus and physical activities routine related information**
- 4.14 Dietary survey of participants**
- 4.15 Fasting Blood Glucose Level of participants**

4.1 GROSS COMPOSITION AND PHYSICO-CHEMICAL EVALUATION OF PROBIOTIC *LASSI* AT THE DAY OF PREPARATION:

4.1.1 Moisture content of probiotic *Lassi* on the day of preparation:

Moisture refers to the presence of water content which has vital effects on dairy products, influencing the quality of the product and its storage life. The moisture content of control and flavoured probiotic *Lassi* at the day of preparation is depicted in Table 4.1 and also presented in Fig. 4.1. The highest average value for moisture content was observed in LS₁ (88.10%) followed by LS₂ (87.90%), LS₃, (87.31%) and LS₀ (87.27%). The effect of adding flavours and stevia extract on the moisture content of goat milk-based flavoured probiotic *Lassi* samples and among control samples was non-significant ($p < 0.05$).

A similar study was conducted by Dhumal *et al.* (2018) and reported that *Lassi* prepared from buffalo milk and water addition of *pudina* extract, in the treatments T₁, T₂, T₃ and T₄ contained moisture content 88.45, 88.68, 88.75 and 88.84 per cent, respectively. Deshpande *et al.* (2020) also prepared the flavoured *Lassi* by blending buttermilk and soymilk at different proportions. Moisture content for four samples

was A, B, C and D was 90.50, 90.36, 90.20 and 90.15 per cent, respectively which was slightly higher than the present investigation.

4.1.2 Fat content of probiotic *Lassi* on the day of preparation:

Fats and oils are esters of fatty acids with glycerol, which account for 90 to 98 per cent of mass. The fat content of control and flavoured probiotic *Lassi* on the day of preparation is presented in Table 4.1 and Fig. 4.1. The highest average value for fat content was observed in LS₂ (3.59%), followed by LS₁ (3.58%), LS₃ (3.57%) and LS₀ (3.50%), respectively. It was slightly more than control. However, The effect of adding flavours and stevia extract on the fat content of goat milk-based flavoured probiotic *Lassi* samples and among control samples was non-significant ($p < 0.05$). Milk's fat content and composition differ naturally due to several factors, including cow feed composition, the amount of time spent grazing, and intrinsic differences between livestock (Heck 2009).

Similar results were found by Ghule *et al.* (2016) analysed the fat content of six brands of market *Lassi* sold in Kolhapur city and found 4.24, 3.00, 2.10, 3.62, 2.46 and 4.03 per cent, respectively. Dhumal *et al.* (2018) investigated average fat content in *pudina* extract *Lassi* was 2.60, 2.53, 2.43 and 2.40 per cent for treatment T₁, T₂, T₃, and T₄, respectively. Kaur *et al.* (2019) reported that the fat content of carrot pulp added to *Lassi* ranged from 2.37- 2.48 per cent, wherein the higher fat content was found in the control treatment. A similar result was reported by Kahandal *et al.* (2020) was prepared *Lassi* with different per cent of strawberry pulp. The mean value of fat content of *Lassi* in five different treatments was 3.55, 3.43, 3.32, 3.23 and 3.13 per cent, respectively. Kumar *et al.* (2020) also reported that the fat content of T₁, T₂, T₃, and T₄ was 3.46, 3.40, 3.46 and 3.31 per cent of herbal honey *Lassi* in different proportions. Deshpande *et al.* (2020) also prepared the flavoured *Lassi* by blending buttermilk and soymilk at different proportions. Fat content for four samples, A, B, C and D, was 0.25, 0.54, 0.71 and 0.88 per cent, respectively.

4.1.3 Carbohydrate content of probiotic *Lassi* on the day of preparation:

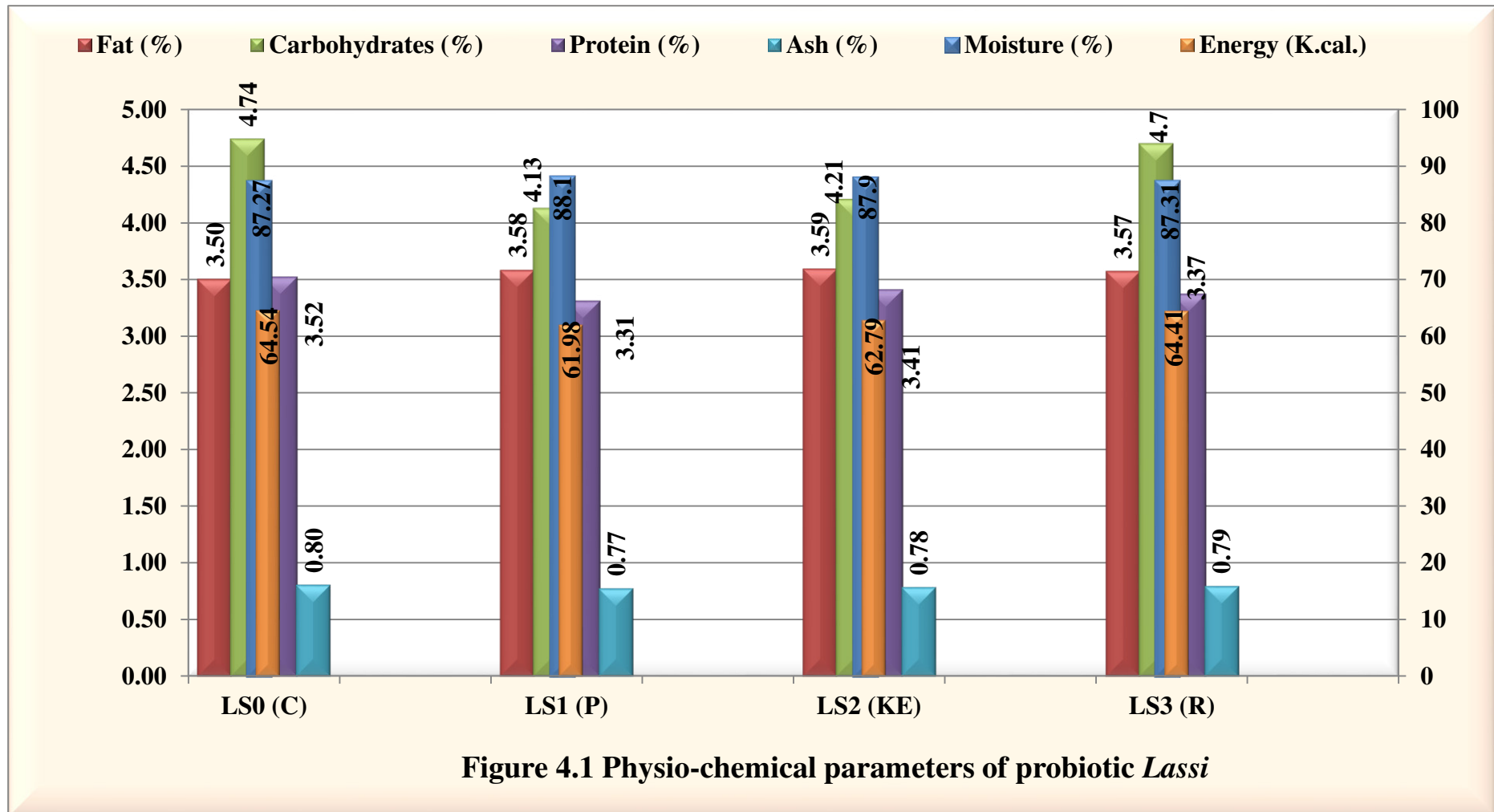
Lactose is dissolved in the serum (whey) phase of fluid milk. Lactose dissolved in the solution is found in two forms, called the α -anomer and β -anomer, which can convert back and forth between each other. The carbohydrate content of

flavoured probiotic *Lassi* along with control on the day of preparation has shown in Table 4.1 and also presented in Fig. 4.1. The highest average value for carbohydrates content was observed in LS₀ (4.74%), followed by LS₃ (4.70%), LS₂ (4.21%) and LS₁ (4.13%), respectively. It was slightly less than control. However, The effect of the addition of flavours and stevia extract on the carbohydrate content of goat milk-based flavoured probiotic *Lassi* samples and between control was non-significant (p<0.05).

Dhumal *et al.* (2018) reported the total sugar content of *pudina Lassi* was 5.28, 5.18, 5.16 and 5.16 per cent for treatment T₁, T₂, T₃ and T₄, respectively. Ghule *et al.* (2015) reported that the total sugar content of strawberry fortified *Lassi* was 13.92 to 16.29 per cent, respectively. Deshpande *et al.* (2020) prepare the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. Carbohydrates content for samples A, B, C and D was 5.32, 4.94, 4.82 and 4.64 per cent, respectively.

Table 4.1 Physio-chemical parameters of fresh probiotic *Lassi*

Sample No.	Moisture (%)	Fat (%)	Carbohydrate (%)	Protein (%)	Ash (%)	Energy (Kcal)
LS ₀ (C)	87.27±2.10	3.50±0.07	4.74±0.14	3.52±0.50	0.80±0.05	64.54±3.47
LS ₁ (P+SE)	88.10±1.47	3.58±0.14	4.13±0.22	3.31±0.21	0.77±0.06	61.98±4.03
LS ₂ (KE+SE)	87.90±1.91	3.59±0.20	4.21±0.31	3.41±0.08	0.78±0.06	62.79±3.21
LS ₃ (R+SE)	87.31±1.75	3.57±0.06	4.70±0.29	3.37±0.07	0.79±0.07	64.41±4.60
Mean	87.64	3.56	4.44	3.40	0.78	63.47
S.Em.±	0.82	0.06	0.25	0.13	0.02	1.73
C.V. (%)	2.09	3.68	12.62	8.23	4.28	6.14
<p>Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.D.), Each value is the average of five replications.</p>						



4.1.4 Protein content of probiotic *Lassi* on the day of preparation:

Proteins are the essential agents of biological functions; the diversity of the thousands of proteins found in nature arises from the commonly occurring 20 amino acids. The protein content of control and flavoured probiotic *Lassi* on the day of preparation is shown in Table 4.1 and Fig. 4.1. The highest average value for protein content was observed in LS₀ (3.52%), followed by LS₂ (3.41%), LS₃ (3.37%) and LS₁ (3.31%), respectively. It was slightly less than control. However, the effect of adding flavours and stevia extract on the protein content of goat milk-based flavoured probiotic *Lassi* samples and among control samples was non-significant ($p < 0.05$).

Similar studies conducted by Ghule *et al.* (2015) studied the protein content varies from 3.64 to 3.78 of strawberry fortified *Lassi*. Kumar *et al.* (2020) reported that the protein content of T₁, T₂, T₃ and T₄ was 3.35, 3.30, 3.25 and 3.20 per cent for herbal honey *Lassi*. Kahandal *et al.* (2020) also analysed five samples of *Lassi* and reported the protein 3.49, 3.29, 3.14, 2.94 and 2.71 per cent, respectively. *Lassi* prepared from cow milk blended strawberry pulp. Deshpande *et al.* (2020) prepare the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. protein content for samples A, B, C and D was 3.43, 3.46, 3.50 and 3.53 per cent, respectively. These results are coincide with the present study.

4.1.5 Ash content of probiotic *Lassi* on the day of preparation

Ash refers to the inorganic residue remaining after either ignition or complete oxidation of organic matter in a foodstuff. Ash content of control and flavoured probiotic *Lassi* on the day of preparation is depicted in Table 4.1 and also presented in Fig. 4.1. The highest average value for ash content was observed in LS₀ (0.80%) followed by LS₃ (0.79%), LS₂ (0.78%) and LS₁ (0.77%) was respectively. It was slightly less than control. However, The effect of adding flavours and stevia extract on the ash content of goat milk-based flavoured probiotic *Lassi* samples and among control samples was non-significant ($p < 0.05$).

Ghule *et al.* (2015) found the similar results from the ash from 0.73 to 0.80 per cent of strawberry fortified *Lassi*. Kumar *et al.* (2020) reported that ash content of T₁, T₂, T₃ and T₄ was 0.77, 0.78, 0.80 and 0.81 per cent for herbal honey *Lassi*. Similar results have been observed by Kahandal *et al.* (2020). They analysed the ash content

and found 0.72, 0.82, 0.91, 1.00 and 1.09 per cent *Lassi* prepared from cow milk blended strawberry pulp.

Deshpande *et al.* (2020) prepare the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. Ash content for samples A, B, C and D was 0.50, 0.70, 0.77 and 0.80 per cent, respectively.

4.1.6 Energy value of probiotic *Lassi* on the day of preparation:

The energy value of control and flavoured probiotic *Lassi* on the day of preparation is depicted in Table 4.1 and Fig. 4.1. The highest average value for energy value was observed in LS₀ (64.54 Kcal) followed by and LS₃ (64.41 Kcal), LS₂ (62.79 Kcal) and LS₁ (61.98 Kcal), was respectively. However, The effect of adding flavours and stevia extract on the energy value of goat milk-based flavoured probiotic *Lassi* samples and control samples was non-significantly lower than control ($p < 0.05$).

Whereas, Deshpande *et al.* (2020) prepared the flavoured *Lassi* with the blending of buttermilk and soymilk at different concentrations, and found the energy value of samples A, B, C and D were 37.25, 38.44, 39.47 and 40.64 Kcal, respectively. Mistry *et al.* (2018) also reported Moringa *Lassi* containing Moringa pod powder as a value-added component and reported energy value of control and *Lassi* prepared with Moringa 77.30 and 77.73 Kcal, respectively.

4.1.7 Total solids content of probiotic *Lassi* on the day of preparation

The total solids content of probiotic *Lassi* on the day of preparation is presented in Table 4.2 and Fig. 4.2. The highest average value for total solids content was observed in LS₀ (12.70%) followed by LS₃ (12.52%), LS₂ (12.05%) and LS₁ (11.79%), respectively. However, the effect of adding flavours and stevia extract on the total solids content of goat milk-based flavoured probiotic *Lassi* samples and among control samples was non significantly lower than control ($p < 0.05$).

Kahandal *et al.* (2020) also reported that total solids were 12.62, 11.47, 11.36, 11.23 and 11.10 per cent of *Lassi* prepared from cow milk blended strawberry pulp. Dhupal *et al.* (2018) Studied at *Lassi* was prepared using buffalo milk and an equal quantity of water with optimized *pudina* extract total solids 16.09, 18.19, 19.75 and 21.78 per cent, respectively for various treatments. Kumar *et al.* (2020) reported that

the total solids content of T₁, T₂, T₃ and T₄ was 17.19, 18.64, 19.52 and 20.48 per cent for herbal honey *Lassi*. Mistry *et al.* (2018) also studied Moringa *Lassi* contained Moringa pod powder (MPP) as a value-added ingredient. Moringa *Lassi* was prepared from *Dahi* made from standardized milk (Total solids (g/100g) control contains 17.27 per cent and moringa *Lassi* contains 15.48 per cent total solids, respectively. Deshpande *et al.* (2020) also prepare the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. Total Solids content for samples A, B, C and D were 8.50, 9.64, 9.80 and 9.85 per cent, respectively.

4.1.8 Solid Non Fat content of probiotic *Lassi* on the day of preparation:

Solids Non-Fat content of flavoured Probiotic *Lassi* on the day of preparation is presented in Table 4.2 and Fig. 4.2. The highest average value for Solids Non-Fat (SNF) content was observed in LS₀ (9.19%) followed by LS₃ (8.93%), LS₁ (8.37%) and LS₂ (8.22%), respectively. However, The effect of the addition of flavours and stevia extract on the Solids Non-Fat content among goat milk-based flavoured probiotic *Lassi* samples and the control sample was non-significant ($p < 0.05$), although the level was found below than control.

4.1.9 pH levels of probiotic *Lassi* on the day of preparation:

pH refers to the quantitative estimation of the acidity or basicity of aqueous or other liquid solutions. The pH of probiotic *Lassi* on the day of preparation is depicted in Table 4.2 and Fig. 4.2. The highest average value for pH level was observed in LS₀ (4.40) followed by LS₁ and LS₃ (4.39) and LS₂ (4.38), respectively. The effect of the addition of flavour on the pH was non-significant ($p < 0.05$) among all flavoured probiotic *Lassi* samples and the control sample on the day of preparation.

Kahandal *et al.* (2020) analysed the pH reported pH levels was 4.31, 4.42, 4.53, 4.61 and 4.71 of *Lassi* prepared from cow milk blended strawberry pulp. Dhupal *et al.* (2018) also analysed and pH level was found to be 4.19, 4.24, 4.25 and 4.27 per cent, respectively, in *Lassi*, which was prepared by using buffalo milk and a similar quantity of water with the addition of different levels of *puddina* extract. Deshpande *et al.* (2020) prepare the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. The pH of flavoured *Lassi* was for four treatments 3.99, 4.17, 4.27 and 4.35, respectively. Ghule *et al.* (2015) pH from 4.13-4.31 in strawberry fortified *Lassi*.

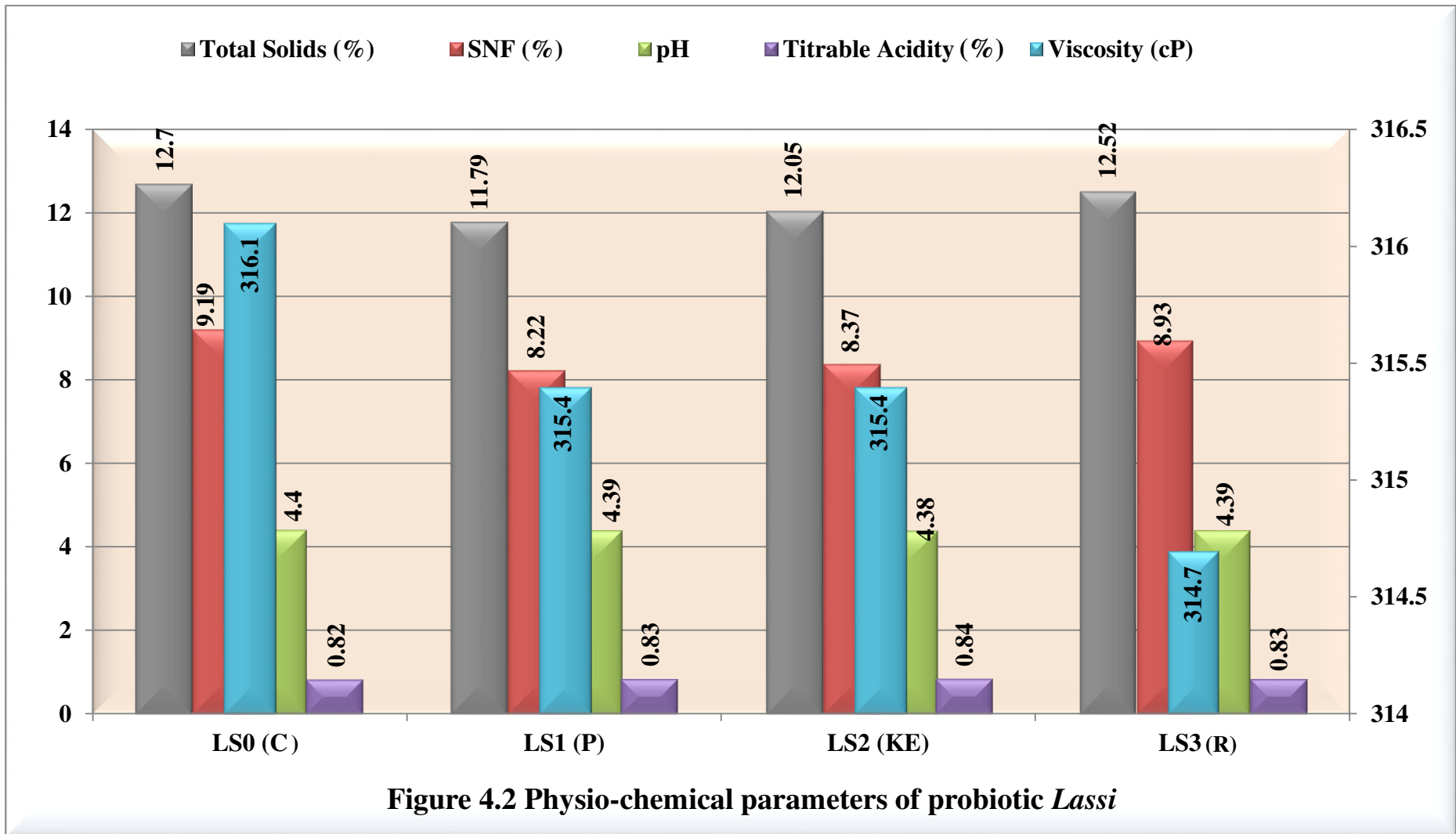
4.1.10 Titratable acidity levels of probiotic *Lassi* on the day of preparation:

Titratable acidity (TA) refers to the absolute concentration of free protons and undissociated acids in a solution that can react with a strong base and can be neutralised. The natural acidity in milk is due to its constituents such as casein, albumin, citrates, phosphates and carbon dioxide. On the other hand, developed acidity is due to the formation of lactic acid from lactose by microbial activity. Practically, natural and developed acidity cannot be distinguished.

The titratable acidity level of the probiotic *Lassi* and the control on the day of preparation is presented in Table 4.2 and Fig. 4.2. The highest average value for titratable acidity level was observed in LS₂ (0.84%), followed by LS₁ and LS₃ (0.83%) and LS₀ (0.82%), respectively. There was no wide variation between the titratable acidity levels in all *Lassi* samples. The effect of the addition of flavour on the titratable acidity level was non-significant ($p < 0.05$) among all flavoured probiotic *Lassi* samples and the control sample on the day of preparation.

Table 4.2 Physio-chemical parameters of fresh probiotic *Lassi*

Sample No.	Total Solids (%)	SNF (%)	pH	Titratable Acidity (%)	Viscosity (cp)
LS ₀ (C)	12.70±0.94	9.19±0.80	4.40±0.03	0.82±0.02	316.10±1.92
LS ₁ (P+SE)	11.79±0.93	8.22±1.06	4.39±0.04	0.83±0.03	315.40±3.76
LS ₂ (KE+SE)	12.05±0.70	8.37±0.81	4.38±0.02	0.84±0.02	315.40±3.83
LS ₃ (R+SE)	12.52±0.92	8.93±0.73	4.39±0.03	0.83±0.03	314.70±3.93
Mean	12.31	8.67	4.39	0.82	315.40
S.Em.±	0.39	0.39	0.04	0.01	1.55
C.V. (%)	7.17	9.95	1.79	3.36	1.09
Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.					



Kumar *et al.* (2020) reported that the titratable acidity content of T₁, T₂, T₃ and T₄ was 0.80, 0.80, 0.79 and 0.77 per cent for herbal honey *Lassi*. Kahandal *et al.* (2020) analysed the titratable acidity content was 1.02, 0.92, 0.82, 0.71 and 0.62 per cent of *Lassi* prepared from cow milk blended strawberry pulp. Ghule *et al.* (2015) reported acidity from 0.90 to 1.02 per cent of strawberry fortified *Lassi*.

4.1.11 Viscosity content of probiotic *Lassi* on the day of preparation:

Viscosity is a measure of a fluid's resistance to flow. It describes the internal friction of a moving fluid. The viscosity of probiotic *Lassi* on the day of preparation is presented in Table 4.2 and Fig. 4.2. The highest mean value for viscosity was observed in LS₀ (316.10 cp) followed by LS₁ and LS₂ (315.40 cp) and LS₃ (314.70 cp), respectively. There was no wide variation in the viscosity in all *Lassi* samples. The effect of the addition of flavour on the viscosity was non-significant ($p < 0.05$) between all flavoured probiotic *Lassi* samples and the control sample on the day of preparation.

Ghule *et al.* (2016) analysed the six brands of *Lassi* sold in Kolhapur city. The highest viscosity was recorded in the *Lassi* from KS₄ brand (381.50 ± 12.99 cp), followed by KS₆ (247.00 ± 8.40 cp) and KS₁ brand (243.33 ± 1.06 cp); however, the difference between viscosity of KS₁ and KS₆ were at par with each other.

Ranjan *et al.* (2015) attempted to utilize the whey solids in the concentrated form and partially hydrolysing its lactose in preparation for *Lassi*. The viscosity of control *Lassi* was estimated to be 363.20 cp, while adding 10 per cent CLHW to milk before fermentation for preparation has reduced the viscosity to 329.90 cp.

4.1.12 Minerals content of *Lassi* at the day of preparation:

Minerals are essential for the human body to stay healthy, keeping bones, muscles, heart and brain working properly.

The calcium content of probiotic *Lassi* on the day of preparation is presented in Table 4.3. The highest average value for calcium content was observed in LS₀ (116.40 mg %), followed by LS₃ (115.40 mg %), LS₂ (114.80 mg %) and LS₁ (112.40 mg %), respectively.

The phosphorus content of probiotic *Lassi* on the day of preparation is presented in Table 4.3. The highest average value for phosphorus content was observed in LS₀ (123.91 mg %), followed by LS₃ (122.60 mg %), LS₁ (121.81 mg %) and LS₂ (121.12 mg %), respectively.

The sodium content of probiotic *Lassi* on the day of preparation is presented in Table 4.3. The highest average value for sodium content was observed in LS₀ (35.87 mg %), followed by LS₃ (35.16 mg %), LS₁ (35.11 mg %) and LS₂ (33.90 mg %), respectively.

Table 4.3 Mineral content of probiotic *Lassi*

Sample No.	Calcium (mg %)	Phosphorus (mg %)	Sodium (mg %)	Potassium (mg %)
LS ₀ (C)	116.40 ±1.34	123.91±1.48	35.87±0.61	162.21±1.89
LS ₁ (P+SE)	112.40 ±0.89	121.81±3.88	35.11±0.99	159.77±0.66
LS ₂ (KE+SE)	114.80 ±2.68	121.12±3.32	33.90±1.23	160.80±1.00
LS ₃ (R+SE)	115.40 ±2.70	122.60±3.43	35.16±0.71	162.09±1.11
Mean	114.75	122.36	35.01	161.21
S.Em.±	0.92	1.42	0.41	0.56
C.V. (%)	1.78	2.59	2.66	0.78
Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.				

The Potassium content of probiotic *Lassi* on the day of preparation is presented in Table 4.3. The highest average value for potassium content was observed in LS₀ (162.21 mg %), followed by LS₃ (162.09 mg %), LS₂ (160.80 mg %) and LS₁ (159.77 mg %), respectively.

The magnesium content of probiotic *Lassi* on the day of preparation is presented in Table 4.4. The highest average value for magnesium content was observed in LS₀ (13.98 mg %), followed by LS₃ (13.95 mg %), LS₂ (13.93 mg %) and LS₁ (13.92 mg %), respectively.

The copper content of probiotic *Lassi* on the day of preparation is presented in Table 4.4. The highest average value for copper content was observed in LS₀ (0.058 mg %), followed by LS₃ (0.057 mg %), LS₂ (0.056 mg %) and LS₁ (0.054 mg %), respectively.

The iron content of probiotic *Lassi* on the day of preparation is presented in Table 4.4. The highest average value for iron content was observed in LS₀ (0.054 mg %), followed by LS₃ (0.053 mg %), LS₂ (0.051 mg %) and LS₁ (0.049 mg %), respectively.

Table 4.4 Mineral content of probiotic *Lassi*

Sample No.	Magnesium (mg %)	Copper (mg %)	Iron (mg %)	Zinc (mg %)
LS ₀ (C)	13.98±0.22	0.058±0.007	0.054±0.00	0.53±0.01
LS ₁ (P+SE)	13.92±0.35	0.054±0.012	0.049±0.006	0.52±0.03
LS ₂ (KE+SE)	13.93±0.65	0.056±0.014	0.051±0.008	0.52±0.02
LS ₃ (R+SE)	13.95±0.21	0.057±0.004	0.053±0.005	0.52±0.02
Mean	13.94	0.056	0.052	0.52
S.Em.±	0.18	0.001	0.01	0.01
C.V. (%)	2.89	9.51	7.13	3.69
Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.				

The zinc content of flavoured probiotic *Lassi* on the day of preparation is presented in Table 4.4. The highest average value for zinc content was observed in LS₀ (0.53 mg %), followed by LS₁, LS₂ and LS₃ (0.52 mg %), respectively.

There was no wide variation among the mineral content in all *Lassi* samples. It was slightly less than control. The effect of the addition of flavour and stevia extract on the calcium, phosphorus, sodium, potassium, magnesium, copper, iron and zinc content was non-significant (p<0.05) among all flavoured probiotic *Lassi* samples and control samples on the day of preparation.

4.2 GROSS COMPOSITION AND PHYSICO-CHEMICAL COMPOSITION OF PROBIOTIC YOGURT AT THE DAY OF PREPARATION:

4.2.1 Moisture content of probiotic yogurt samples on the day of preparation:

Moisture is an essential component in food products, especially dairy products which affect the perishability and viscosity. The moisture content of flavoured probiotic yogurt on the day of preparation is presented in Table 4.5. The highest average value for moisture content was observed in YS₂ (86.95%), followed by YS₃ (86.68%), YS₁ (86.52%) and YS₀ (85.98%), respectively, which was slightly more than the control. The effect of adding flavours and stevia extract on the moisture content among goat milk-based flavoured probiotics yogurt samples and the control sample was non-significant ($p < 0.05$).

Joseph *et al.* (2011) analysed random selected nine commercial yogurts from the Nigerian market; the moisture content of the samples ranged from 78.20 to 87.10 per cent which was agreed with the results of present investigation. Similar study conducted by Mukhekar and Desale (2018) prepared yogurt from cow milk with the addition of aloe vera at 12, 14, 16 and 18 per cent. Moisture content in treatments T₀, T₁, T₂, T₃, T₄ and T₅ were 83.44, 84.15, 85.05, 85.73 and 86.09 per cent, respectively.

Lesotho *et al.* (2019) studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The moisture content of yogurt samples from manufacture no. one for A, B and C were reported to be 79.16, 79.74 and 80.07 per cent from manufacture no. two for A, B and C were 76.08, 76.98 and 76.44 per cent and from manufacture no three two for A, B and C 79.64, 79.65 and 79.63 per cent, respectively.

4.2.2 Fat content of probiotic yogurt samples on the day of preparation:

In lacticinia, fat is an important constituent, which provides specific flavour and mouth feel to the product. The fat content of flavoured probiotic yogurt on the day of preparation is presented in Table 4.5. The highest average value for fat content was observed in YS₂ (4.03%) followed by YS₃ (3.94%), YS₁ (3.92%) and YS₀ (3.70%), which was to some extent higher than control. The effect of the addition of flavours and stevia extract on the fat content of goat milk-based flavoured probiotic yogurt samples and in the control sample was non-significant ($p < 0.05$).

Mukhekar and Desale (2018) prepared yogurt from cow milk with the addition of Aloe vera at 12, 14, 16 and 18 per cent, respectively. Fat content in yogurt was found to be 3.10, 1.55, 1.15, 0.82 and 0.80 per cent for treatment T₀, T₁, T₂, T₃ and T₄, respectively.

Joseph *et al.* (2011) analysed randomly selected nine commercial yogurts samples from the Nigerian market and reported that the fat content was 4.00 per cent and the lowest average fat content was 1.88 per cent.

Lesotho *et al.* (2019) studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The fat content of yogurt samples from manufacture no. one for A, B and C were reported to be 1.61, 1.49 and 1.55 per cent from manufacture no. two for A, B and C were 2.24, 2.08 and 2.10 per cent and from manufacture no. three for A, B and C 3.41, 3.50 and 3.34 per cent, respectively.

4.2.3 Carbohydrate content of probiotic yogurt samples on the day of preparation:

In lacticinia, the carbohydrate content of probiotic yogurt on the day of preparation is presented in Table 4.5. The highest average value for carbohydrates content was observed in YS₀ (5.37%) followed by YS₁ (4.97%), YS₃ (4.79%) and YS₂ (4.55%), respectively. Which was at some level lower than control. The effect of the addition of flavours and stevia extract on the carbohydrate content among goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant ($p < 0.05$).

Lesotho *et al.* (2019) studied nine commercially available yoghurt samples purchased from Maseru, the Kingdom of Lesotho. The carbohydrate content of yogurt samples from manufacture no. one for A, B and C were reported to be 16.53, 16.08 and 15.86 per cent, from manufacture no. two for A, B and C were 19.20, 18.35 and 18.30 per cent and from manufacture no. three for A, B and C 13.66, 13.65 and 14.12 per cent, respectively.

4.2.4 Protein content of probiotic yogurt samples on the day of preparation:

The protein content of flavoured probiotic yogurt, along with control on the day of preparation, is presented in Table 4.5. The highest average value for protein

content was observed in YS₀ (3.98%), followed by YS₁ (3.76%), YS₃ (3.75%) and YS₂ (3.65%), respectively. Which was slightly less than the control. The effect of adding flavours and stevia extract on the protein content among goat milk-based flavoured between probiotic yogurt samples and control samples was non-significant ($p < 0.05$).

Similar results were reported by Mukhekar and Desale (2018) prepared yogurt from cow milk with the blending of aloe vera (12, 14, 16 and 18%). The mean values of the protein content of the treatment T₀, T₁, T₂, T₃, T₄ and T₅ were 3.15, 2.61, 2.37, 2.17 and 2.10 per cent, respectively.

Lesotho *et al.* (2019) studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The protein content of yogurt samples from manufacture no. one for A, B and C were reported to be 2.39, 2.33 and 2.13 per cent from manufacture no. two for A, B and C were 1.95, 2.07 and 2.70 per cent and from manufacture no three two for A, B and C 2.35, 2.27 and 1.95 per cent, respectively.

4.2.5 Ash content of probiotic yogurt samples on the day of preparation:

Ash content of flavoured probiotic yogurt along with control on the day of preparation is presented in Table 4.5. The highest average value for ash content was observed in YS₀ (0.84%) followed by YS₁, YS₃ (0.83%) and YS₂ (0.80%) which was slightly lower with compare to the control. The effect of the addition of flavours and stevia extract on the ash content among goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant ($p < 0.05$).

Joseph *et al.* (2011) analysed random chosen nine commercial yogurts from the Nigerian market and reported the mean ash content range was from 0.26-0.71 per cent.

Lesotho *et al.* (2019) studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The ash content of yogurt samples from manufacture no. one for A, B and C were reported to be 0.28, 0.31 and 0.31 per cent, respectively from manufacture no. two for A, B and C were 0.45, 0.46 and 0.45 per cent, respectively and from manufacture no three for A, B and C 0.94, 0.93 and 0.95 per cent, respectively.

Table 4.5 Physio-chemical parameters of fresh probiotic yogurt

Sample No.	Moisture (%)	Fat (%)	Carbohydrates (%)	Protein (%)	Ash (%)	Energy (Kcal)
YS ₀ (C)	85.98±2.13	3.70±0.21	5.37±0.49	3.98±0.20	0.84±0.03	70.70±4.11
YS ₁ (K+SE)	86.52± 1.39	3.92±0.09	4.97±0.32	3.76±0.06	0.83±0.06	70.20±4.44
YS ₂ (KP+SE)	86.95±1.41	4.03±0.12	4.55±0.10	3.65±0.03	0.80±0.07	69.07±3.65
YS ₃ (R+SE)	86.68±0.52	3.94±0.14	4.79±0.72	3.75±0.21	0.83±0.06	69.62±0.80
Mean	86.53	3.93	4.96	3.78	0.82	69.89
S.Em.±	0.66	0.07	0.21	0.15	0.02	1.59
C.V. (%)	1.71	3.82	9.49	8.69	5.97	5.10
Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.						

4.2.6 Energy value of probiotic yogurt samples on the day of preparation:

The energy value of flavoured probiotic yogurt, along with control on the day of preparation is presented in Table 4.5. The highest average value for energy value was observed in YS₀ (70.70 Kcal) followed by YS₁, (70.20 Kcal) YS₃ (69.62 Kcal) and YS₂ (69.07 Kcal), respectively; which was slightly lower with compare to the control. The effect of the addition of flavours and stevia extract on the energy value among goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant (p<0.05).

Lesotho *et al.* (2019) studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The energy of yogurt samples from manufacture no. one for A, B and C were reported to be 90.19, 87.09 and 85 Kcal per cent from manufacture no. two for A, B and C were 104.75, 100.36 and 102.89 Kcal and from manufacture no three for A, B and C 94.71, 95.23 and 94.39 Kcal, respectively.

4.2.7 Total solids content of probiotic yogurt samples on the day of preparation:

The total solids (TS) content of flavoured probiotic yogurt, along with control on the day of preparation, is presented in Table 4.6. The highest average value for total solids content was observed in YS₀ (13.97%), followed by YS₁, (13.47%), YS₃ (13.31%) and YS₂ (13.04%) which was slightly lower with compare to the control. Adding flavours and stevia extract to the total solids content of goat milk-based flavoured probiotic yogurt samples and among control samples was non-significant ($p < 0.05$).

The study conducted by Mukhekar and Desale (2018) who prepared yogurt from cow milk with the addition of aloe vera (12, 14, 16 and 18%). The total solids content of the product treatments was found to be 16.56, 15.85, 14.95, 14.27 and 13.92 per cent for five treatments as T₀, T₁, T₂, T₃, T₄ and T₅, respectively.

Lesotho *et al.* (2019) also studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The total solids content of yogurt samples from manufacture no. one for A, B and C were reported to be 20.84, 20.26 and 19.93 per cent from manufacture no. two for A, B and C were 23.92, 23.02 and 23.56 per cent and from manufacture no three for A, B and C 20.36, 20.35 and 20.37 per cent, respectively. The results from both studies shows higher mean values than the present research.

4.2.8 Solid Non-Fat (SNF) content of probiotic yogurt samples on the day of preparation:

Solid Non-Fat (SNF) content of flavoured probiotic yogurt along with control on the day of preparation is presented in Table 4.6. The highest average value for Solid Non-Fat content was observed in YS₀ (10.20%), followed by YS₁, (9.55%), YS₃ (9.35%) and YS₂ (8.99%) which was slightly lower with compare to the control. The effect of adding flavours and stevia extract on the Solid Non-Fat content of goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant ($p < 0.05$). Lesotho *et al.* (2019) studied nine commercially accessible yoghurt samples purchased from Maseru (the Kingdom of Lesotho). The total Solids Non-Fat content of yogurt samples from manufacture no. one for A, B and C were reported to be 19.24, 18.77 and 18.38 per cent, respectively from manufacture no. two for A, B and

C were 21.68, 20.95 and 21.46 per cent, respectively and from manufacture no. three for A, B and C 16.95, 16.85 and 17.03 per cent, respectively.

4.2.9 pH level of probiotic yogurt samples on the day of preparation:

The pH level of flavoured probiotic yogurt along with control on the day of preparation is presented in Table 4.6. The highest average value for pH level was observed in YS₁ (4.38), followed by YS₂, YS₃ (4.37) YS₁, (4.36), which was slightly higher with compare to the control. The effect of the addition of flavours and stevia extract on the pH level among goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant (p<0.05).

Table 4.6 Physio-chemical parameters of fresh probiotic yogurt

Sample No.	Total Solids (%)	SNF (%)	pH	Titrateable Acidity (%)	Viscosity (cp)
YS ₀ (C)	13.97±0.45	10.20±0.50	4.36±0.02	0.86±0.03	376.20±5.16
YS ₁ (K+SE)	13.47±0.20	9.55±0.64	4.38±0.03	0.85±0.04	375.50±1.67
YS ₂ (KP+SE)	13.04±1.01	8.99±1.13	4.37±0.02	0.85±0.03	375.40±4.66
YS ₃ (R+SE)	13.31±0.47	9.35±0.42	4.37±0.03	0.84±0.01	375.80±1.63
Mean	13.44	9.52	4.37	0.85	375.47
S.Em.±	0.61	0.33	0.02	0.01	7.00
C.V. (%)	10.17	7.65	1.26	3.63	4.19
Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.					

Similar results found by Mukhekar and Desale (2018) who prepared yogurt from cow milk with the addition of aloe vera at (12, 14, 16 and 18%). pH of the product was found to be 4.24, 4.08, 3.96, 3.93 and 3.86 for the treatment T₀, T₁, T₂, T₃, T₄ and T₅, respectively. Joseph *et al.* (2011) also examined random chosen nine commercial yogurts from the Nigerian market and reported the average pH ranged from 4.08-3.70.

Oktavia *et al.* (2016) studied and depicted the effect of using single culture (*L. bulgaricus*, *S. thermophilus*) and mixed culture (*L. bulgaricus* and *S. thermophilus*), (*S. thermophilus*, *L. bulgaricus* and *Bifidobacterium*) as much as 3 per cent to measure the pH was for P₁, P₂, P₃ and P₄ as 3.73, 3.69, 3.67 and 3.98, respectively.

4.2.10 Titratable acidity (%) of probiotic yogurt samples on the day of preparation:

The titratable acidity of flavoured probiotic yogurt samples along with the control on the day of preparation is presented in Table 4.6. The highest average value for titratable acidity level was observed in YS₀ (0.86%), followed by YS₁, YS₂ (0.85%) and YS₂ (0.84%), which was slightly lower with compare to the control. The effect of the addition of flavours and stevia extract on the titratable acidity level among goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant ($p < 0.05$).

4.2.11 Viscosity of probiotic yogurt samples on the day of preparation:

The viscosity of flavoured probiotic yogurt along with control on the day of preparation is presented in Table 4.6. The highest average value for viscosity was observed in YS₀ (376.20 cp), followed by YS₃ (375.80 cp), YS₁, (375.50 cp) and YS₂ (375.40 cp) which was slightly lower with compare to the control. The effect of the addition of flavours and stevia extract on the viscosity of goat milk-based flavoured probiotic yogurt samples and the control sample was non-significant ($p < 0.05$).

There are wide ranges of viscosity in the various researches. Oktavia *et al.* (2016) conducted research to determine the effect of using single culture (*L. bulgaricus*, *S. thermophilus*) and mixed cultures (*L. bulgaricus* and *S. thermophilus*), (*S. thermophilus*, *L. bulgaricus* and *Bifidobacterium*) highest 3 per cent for evaluation of viscosity was for four treatments 257.30, 260.30, 285.30 and 1220.30 cp respectively.

Lutfiye *et al.* (2017) also studied buffalo milk yogurt and buffalo milk yogurt mixed with cow's and ewe's milk combination yogurts. The index of viscosity for mixed yogurt samples was between 244.71 and 810.04, 5532.92 and 14846.80; 146.54 and 537.89, and 256.84 and 1040.75 cp, respectively.

In another study Pappa *et al.* (2018) analysed the yogurt prepared with beta-glucan for four samples was 2030, 2020, 2015 and 2015 cp, respectively

4.2.12 Minerals content of probiotic yogurt samples at the day of preparation:

Minerals are important for the human body to stay healthy. The body uses minerals for many jobs, including keeping bones, muscles, heart, and brain working properly.

The calcium content of probiotic yogurt on the day of preparation is presented in Table 4.7 and Fig. 4.3. The highest average value for calcium content was observed in YS₀ (130.89 mg %), followed by YS₁ (130.74 mg %), YS₃ (129.54 mg %) and YS₂ (129.37 mg %), respectively.

The phosphorus content of probiotic yogurt on the day of preparation is presented in Table 4.7 and Fig. 4.3. The highest average value for phosphorus content was observed in YS₀ (139.49 mg %), followed by YS₁ (139.47 mg %), YS₃ (139.84 mg %) and YS₂ (139.01 mg %), respectively.

The sodium content of probiotic yogurt on the day of preparation is presented in Table 4.7 and Fig. 4.3. The highest average value for sodium content was observed in YS₀ (40.28 mg %), followed by YS₃ (40.22 mg %), YS₁ (39.81 mg %) and YS₂ (38.75 mg %), respectively.

The potassium content of probiotic yogurt on the day of preparation is presented in Table 4.7 and Fig. 4.3. The highest average value for potassium content was observed in YS₀ (175.55 mg %), followed by YS₁ (175.46 mg %), YS₃ (175.04 mg %) and YS₂ (174.46 mg %), respectively.

The magnesium content of probiotic yogurt on the day of preparation is presented in Table 4.8 and Fig. 4.3. The highest average value for magnesium content was observed in YS₀ (15.11 mg %), followed by YS₁ (14.91 mg %), YS₃ (14.86 mg %) and YS₂ (14.82 mg %), respectively.

Table 4.7 Mineral content of probiotic yogurt

Sample No.	Calcium (mg %)	Phosphorus (mg %)	Sodium (mg %)	Potassium (mg %)
YS ₀ (C)	130.89±1.81	139.49±1.97	40.28±0.71	175.55±3.46
YS ₁ (K+SE)	130.74±2.03	139.47±1.05	39.81±1.16	175.46±2.44
YS ₂ (KP+SE)	129.37±2.41	139.01±1.77	38.75±2.15	174.46±0.79
YS ₃ (R+SE)	129.54±1.95	139.84±2.40	40.22±0.81	175.04±3.87
Mean	130.13	139.45	39.76	175.12
S.Em.±	0.92	0.83	0.60	1.30
C.V. (%)	1.59	1.34	3.39	1.65
Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.				

The copper content of probiotic yogurt on the day of preparation is presented in Table 4.8 and Fig. 4.3. The highest average value for copper content was observed in YS₀ (0.072 mg %), followed by YS₁ and YS₃ (0.071 mg %) and YS₂ (0.069 mg %), respectively.

The iron content of probiotic yogurt on the day of preparation is presented in Table 4.8 and Fig. 4.3. The highest average value for iron content was observed in YS₀ (0.081 mg %), followed by YS₁, YS₃ (0.079 mg %) and YS₁ (0.077 mg %), respectively.

The zinc content of probiotic yogurt on the day of preparation is presented in Table 4.8 and Fig. 4.3. The highest average value for zinc content was observed in YS₀ (0.54 mg %), followed by YS₁ (0.53 mg %), YS₃ (0.52 mg %) and YS₃ (0.51 mg %), respectively.

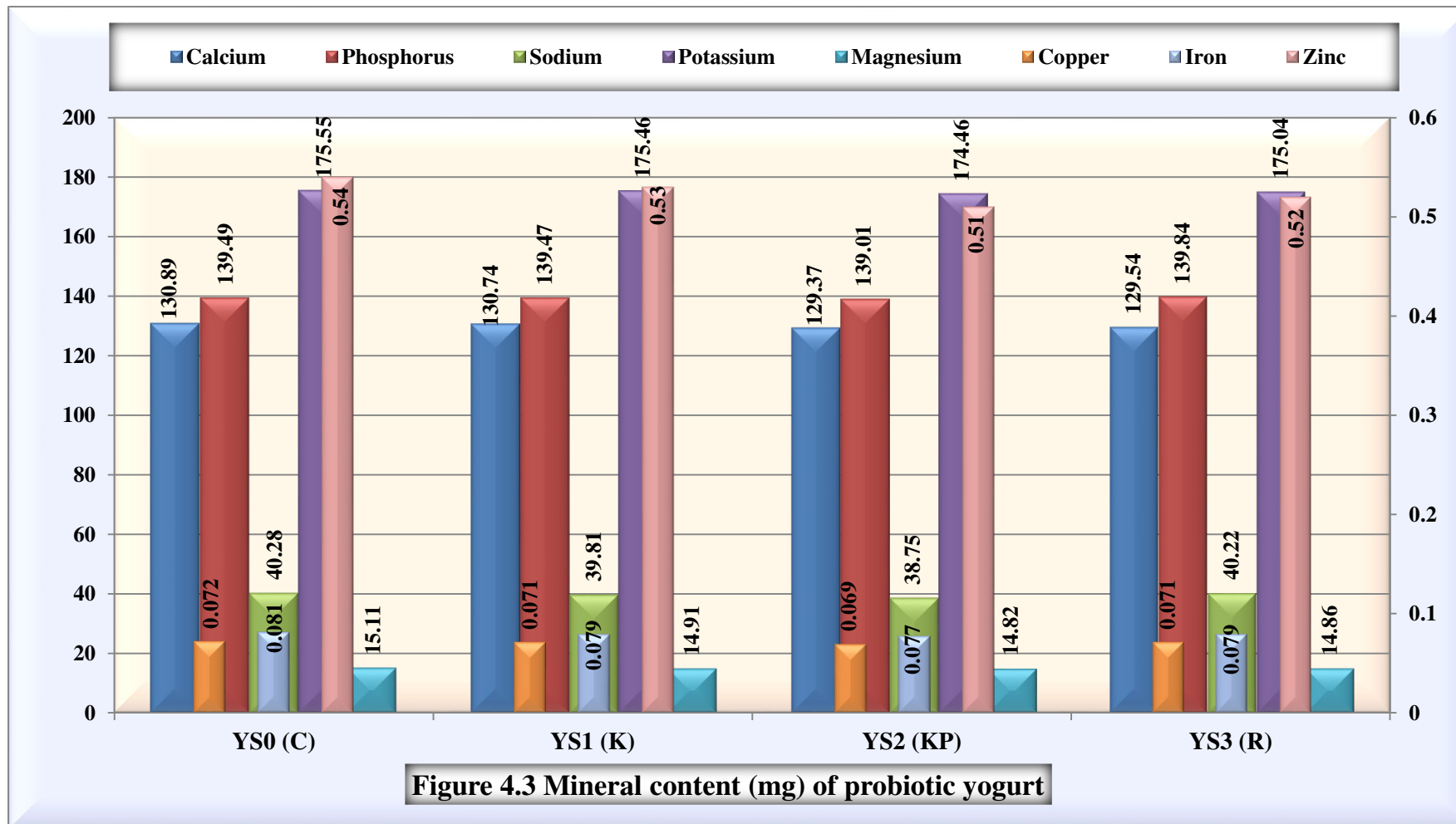


Table 4.8 Mineral content of probiotic yogurt

Sample No.	Magnesium (mg %)	Copper (mg %)	Iron (mg %)	Zinc (mg %)
YS ₀ (C)	15.11±1.22	0.072±0.01	0.081±0.02	0.54±0.01
YS ₁ (K+SE)	14.91±0.91	0.071±0.01	0.079±0.01	0.53±0.01
YS ₂ (KP+SE)	14.82±0.93	0.069±0.01	0.077±0.04	0.51±0.02
YS ₃ (R+SE)	14.86±0.97	0.071±0.02	0.079±0.01	0.52±0.01
Mean	14.92	0.071	0.079	0.53
S.Em.±	0.45	0.01	0.01	0.01
C.V. (%)	6.82	13.26	7.70	3.04
Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.				

There was no wide variation in the mineral content in all probiotic yogurt samples. It was slightly less than control. The effect of the addition of flavour and stevia extract on the calcium, phosphorus, sodium, potassium, magnesium, copper, iron and zinc content was non-significant ($p < 0.05$) between all flavoured probiotic yogurt samples along with control on the day of preparation. Deeth and Tamime, (1980) reported that yogurt contains 145 mg of calcium, 114 mg phosphorus, 47 mg sodium and 186 mg potassium (per 100 g).

4.3 SENSORY PROFILE OF PROBIOTIC LASSI AT THE DAY OF PREPARATION:

Sensory evaluation is a critical tool for quality control as well as research. Customers perceive product quality with their senses. Along with taste, other sensory attributes like flavour, appearance, texture and mouth feel are essential properties of lacticinia that cannot be sellable if the fondness and other visual characteristics are not satisfied the panel members as well as consumers (Kumthekar *et al.* 2021).

Sensory methods of analysis are evolving and necessary in reckoning the acceptability of food products. Hence, to get acceptable data, these tests are scientifically designed and tasters called ‘panellists’ carefully selected. Semi-trained but experienced panellists have judged particular sensory characteristics.

Results for the sensory profile of *Lassi* and yogurt, which were prepared with different flavours (*Lassi* -Pineapple, *Kesar Elaichi*, Rose and yogurt - Kevda, Kesar Pista, Raspberry) and incorporated with stevia extract and powder are as follows:

4.3.1 Colour and Appearance score of probiotic *Lassi* on the day of preparation:

Appearance is a crucial feature of sensory assessment, and the vision system is the sensor of appearance. The colour and appearance score of *Lassi* on the day of preparation is presented in Table 4.9 and Fig. 4.4. The highest average value for colour and appearance was observed in LS₂ (8.33) which was “liked very much”, followed by LS₅ (8.27), LS₃, LS₄ and LS₆ (8.07) lowest score was found in LS₀ and LS₁ (8.00). *Lassi* samples prepared with stevia extract and powder did not differ significantly ($p < 0.05$) along with control sample.

Similar results were reported by Poul *et al.* (2019) composed *Lassi* by using standardized milk where T₀ was control (cane sugar), T₁ (8% Honey), T₂ (75 µl Stevia) and T₃ (8% Honey + 75 µl Stevia) added. The colour score for T₀, T₁, T₂ and T₃ was 8.83, 8.81, 8.80 and 8.87, respectively. The non-significant divergence ($P < 0.05$) was observed in the mean values of the scores rationed to the colour and appearance of *Lassi* prepared under different treatments. Whereas Dhumal *et al.* (2018) reported that pudina *Lassi*'s colour and appearance score for the treatments T₁, T₂, T₃ and T₄ was 7.50, 7.63, 7.19 and 6.88, respectively. Kakde *et al.* (2019) prepared *Lassi* from buffalo milk by adding wheatgrass extract colour and appearances score were observed in treatments T₁, T₂, T₃ and T₄ were 7.50, 7.63, 7.19 and 6.88, respectively. Deshpande *et al.* (2020) prepare the flavoured *Lassi* by blending buttermilk and soymilk in different portions and found the similar score. The colour and appearance score for four samples was 9.0, 8.0, 8.5 and 7.0, respectively.

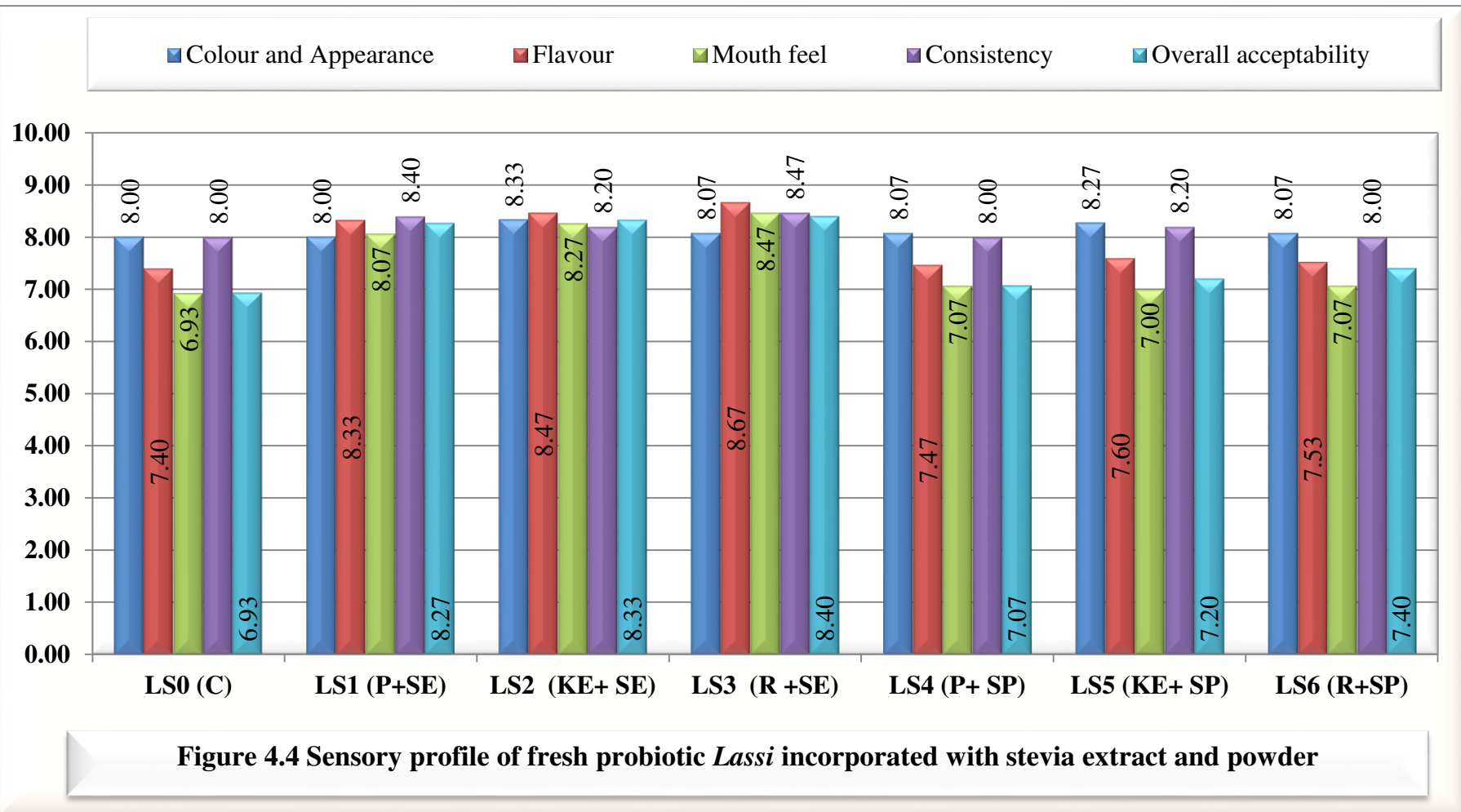
Table 4.9 Sensory profile of fresh probiotic *Lassi* incorporated with stevia extract and powder

Sample No.	Colour and Appearance	Flavour	Consistency	Mouth feel	Overall acceptability
LS ₀ (C)	8.00 ±0.20	7.40 ±0.19	8.00±0.22	6.93±0.21	6.93±0.18
LS ₁ (P+SE)	8.00±0.21	8.33±0.23	8.40±0.18	8.07±0.22	8.27±0.21
LS ₂ (KE+ SE)	8.33±0.19	8.47±0.24	8.20±0.20	8.27±0.23	8.33±0.11
LS ₃ (R +SE)	8.07±0.18	8.67±0.12	8.47±0.13	8.47±0.17	8.40±0.21
LS ₄ (P+ SP)	8.07±0.16	7.47±0.21	8.00±0.22	7.07±0.11	7.07±0.21
LS ₅ (KE+ SP)	8.27±0.18	7.60±0.20	8.20±0.21	7.00±0.13	7.20±0.15
LS ₆ (R+SP)	8.07±0.15	7.53±0.25	8.00±0.19	7.07±0.19	7.40±0.21
S.Em. ±	0.19	0.16	0.20	0.19	0.18
C.D. (P = 0.05)	NS	0.46*	NS	0.53*	0.50*
C.V. %	9.05	8.62	9.53	9.64	9.00
<p>Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract, SP- Stevia Powder, (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance.</p>					

4.3.2 Flavour score of probiotic *Lassi* on the day of preparation:

The flavour of any food substance plays a vital role in the acceptability of particular food and makes the foodstuff innovative. The Flavour score of *Lassi* on the day of preparation is presented in Table 4.9 and Fig. 4.4.

The highest average value for flavour was observed in LS₃ (8.67) which was “liked very much”, followed by LS₂ (8.47), LS₁ (8.33), LS₃ and LS₅ (7.60) lowest score was observed in LS₀ (7.40), respectively which was “like moderately”.



flavoured *Lassi* obtained a higher score compared to the control. There was a significant difference ($p < 0.05$) between the flavour score provided by panel members to the samples of *Lassi* prepared with stevia extract and powder along with control.

Results were inclined with Poul *et al.* (2019) prepared *Lassi* by using standardized milk where T₀ was control (cane sugar), T₁ (8% Honey), T₂ (75 µl Stevia) and T₃ (8% Honey + 75 µl Stevia) added to *Lassi*. The flavour score for four different treatments was 8.87, 8.81, 8.67 and 8.54, respectively. However, Dhumal *et al.* (2018) assessed the flavour score of *pudiva Lassi* for the T₁, T₂, T₃ and T₄ as 7.68, 7.88, 7.38 and 7.06, respectively. Kakde *et al.* (2019) prepared *Lassi* from buffalo milk by adding wheatgrass extract flavour were observed as in treatment T₁, T₂, T₃ and T₄ were 8.38, 7.13, 7.50 and 7.75, respectively. Deshpande *et al.* (2020) prepared the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. The flavour scores for A, B, C and D was 8.5, 8.0, 8.0 and 7.0, respectively.

4.3.3 Consistency score of probiotic *Lassi* on the day of preparation:

Consistency in dairy products refers to the uniformity of food products and their compatibility with other ingredients. The consistency score of *Lassi* on the day of preparation is presented in Table 4.9 and Fig. 4.4. The highest average value for consistency was observed in LS₃ (8.47), followed by LS₁ (8.40), LS₂ and LS₅ (8.20) lowest score determined in LS₀ LS₄, and LS₆ (8.00), respectively. No significant difference ($p < 0.05$) was found between the samples of *Lassi* prepared with stevia extract and powder as well as in control sample.

Similar results were reported by Poul *et al.* (2019) prepared *Lassi* by using standardized milk where T₀ was control (cane sugar), T₁ (8% Honey), T₂ (75 µl Stevia) and T₃ (8% Honey + 75 µl Stevia) added to *Lassi* and reported the consistency score for T₀, T₁, T₂ and T₃ was 8.82, 8.86, 8.84 and 8.44, respectively. Kakde *et al.* (2019) *Lassi* prepared from buffalo milk with the addition of wheatgrass extract, the average score of *wheatgrass Lassi* in T₁, T₂, T₃ and T₄ was 8.38, 7.13, 7.50 and 7.75, respectively by using 9 point hedonic rating scale.

4.3.4 Mouth feel score of probiotic *Lassi* on the day of preparation:

Mouth feel is a fundamental sensory characteristic for analysing the overall flavour and taste. Sometimes it is referred to as texture. The mouth feel score

of *Lassi* on the day of preparation is presented in Table 4.9 and Fig. 4.4. The highest average value for mouth feel was observed in LS₃ (8.47) which was “liked very much” followed by LS₂ (8.27), LS₁ (8.07), LS₄, LS₆ (7.07), however, lowest score was determined in LS₅ (7.00) LS₀ (6.93), respectively. There was a significant difference ($p < 0.05$) found between the samples of *Lassi* prepared with different flavours, stevia extract and powder along with control sample.

Similar results were reported by Dhumal *et al.* (2018) the mouth feel score of *pudina Lassi* for the T₁, T₂, T₃ and T₄ was 8.13, 8.25, 7.63 and 7.50, respectively.

4.3.5 Overall acceptability score of probiotic *Lassi* on the day of preparation:

The acceptance or rejection of food entirely depends on whether it corresponds to consumer expectations and needs (Mosca *et al.* 2015). The process through which an individual accepts or rejects food is considered multi-dimensional and is measured by overall acceptability. The overall acceptability score of *Lassi* on the day of preparation is presented in Table 4.9 and Fig. 4.4. The highest average value for overall acceptability was observed in LS₃ (8.40) which was “liked very much”, followed by LS₂ (8.33), LS₁ (8.27), LS₆ (7.40), the lowest score was determined in LS₅ (7.20) and LS₀ (6.93) respectively. A significant difference ($p < 0.05$) was found between the overall acceptability score allotted by panel members to the samples of *Lassi* prepared with stevia extract and powder along with control sample.

Poul *et al.* (2019) reported that the *Lassi* was prepared with the addition of T₀ – Control cane sugar, T₁ - 8 per cent Honey, T₂ - 75 µl Stevia and T₃ - 8 per cent Honey + 75 µl Stevia. Overall acceptability score for T₀, T₁, T₂ and T₃ was 8.79, 8.86, 8.87 and 8.62, respectively.

Dhumal *et al.* (2018) found the overall score of acceptability of *pudina Lassi* prepared with different amounts of *pudina* extract for the four treatments was 7.80, 8.00, 7.36 and 7.17, respectively.

Deshpande *et al.* (2020) prepare the flavoured *Lassi* by blending buttermilk and soymilk at different concentrations. Overall acceptability for A, B, C and D was 8.5, 8.0, 8.5 and 7.0, respectively.

4.4 SENSORY PROFILE OF PROBIOTIC YOGURT AT THE DAY OF PREPARATION:

4.4.1 Colour and appearance score of probiotic yogurt on the day of preparation:

Appearance is a key component of sensory assessment. The vision system is the detector of appearance. Using the eyes, we can measure optical and physical characteristics. The colour and appearance score of yogurt on the day of preparation is presented in Table 4.10. The highest average value for colour and appearance was observed in YS₂ (8.40) which was “liked very much”, followed by YS₁ (8.27), YS₃, YS₄ and YS₅ (8.20) lowest score was found in YS₆ and YS₀ (8.13), respectively which was also obtained score “liked very much” category. Yogurt samples prepared with stevia extract and powder did not differ significantly ($p < 0.05$) along with control sample.

Phalgun *et al.* (2020) investigated the effects of utensils used in curd preparation on sensory profile. The curd was prepared by T₁- utilising earthen, T₂- stainless steel, T₃- aluminium, T₄- plastic and T₅- china clay utensils with colour and appearance scores recorded for five treatments 8.70, 7.73, 7.11, 6.91 and 8.45, respectively.

4.4.2 Flavour score of probiotic yogurt on the day of preparation:

The flavour of any food substance plays a vital role in the acceptability of particular food and makes the foodstuff innovative. The flavour score of yogurt on the day of preparation is presented in Table 4.10. The highest average value for flavour was observed in YS₂ (8.00) which was “liked very much” followed by YS₁, YS₃ (7.93), YS₅ (7.87), YS₄ (7.73) and YS₆ (7.60) lowest score was observed in YS₀ (6.87), respectively which was “liked slightly”. Flavoured yogurt obtained more scores compared to control. A significant difference ($p < 0.05$) was found among the samples of yogurt prepared with stevia extract and powder as well as in control sample.

Phalgun *et al.* (2020) investigated the effects of utensils used in curd preparation on sensory profile. The curd was prepared by T₁- utilising earthen, T₂- stainless steel, T₃ - aluminium, T₄-plastic and T₅-china clay utensils with flavour scores were recorded for five treatments 8.33, 8.14, 7.93, 7.65 and 8.58, respectively.

4.4.3 Consistency score of probiotic yogurt on the day of preparation:

Consistency in dairy products refers to uniformity and its compatibility with other components. The consistency score of yogurt on the day of preparation is presented in Table 4.10. The highest average value for consistency was observed in YS₂ (8.40) which was “liked very much” followed by YS₁ (8.20), YS₃ (8.13) and YS₄ (8.07) lowest score was determined in YS₀ and YS₆ (8.00), respectively which was also found in category “liked very much. No significant difference in consistency score ($p < 0.05$) was found among the samples of yogurt prepared with stevia extract and powder along with control sample.

Phalgun *et al.* (2020) investigated the effects of utensils used in curd preparation on sensory profile. The curd was prepared by T₁- utilising earthen, T₂- stainless steel, T₃- aluminium, T₄-plastic and T₅-china clay utensils with body and texture were recorded for five treatments 8.67, 7.54, 7.24, 6.92 and 8.34, respectively.

4.4.4 Mouth feel score of probiotic yogurt on the day of preparation:

Mouth feel is a crucial sensory facet of exploring the overall flavour and taste. Sometimes it is referred to as texture. The mouth feels score of yogurt on the day of preparation is presented in Table 4.10. The highest average value for mouth feel was observed in YS₂ (8.00), which was “liked very much” followed by YS₃ (8.93), YS₁ (7.80), YS₅ (7.20), YS₄ (7.13), YS₆ (7.07) however, the lowest score was determined in YS₀ (6.87), respectively which was “liked slightly”. A significant difference ($p < 0.05$) was found among the samples of yogurt prepared with different flavours, stevia extract and powder along with control sample.

4.4.5 Overall acceptability score of probiotic yogurt on the day of preparation:

The acceptance or rejection of food entirely depends on whether it corresponds to consumer expectations and needs (Mosca *et al.* 2015). The process through which an individual accepts or rejects food is considered to be of a multi-dimensional nature and is measured by overall acceptability. The overall acceptability score of yogurt on the day of preparation is presented in Table 4.10. The highest average value for overall acceptability was observed in YS₂ (8.12) which was

“liked very much”, followed by YS₃ (8.07), YS₁ (8.00), YS₅ (7.20), YS₄ (7.13) YS₆ (7.07) however lowest score was determined in YS₀ (7.00), respectively which was “moderately liked” by the panel members. There was a significant difference ($p < 0.05$) found between the score of overall acceptability allotted by panel members to the samples of yogurt prepared with stevia extract and powder as well as with control sample.

The satiety of yogurts was directly correlated to sweetness level. Liking for chocolate bars and peaches declined after panellists were served yogurt with a high sweetness level. The reason behind this decrease in liking was found to be the overwhelming sweetness which concealed the characteristic yogurt sour taste (Desai, 2012).

Table 4.10 Sensory profile of fresh probiotic yogurt incorporated with stevia extract and powder

Sample No.	Colour and Appearance	Flavour	Consistency	Mouth feel	Overall acceptability
YS ₀ (C)	8.13±0.19	6.87±0.13	8.00±0.23	6.87±0.22	7.00±0.23
YS ₁ (K+SE)	8.27±0.18	7.93±0.22	8.20±0.16	7.80±0.13	8.00±0.17
YS ₂ (KP+SE)	8.40±0.15	8.00±0.26	8.40±0.14	8.00±0.24	8.12±0.18
YS ₃ (R+SE)	8.20±0.18	7.93±0.22	8.13±0.17	7.93±0.18	8.07±0.18
YS ₄ (C+SP)	8.20±0.21	7.73±0.23	8.07±0.18	7.13±0.17	7.13±0.25
YS ₅ (K+SP)	8.20±0.19	7.87±20	8.07±0.19	7.20±0.26	7.20±0.26
YS ₆ (KP+SP)	8.13±0.17	7.60±21	8.00±0.26	7.07±0.16	7.07±0.24
S.Em. ±	0.18	0.14	0.20	0.20	0.22
C.D. (P = 0.05)	NS	0.40*	NS	0.57*	0.63*
C.V. %	8.64	7.63	9.44	10.00	11.60

Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance.

Phalgun *et al.* (2020) investigated the effects of utensils used in curd preparation on sensory profile. The curd was prepared by T₁- utilising earthen, T₂- stainless steel, T₃- aluminium, T₄-plastic and T₅-china clay utensils with overall acceptability were recorded for five treatments 8.78, 8.15, 7.93, 7.53 and 8.54, respectively.

By observing the mean score of overall acceptability of both *Lassi* and yogurt prepared with stevia powder and extract, *Lassi* and yogurt prepared with stevia extract obtained more scores compared with *Lassi* and yogurt prepared with stevia powder; however, control obtained lower scores compared with the both products. Among all the samples highest score was obtained, i.e. Rose flavour followed by Kesar Elaichi and Pineapple, whereas in yogurt highest score was Kevda, Kesar Pista and Raspberry which was prepared with stevia extract. So stevia extract incorporated products were selected for further work.

4.5 SENSORY PROFILE OF PROBIOTIC *LASSI* DURING STORAGE:

Based on the acceptability stevia extract incorporated in different flavoured *Lassi*, yogurt was selected for storage studies. Along with their control samples, both *Lassi* and yogurt samples were stored at refrigeration temperature ($4\pm 2^{\circ}$ C) in glass bottles and jars, respectively. They were analysed for the physio-chemical parameter, sensory score (*viz.* Colour and appearance, Flavour, Consistency, Mouth feel and Overall acceptability) and microbiological (*i.e.* Lactobacilli count, Streptococci count, yeast and mould count, coliform count). The analysis of selected *Lassi* and yogurt samples was stopped to study when it was found to be unacceptable by the panellists.

4.5.1 Colour and appearance score of probiotic *Lassi* during storage:

The data in the Table 4.11 and Fig. 4.5 showed colour and appearance score range for LS₀, LS₁, LS₂ and LS₃ was 6.60-8.00, 6.67-8.00, 7.00-8.33 and 6.73-8.07, respectively. On the day of preparation highest score was LS₂ (8.33) which was “liked very much” and the lowest score was LS₀ and LS₁ (8.00), which was “liked very much”, at the 11th day of storage, the highest score was observed in LS₂ (7.80) and lowest score LS₀ (6.60) which was “liked slightly”. The colour and appearance of all the flavoured probiotic *Lassi* and control non-significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples.

Shuwu *et al.* (2011) reported there was a gradual decrease in colour and appearance of the honey *Lassi* from 8.57 on the first day of storage to 7.64 on the 10th day.

Jadhav *et al.* (2014) reported the mean score varied from 7.10 to 8.48 during the storage period. The blending of orange juice in different levels of the *Lassi* samples did not affect significantly.

Table 4.11 Colour and appearance score of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.E m. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	8.00 ±0.20	8.00 ±0.19	7.93 ±0.18	7.87 ±0.19	6.80 ±0.22	6.60 ±0.15	7.53	0.19	NS	9.53
LS ₁ (P+SE)	8.00 ±0.21	8.00 ±0.19	7.93 ±0.20	7.87 ±0.20	6.80 ±0.17	6.67 ±0.16	7.54	0.19	NS	9.35
LS ₂ (KE+SE)	8.33 ±0.19	8.13 ±0.19	8.13 ±0.19	8.07 ±0.19	7.00 ±0.19	7.00 ±0.22	7.77	0.20	NS	9.74
LS ₃ (R+SE)	8.07 ±0.18	8.00 ±0.18	8.07 ±0.26	7.93 ±0.18	6.93 ±0.20	6.73 ±0.22	7.62	0.21	NS	10.11
Mean	8.10	8.03	8.01	7.93	6.88	6.75	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em).			
S.Em. ±	0.20	0.19	0.22	0.19	0.20	0.20				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	9.26	9.08	10.35	9.37	9.73	9.86				

4.5.2 Flavour score of probiotic *Lassi* during storage:

The data in the Table 4.12 showed flavour score range for LS₀, LS₁, LS₂ and LS₃ was 5.00-7.40, 5.53-8.33, 5.20-8.47 and 5.40-8.67, respectively. On the day of preparation highest score was LS₃ (8.67) which was “liked very much” and the lowest score was LS₀ (7.47), which was “liked moderately”, at the 11th day of storage, the highest score was observed in LS₃ (5.40) and lowest score LS₀ (5.00) which was “neither liked nor disliked”. The flavour of all the flavoured probiotic *Lassi* and control samples were significantly (p<0.05) decreased throughout the storage period from the day of preparation to the 11th day of storage.

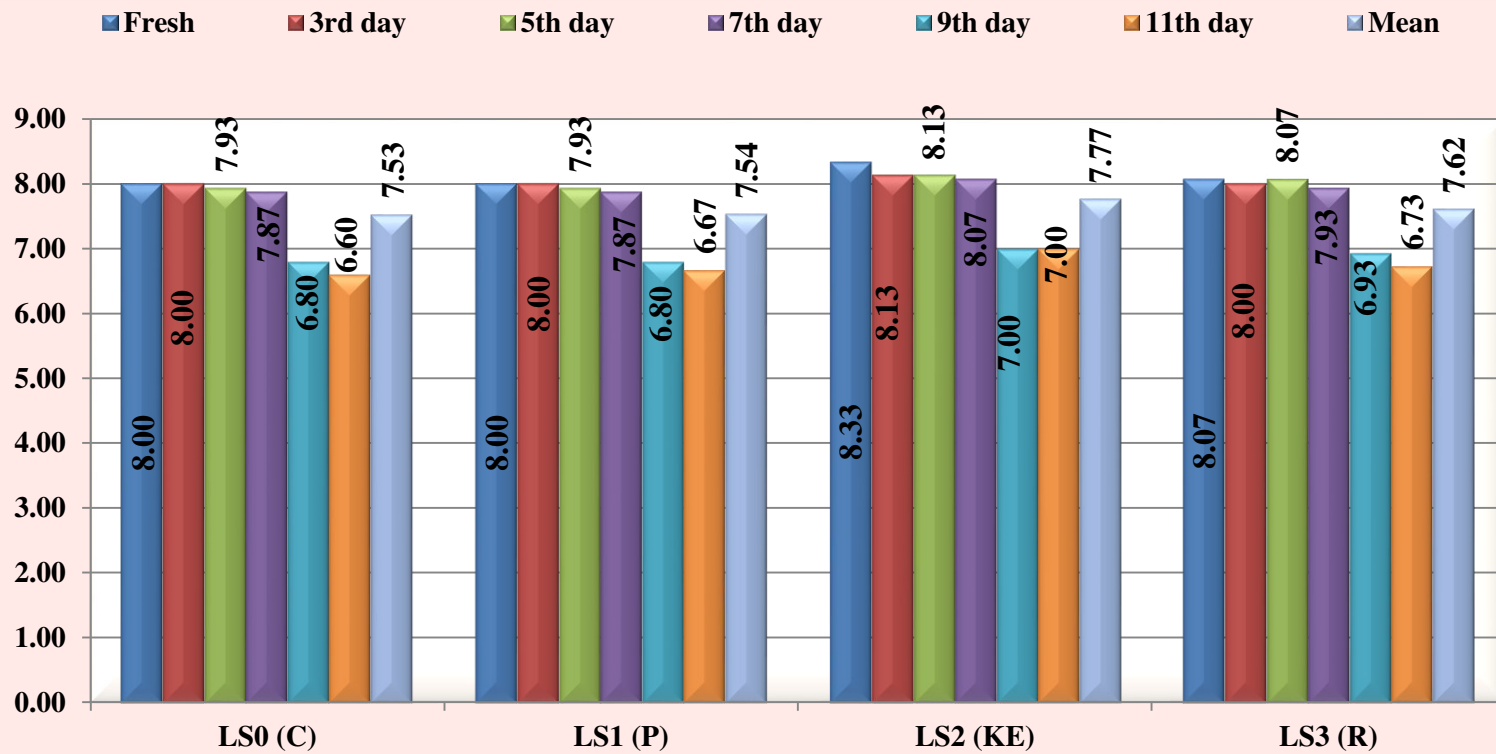


Figure 4.5 Colour and appearance score of probiotic *Lassi* during storage

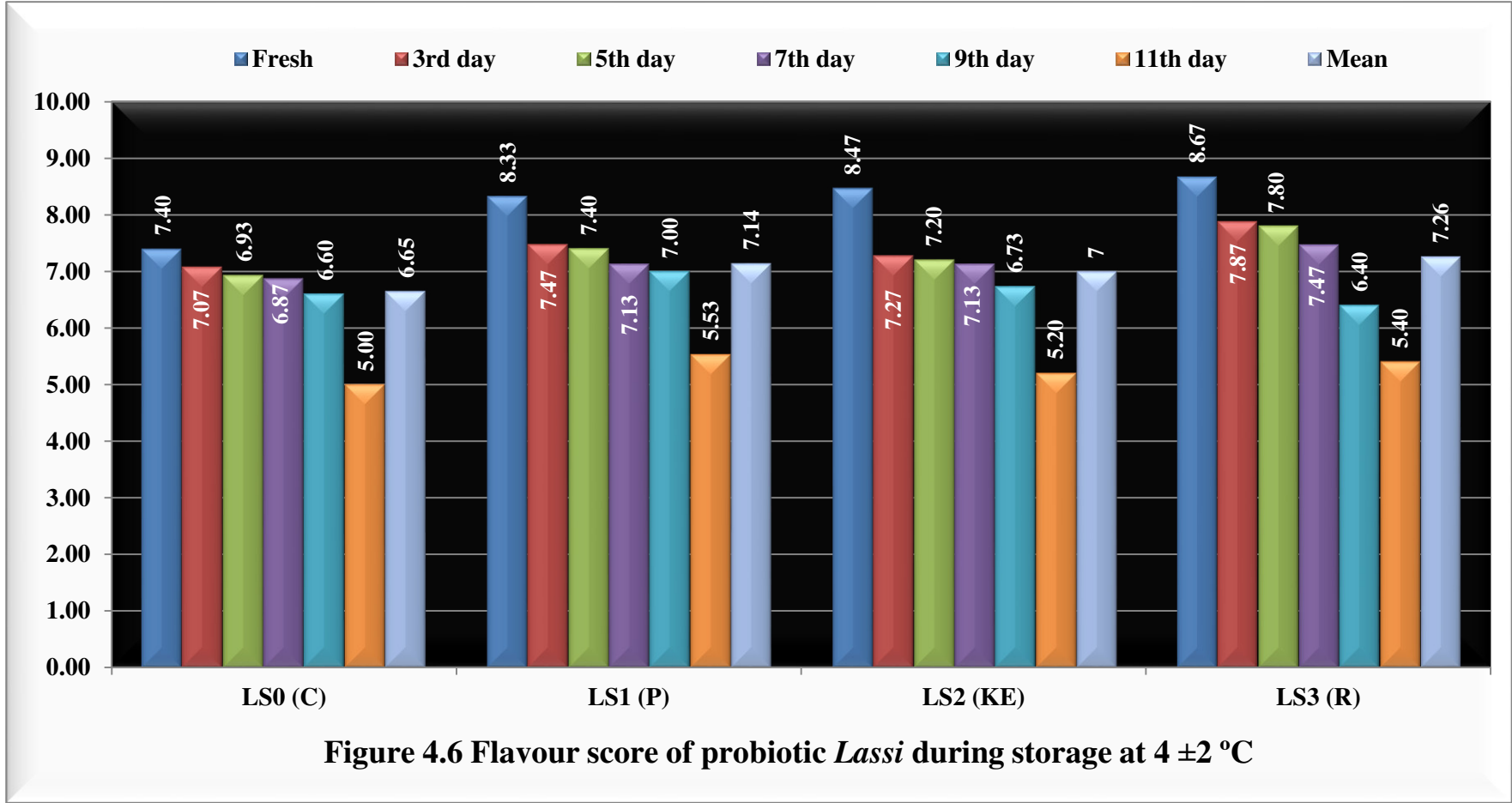
Yadav *et al.* (2007) assessed the storage modifications in probiotic *Dahi* stored at 7°C up to 8 days. On a fresh day, the flavour score was 21.6 out of 25 and on day 8, they observed a little decline in flavour and the score was 19.6 out of 25.

Sharma (2010) found the flavour score decreased slowly in both control and synbiotic *Lassi* from 7.78 to 5.59 and 7.90 to 5.68, respectively, during storage of 28 days at refrigerated temperature. Shukla *et al.* (2013) developed probiotic beverages from whey and pineapple juice. They found that the initial flavour score was 8.82 and after 8 days of storage under cold storage, it decreased to 8.64.

Table 4.12 Flavour score of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.E m. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	7.40 ±0.19	7.07 ±0.11	6.93 ±0.21	6.87 ±0.09	6.60 ±0.12	5.00 ±0.12	6.65	0.15	0.42*	8.40
LS ₁ (P+SE)	8.33 ±0.23	7.47 ±0.21	7.40 ±0.19	7.13 ±0.16	7.00 ±0.10	5.53 ±0.13	7.14	0.16	0.45*	9.68
LS ₂ (KE+SE)	8.47 ±0.24	7.27 ±0.18	7.20 ±0.17	7.13 ±0.11	6.73 ±0.19	5.20 ±0.09	7.00	0.17	0.48*	9.11
LS ₃ (R+SE)	8.67 ±0.12	7.87 ±0.19	7.80 ±0.20	7.47 ±0.16	6.40 ±0.19	5.40 ±0.10	7.26	0.17	0.47*	8.44
Mean	8.23	7.42	7.33	7.15	6.68	5.28	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (Mean ±S.Em), *Significant at 5% level of significance.			
S.Em. ±	0.19	0.18	0.19	0.14	0.16	0.12				
C.D. (P = 0.05)	0.53*	0.51*	0.55*	0.39*	0.44*	0.33*				
C.V.%	9.41	9.42	10.20	7.52	8.62	6.81				

Singh *et al.* (2013) studied the storage changes of whey-based pineapple and bottle gourd mixed herbal drinks. They found that the taste and flavour reduced significantly with the storage period, and the mean value of 7.50 for flavour and taste in the on the day of preparation, beverage score decreased 5.90 and 4.90 respectively after 20 days of storage.



4.5.3 Consistency score of probiotic *Lassi* during storage:

The data in the Table 4.13 exhibited the consistency score range for LS₀, LS₁, LS₂ and LS₃ was 6.80-8.00, 7.00-7.93, 6.87-8.20 and 7.20-8.47, respectively. On the day of preparation highest score was LS₃ (8.47) which was “liked very much” and the lowest score was LS₁ (7.93), which was “liked moderately”, whereas, on the 11th day of storage, the highest score was observed in LS₃ (7.20) and lowest score LS₀ (6.80) which was “liked slightly”. The consistency score of all the flavoured probiotic *Lassi* and control non-significantly ($p < 0.05$) gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples.

Table 4.13 Consistency score of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.E m. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	8.00 ±0.22	7.93 ±0.27	7.73 ±0.24	7.60 ±0.37	7.00 ±0.35	6.80 ±0.09	7.51	0.28	NS	14.12
LS ₁ (P+SE)	7.93 ±0.18	7.73 ±0.23	7.60 ±0.23	7.60 ±0.23	7.00 ±0.19	7.00 ±0.15	7.47	0.26	NS	13.66
LS ₂ (KE+SE)	8.20 ±0.20	8.20 ±0.22	8.07 ±0.26	7.93 ±0.27	7.60 ±0.27	6.87 ±0.13	7.81	0.33	NS	16.07
LS ₃ (R+SE)	8.47 ±0.13	8.20 ±0.26	8.00 ±0.25	7.80 ±0.26	7.73 ±0.28	7.20 ±0.19	7.90	0.29	NS	14.33
Mean	8.15	8.01	7.85	7.73	7.33	6.96	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em).			
S.Em. ±	0.19	0.25	0.25	0.29	0.25	0.15				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V.%	8.84	11.87	12.45	14.51	12.94	8.21				

4.5.4 Mouth feel score of probiotic *Lassi* during storage:

The data in the Table 4.14 exhibited the mouth feel score range for LS₀, LS₁, LS₂ and LS₃ was 4.90-6.93, 5.33-8.07, 5.20-8.07 and 5.27-8.47, respectively. On the day of preparation highest score was LS₃ (8.47) which was “liked very much” and the

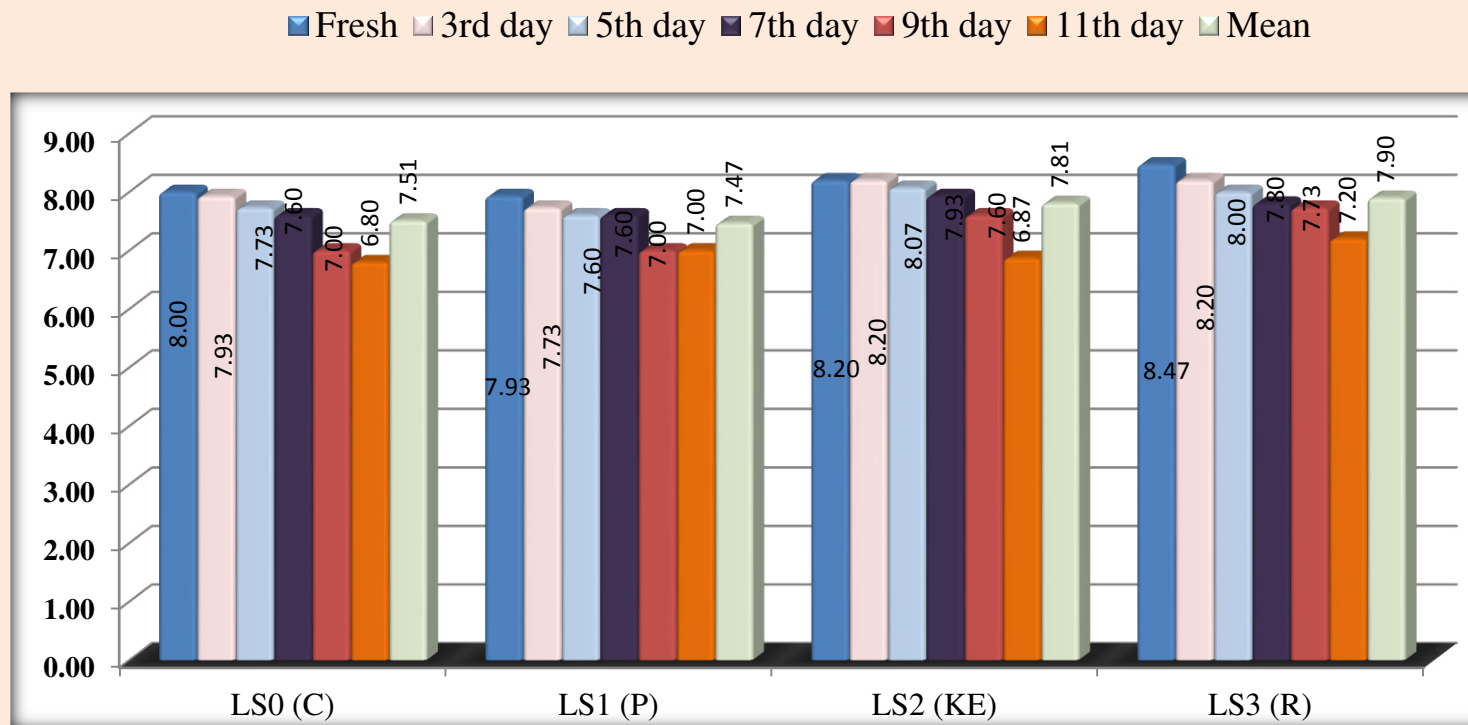


Figure 4.7 Consistency score of probiotic *Lassi* during storage at 4 ± 2 °C

lowest score was LS₁ (6.93), which was “liked slightly”, at the 11th day of storage, the highest score was observed in LS₃ (5.27) and lowest score LS₀ (4.90) which was “disliked slightly”. The mouth feel score of all the flavoured probiotic *Lassi* and control sample significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage.

Table 4.14: Mouth feel score of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.E. m. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	6.93 ±0.20	6.87 ±0.24	6.80 ±0.25	6.73 ±0.39	6.27 ±0.28	4.90 ±0.27	6.41	0.23	0.64*	13.31
LS ₁ (P+SE)	8.07 ±0.23	8.13 ±0.28	8.00 ±0.22	7.87 ±0.26	7.67 ±0.33	5.33 ±0.27	7.51	0.16	0.43*	12.98
LS ₂ (KE+S E)	8.07 ±0.22	8.00 ±0.21	8.00 ±0.21	7.93 ±0.33	7.80 ±0.33	5.20 ±0.31	7.05	0.18	0.49*	8.94
LS ₃ (R+SE)	8.47 ±0.16	8.27 ±0.16	8.07 ±0.24	8.03 ±0.32	7.73 ±0.32	5.27 ±0.25	7.65	0.25	0.70*	12.31
Mean	7.88	7.81	7.71	7.66	7.36	5.17	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (Mean ±S.Em), *Significant at 5% level of significance.			
S.E m. ±	0.21	0.23	0.23	0.33	0.31	0.29				
C.D. (P = 0.05)	0.59*	0.65*	0.66*	0.92*	0.87*	0.81*				
C.V.%	10.25	11.39	11.66	16.47	16.18	15.83				

4.5.5 Overall acceptability score of probiotic *Lassi* during storage:

The data in the Table 4.15 exhibited the overall acceptability score range for LS₀, LS₁, LS₂ and LS₃ was 5.00-6.93, 5.40-8.27, 5.20-8.33 and 5.80-8.40, respectively. On the day of preparation highest score was LS₃ (8.40) which was “liked very much” and the lowest score was LS₁ (6.93), which was “liked slightly”, at the 11th day of storage, the highest score was observed in LS₃ (5.80) neither liked nor disliked and lowest score LS₀ (5.00) which was also “neither liked nor disliked. The

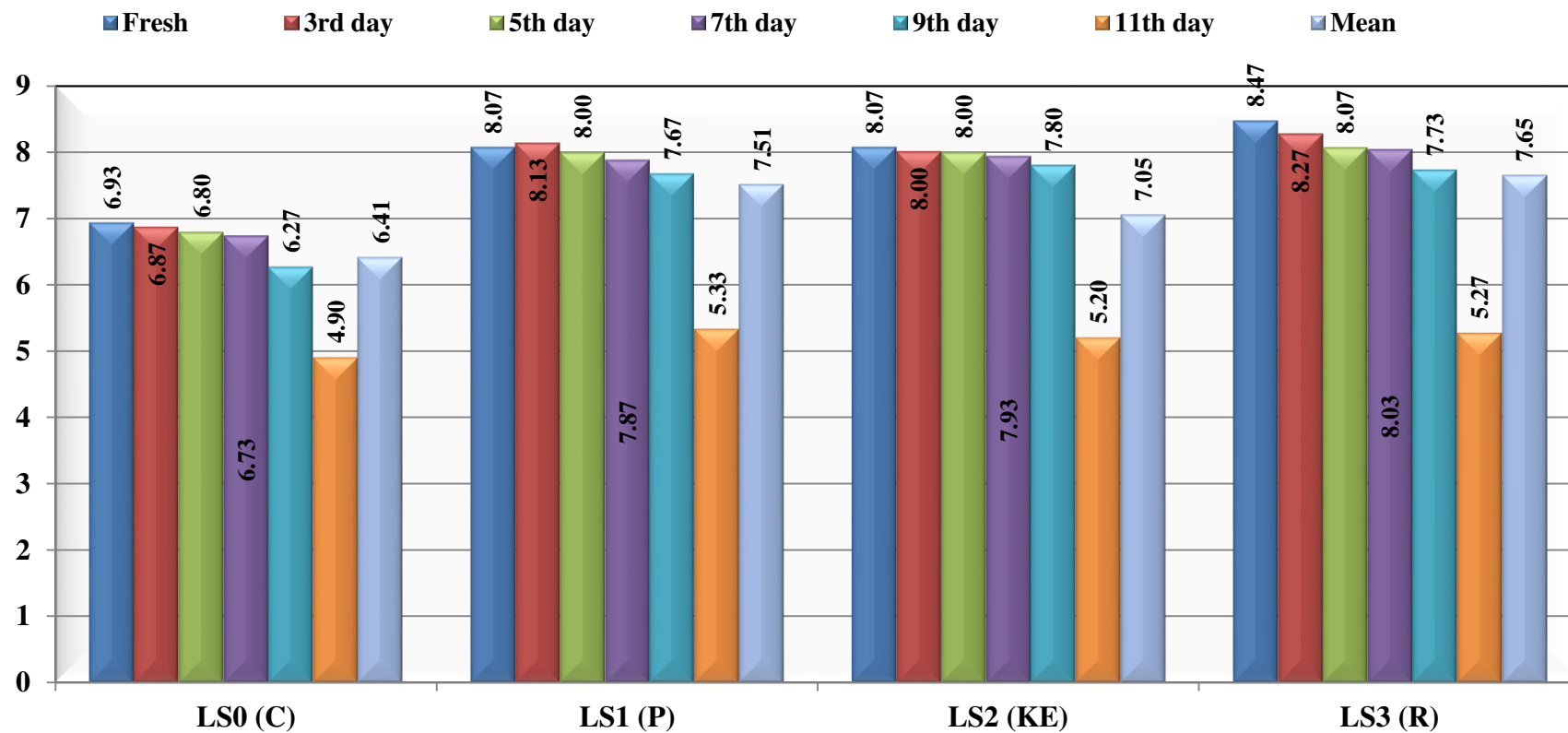


Figure 4.8 Mouth feel score of probiotic *Lassi* during storage at 4 ± 2 °C

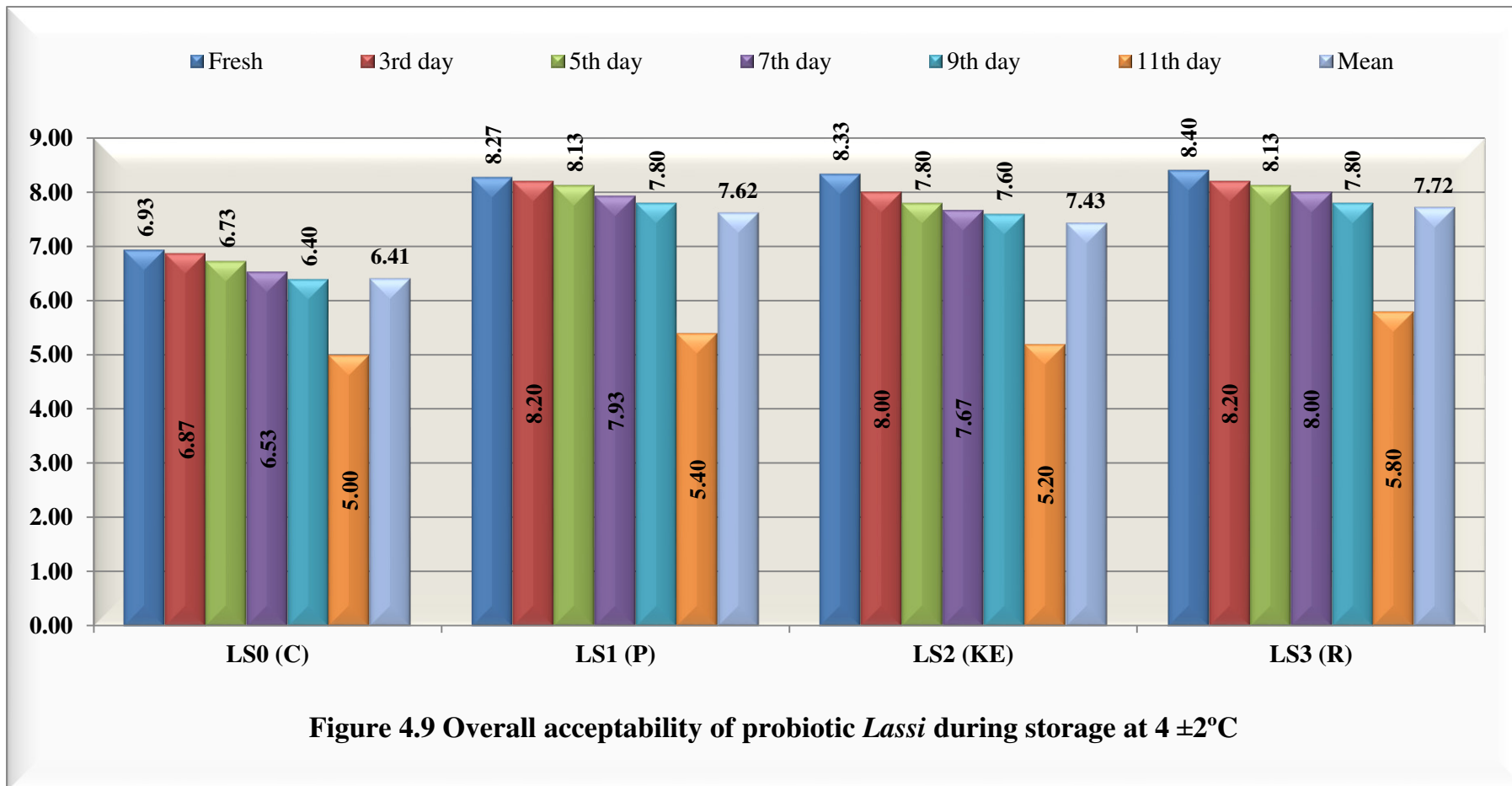
overall acceptability score of all the flavoured probiotic *Lassi* and control significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage. Shah and Prajapati (2014) studied all the sensory characteristics, the score was higher for the product S₃ (15% Sugar) followed by S₂ (12% Sugar) and S₁ (10% Sugar) and the average overall acceptability scores were 8.22, 7.52, and 7.11, respectively.

Singh *et al.* (2013) studied the modifications during the shelf life of whey-based pineapple and bottle gourd mixed herbal (WPBH) beverages. They reported that the overall acceptability of the WPBH beverage during 20 days of storage duration reduced significantly from 8.02 to 5.80 and the product was slightly acceptable up to 15 days, having a score of 6.45 for overall acceptability.

Table 4.15 Overall acceptability of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	6.93 ±0.18	6.87 ±0.21	6.73 ±0.22	6.53 ±0.21	6.40 ±0.21	5.00 ±0.21	6.41	0.21	0.39*	12.34
LS ₁ (P+SE)	8.27 ±0.21	8.20 ±0.24	8.13 ±0.27	7.93 ±0.31	7.80 ±0.31	5.40 ±0.28	7.62	0.28	0.62*	13.48
LS ₂ (KE+SE)	8.33 ±0.11	8.00 ±0.16	7.80 ±0.17	7.67 ±0.18	7.60 ±0.27	5.20 ±0.25	7.43	0.20	0.58*	10.13
LS ₃ (R+SE)	8.40 ±0.21	8.20 ±0.22	8.13 ±0.21	8.00 ±0.23	7.80 ±0.26	5.80 ±0.22	7.72	0.23	0.63*	11.06
Mean	7.98	7.81	7.69	7.53	7.40	5.35	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract, (Mean ±S.Em), *Significant at 5% level of significance.			
S.Em. ±	0.19	0.21	0.23	0.24	0.27	0.24				
C.D. (P = 0.05)	0.54*	0.60*	0.64*	0.69*	0.76*	0.69*				
C.V. %	9.29	10.62	11.35	12.54	13.99	13.08				

Desai *et al.* (2016) reported that the overall acceptability of cultured buttermilk made with the blending of *Dahi* and fermented whey in 60:40 ratio declined from 7.94 on the first day to 6.57 on the 5th day of storage, which was stored in PET bottles at 7±2° C.



The current study also follows similar trends for the acceptability of the product. The potential reasons for a significant decline in acceptability scores would be the formation of flaxes in the product, hardening of fat globules, growth of non-uniform curd particles, clinging of the product at the walls of the glass container, increase in acidity, continuous proteolysis, dearth of freshness, after 11 days of storage.

4.6 PHYSIO-CHEMICAL PROPERTIES OF PROBIOTIC *LASSI* DURING STORAGE:

4.6.1 Moisture content of probiotic *Lassi* during storage:

The data in the Table 4.16 showed moisture content range for LS₀, LS₁, LS₂ and LS₃ was 85.94-87.27, 86.10-88.18, 86.12-87.90 and 86.06-87.31 per cent, respectively. The moisture content of all the samples decreased non significantly ($p < 0.05$) throughout the storage period from fresh yogurt to the 11th day of storage in all *Lassi* samples. Kakde *et al.* (2019) reported that treatments T₁, T₂, T₃, T₄ and T₅ were prepared on the 0 days of *Lassi* moisture percentage were reduced with the amount of incorporation of sapota pulp in four treatments i.e. 87.60, 87.10, 86.38, 85.73 and 85.06 per cent, respectively and on the eighth day of storage, mean moisture percentages were noted as 81.55, 80.93, 80.09, 79.56 and 79.14 per cent, respectively.

4.6.2 Total solids content of probiotic *Lassi* during storage:

The data in Table 4.17 showed that the total solids content range for LS₀, LS₁, LS₂ and LS₃ was 12.70-14.01, 11.79-13.90, 12.05-13.78 and 12.52-13.92 per cent, respectively. As moisture content turned down due to that, the total solids content was non-significantly ($p < 0.05$) more than before. Similar developments followed by Shinde *et al.* 2015 observed that total solids were increased as the increase in the rate of addition of sapota pulp, i.e. at the 0th day of preparation 12.40, 12.90, 13.62, 14.27 and 14.94, which increases at the eighth day of preparation 18.45, 19.07 19.91, 20.44 during storage for *Lassi* prepared under treatments T₁, T₂, T₃, T₄ and T₅, respectively.

Table 4.16 Moisture content (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	87.27 ±0.94	87.06 ±0.77	86.96 ±0.51	86.17 ±0.97	86.02 ±0.55	85.94 ±0.40	86.57	0.73	NS	1.88
LS ₁ (P+SE)	88.10 ±0.65	87.70 ±0.69	87.12 ±0.61	86.64 ±0.63	86.15 ±0.35	86.10 ±0.44	86.98	0.58	NS	1.49
LS ₂ (KE+SE)	87.90 ±0.85	87.27 ±0.53	87.04 ±0.34	86.93 ±0.59	86.37 ±0.35	86.12 ±0.31	86.93	0.57	NS	1.45
LS ₃ (R+SE)	87.31 ±0.78	87.14 ±0.41	86.99 ±0.55	86.73 ±0.23	86.08 ±0.54	86.06 ±0.42	86.71	0.52	NS	1.34
Mean	87.64	87.29	87.02	86.61	86.15	86.05	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.82	0.62	0.51	0.66	0.46	0.40				
C.V. %	2.09	1.59	1.32	1.71	1.20	1.04				

Table 4.17 Total solids content (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	12.70 ±0.42	12.93 ±0.37	13.03 ±0.31	13.80 ±0.29	13.93 ±0.28	14.01 ±0.38	13.40	0.37	NS	6.21
LS ₁ (P+SE)	11.79 ±0.41	12.28 ±0.36	12.87 ±0.34	13.35 ±0.41	13.84 ±0.25	13.90 ±0.31	13.05	0.36	NS	6.13
LS ₂ (KE+SE)	12.05 ±0.31	12.71 ±0.48	12.93 ±0.32	13.06 ±0.27	13.72 ±0.24	13.78 ±0.41	13.04	0.34	NS	6.01
LS ₃ (R+SE)	12.52 ±0.40	12.84 ±0.43	13.00 ±0.15	13.22 ±0.14	13.90 ±0.16	13.92 ±0.26	13.23	0.27	NS	4.69
Mean	12.31	12.69	12.95	13.36	13.84	13.90	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.39	0.42	0.29	0.29	0.24	0.35				
C.V. %	7.17	7.40	4.92	4.91	3.87	5.50				

4.6.3 Protein content of *Lassi* during storage:

The data in Table 4.18 showed protein content range for LS₀, LS₁, LS₂ and LS₃ was 3.52-4.82, 3.31-4.78, 3.41-4.80 and 3.37-4.64 per cent, respectively. Protein content was increased non-significantly ($p < 0.05$) during the storage period in all the samples of flavoured probiotic *Lassi* (Table 4.18). The proteolytic activity of Lactic acid bacteria, which hydrolyses proteins into peptides and amino acids, affects the protein level. Lactic acid bacteria require a wide range of amino acids for growth and their proteolytic enzyme complement is able to split most types of peptide bonds (Thomas and Mills, 1981). Probably, the free amino acid present in *Lassi* was the result of hydrolysis of protein under the influence of proteolytic enzymes. During the storage period, these free amino acids again link to form the peptide bonds that transform into protein. Hence the protein contents of *Lassi* increased during storage. The tyrosine value significantly ($P < 0.05$) increased in fermented milk from 2.08 to 2.89 mg/100 g for the period of 28 days of storage. This could be because of the initiation of proteolysis by starters themselves or mediated by enzymatic hydrolysis of certain metabolites (Shah and Prajapati 2004). Shinde *et al.* 2015 observed that on the 0 day of *Lassi* preparation, protein percentage was reduced with an increase in the rate of addition of sapota pulp *i.e.* 2.30, 2.21, 2.14, 2.06 and 1.97 per cent for *Lassi* on the eighth day of storage, average protein percentage were observed as 2.91, 2.82, 2.71, 2.60 and 2.58 per cent for *Lassi* T₁, T₂, T₃, T₄ and T₅, respectively which was increased during the storage period.

4.6.4 Fat content of *Lassi* during storage:

Fat content was slightly increased during the storage period as presented in Table 4.19. The data in the Table 4.19 showed the fat content range for LS₀, LS₁, LS₂ and LS₃ 3.50-3.89, 3.58-3.97, 3.59-4.00 and 3.57-3.95 per cent, respectively. There was no significant ($p < 0.05$) difference due to the addition of different flavours and duration of storage. The justification for the increase in fat percentage is due to the decline in the moisture content, which led to an increase in the percentage of total solids, including fat.

Table 4.18 Protein content (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	3.52 ±0.03	3.98 ±0.25	4.17 ±0.13	4.26 ±0.19	4.48 ±0.13	4.82 ±0.20	4.20	0.18	NS	9.68
LS ₁ (P+SE)	3.31 ±0.09	3.76 ±0.22	3.91 ±0.32	3.92 ±0.30	4.17 ±0.32	4.78 ±0.19	3.97	0.24	NS	13.42
LS ₂ (KE+SE)	3.41 ±0.04	3.61 ±0.17	3.96 ±0.20	4.20 ±0.26	4.52 ±0.20	4.80 ±0.19	4.08	0.22	NS	11.69
LS ₃ (R+SE)	3.37 ±0.03	3.61 ±0.20	3.71 ±0.21	4.21 ±0.18	4.38 ±0.21	4.64 ±0.15	3.98	0.16	NS	8.96
Mean	3.40	3.74	3.93	4.14	4.38	4.76	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.13	0.22	0.22	0.24	0.18	0.16				
C.V. %	8.23	12.98	12.49	13.07	8.99	7.68				

Table 4.19 Fat content (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	3.50 ±0.02	3.53 ±0.03	3.58 ±0.10	3.63 ±0.17	3.79 ±0.10	3.89 ±0.10	3.65	0.10	NS	6.32
LS ₁ (P+SE)	3.58 ±0.06	3.61 ±0.07	3.65 ±0.07	3.72 ±0.11	3.91 ±0.14	3.97 ±0.11	3.74	0.11	NS	5.91
LS ₂ (KE+SE)	3.59 ±0.09	3.73 ±0.16	3.80 ±0.15	3.89 ±0.12	3.95 ±0.05	4.00 ±0.20	3.82	0.14	NS	8.28
LS ₃ (R+SE)	3.57 ±0.02	3.61 ±0.13	3.73 ±0.16	3.76 ±0.11	3.89 ±0.06	3.95 ±0.07	3.79	0.10	NS	6.06
Mean	3.56	3.62	3.69	3.75	3.88	3.95	Note: LS: <i>Lassi</i> Sample, C- Control, R – Rose Flavour, KE- <i>Kesar Elachi</i> Flavour, P-Pineapple Flavour (NS)- Non Significant Each value is the average of five replications.			
S.Em. ±	0.06	0.11	0.13	0.13	0.10	0.13				
C.V. %	3.68	7.06	7.68	7.97	5.68	7.41				

4.6.5 Ash content of *Lassi* during storage:

The data in Table 4.20 showed ash content range for LS₀, LS₁, LS₂ and LS₃ 0.80-0.89, 0.77-0.82, 0.78-0.83 and 0.79-0.85 per cent, respectively. The ash content of flavoured probiotic *Lassi* as well as in the control sample was increased with the progression of the storage period. The increase in ash content could be attributed to a decrease in moisture (Abdalla and Abdel Razig, 1997).

4.6.6 Carbohydrate content of *Lassi* during storage:

The data in Table 4.21 showed the carbohydrate content range for LS₀, LS₁, LS₂ and LS₃ 4.17-4.74, 3.89-4.13, 4.01- 4.21 and 4.26-4.70 per cent, respectively. Carbohydrate content was slightly decreased during the storage period (Table 4.21). There was no significant (p<0.05) difference found due to the addition of different flavours and stevia extract during the storage period.

Table 4.20 Ash content (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P=0.05)	C.V. (%)
LS ₀ (C)	0.80 ±0.02	0.81 ±0.01	0.84 ±0.02	0.85 ±0.01	0.86 ±0.01	0.89 ±0.02	0.84	0.02	NS	4.11
LS ₁ (P+SE)	0.77 ±0.02	0.78 ±0.02	0.79 ±0.01	0.80 ±0.03	0.82 ±0.01	0.82 ±0.04	0.79	0.03	NS	5.09
LS ₂ (KE+SE)	0.78 ±0.02	0.79 ±0.02	0.80 ±0.01	0.82 ±0.02	0.82 ±0.03	0.83 ±0.03	0.81	0.02	NS	5.18
LS ₃ (R+SE)	0.79 ±0.01	0.80 ±0.01	0.82 ±0.01	0.84 ±0.02	0.85 ±0.02	0.85 ±0.03	0.82	0.03	NS	5.92
Mean	0.78	0.79	0.81	0.82	0.83	0.85	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.02	0.01	0.01	0.02	0.02	0.03				
C.V. %	4.28	3.80	3.15	5.28	5.25	7.39				

Table 4.21 Carbohydrate content (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P= 0.05)	C.V. (%)
LS ₀ (C)	4.74 ±0.14	4.57 ±0.18	4.42 ±0.21	4.29 ±0.20	4.20 ±0.28	4.17 ±0.27	4.39	0.23	NS	11.46
LS ₁ (P+SE)	4.13 ±0.22	4.11 ±0.17	4.06 ±0.36	4.04 ±0.22	4.01 ±0.14	3.89 ±0.11	4.05	0.22	NS	12.26
LS ₂ (KE+SE)	4.21 ±0.31	4.19 ±0.27	4.16 ±0.22	4.14 ±0.17	4.03 ±0.19	4.01 ±0.26	4.12	0.23	NS	12.22
LS ₃ (R+SE)	4.70 ±0.29	4.73 ±0.37	4.68 ±0.13	4.58 ±0.23	4.53 ±0.15	4.26 ±0.18	4.58	0.24	NS	11.36
Mean	4.44	4.40	4.33	4.26	4.19	4.08	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.25	0.26	0.25	0.18	0.18	0.22				
C.V. (%)	12.62	13.14	12.85	9.47	9.68	12.19				

4.6.7 pH level of *Lassi* during storage:

The data in Table 4.22 showed that the pH level varies for LS₀, LS₁, LS₂ and LS₃ was 4.07-4.40, 4.02-4.39, 4.01-4.38 and 4.05-4.39, respectively. pH content was slightly decreased significantly (p<0.05) during the storage period (Table 4.22). The continuous growth and metabolic activity of lactic acid bacteria lead to the accumulation of organic acids and cause the reduction in the pH of fermented milk (Ruggeri *et al.* 2008). There was a significant difference found in all samples of probiotic *Lassi* during the storage period.

Similar trends has been reported by Momin (2009) who carried out the storage study of mushli mixed herbal probiotic *Lassi* and control (without herb) probiotic *Lassi* at 5±2 °C for 21 days. He reported that the acidity for fresh products was 0.61 to 0.62 per cent lactic acid, which increased to 1.03 per cent lactic acid in both cases after 21 days of refrigerated storage and the pH declined slowly during the storage from 4.84 to 3.96 in herbal probiotic *Lassi* and from 4.78 to 3.95 in control *Lassi*.

Shuwu *et al.* (2011) also reported a gradual decline in pH in both regular *Lassi* and herbal honey *Lassi* was 4.53 to 3.91 and 4.57 to 3.89, respectively, during storage at 7±1°C. A significant reduction in pH of the control sample (regular *Lassi*) was observed from the 0th day to the 21st day and then on the 28th day of storage, the pH change was non-significant. In Herbal honey *Lassi*, till the 7th day, the alteration was significant, followed by a non-significant difference on the 14th day. Later, on the 21st day of storage, pH significantly decreased; likewise, on the 28th day, the change in pH was non-significant.

Menon *et al.* (2014) prepared carbonated sweetened fermented dairy drinks stored in glass bottles. pH values for control and carbonated beverages were 4.51 and 4.47, respectively. There was a marginal reduction in the pH value of the carbonated sample reaching 3.83 after 12 weeks of storage.

Table 4.22 pH levels of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	4.40 ±0.03	4.37 ±0.02	4.31 ±0.03	4.25 ±0.02	4.16 ±0.05	4.07 ±0.01	4.26	0.05	0.13*	2.41
LS ₁ (P+SE)	4.39 ±0.04	4.36 ±0.02	4.29 ±0.02	4.24 ±0.03	4.15 ±0.04	4.02 ±0.07	4.24	0.07	0.21*	2.79
LS ₂ (KE+SE)	4.38 ±0.02	4.34 ±0.04	4.29 ±0.00	4.22 ±0.04	4.12 ±0.03	4.01 ±0.02	4.22	0.02	0.07*	1.31
LS ₃ (R+SE)	4.39 ±0.03	4.34 ±0.05	4.30 ±0.04	4.21 ±0.03	4.14 ±0.05	4.05 ±0.03	4.23	0.04	0.11*	2.06
Mean	4.39	4.35	4.29	4.23	4.14	4.03	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance. Each value is the average of five replications.			
S.Em. ±	0.04	0.04	0.03	0.03	0.04	0.08				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	1.79	1.92	1.69	1.83	2.35	4.69				

4.6.8 Titratable acidity of probiotic *Lassi* during storage:

The data in the Table 4.23 showed titratable acidity level range for LS₀, LS₁, LS₂ and LS₃ 0.82-1.09, 0.83-1.17, 0.84-1.22 and 0.83-1.19 per cent, respectively. Titratable acidity level was increased significantly ($p < 0.05$) during the storage period. There was a significant difference due to the addition of different flavours and stevia extract.

Table 4.23 Titratable acidity (%) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
LS ₀ (C)	0.82 ±0.01	0.83 ±0.01	0.85 ±0.01	0.88 ±0.01	0.93 ±0.02	1.09 ±0.02	0.90	0.03	0.07*	4.28
LS ₁ (P+SE)	0.83 ±0.01	0.84 ±0.01	0.85 ±0.06	0.92 ±0.02	0.94 ±0.01	1.17 ±0.04	0.92	0.04	0.13*	8.70
LS ₂ (KE+SE)	0.84 ±0.02	0.84 ±0.02	0.86 ±0.04	0.90 ±0.00	0.93 ±0.03	1.22 ±0.01	0.93	0.03	0.11*	10.80
LS ₃ (R+SE)	0.83 ±0.01	0.85 ±0.01	0.87 ±0.09	0.88 ±0.07	0.97 ±0.00	1.19 ±0.03	0.93	0.05	0.14*	9.68
Mean	0.82	0.83	0.85	0.90	0.94	1.16	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- <i>Kesar Elachi</i> Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance. Each value is the average of five replications.			
S.Em. ±	0.01	0.01	0.04	0.03	0.02	0.02				
CD (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. (%)	3.36	2.54	2.40	3.72	4.16	3.68				

Similar trend followed by Momin (2009) study of the herbal probiotic *Lassi* and control (without herb) was stored at $5 \pm 2^\circ\text{C}$ for 21 days. He found that the acidity for fresh products was 0.61 to 0.62 per cent lactic acid, which increased to 1.03 per cent lactic acid in both subjects after 21 days of refrigerated storage.

Shukla *et al.* (2013) devised a probiotic drink from whey and pineapple juice. They found that the acidity level raised during the refrigerated storage from 0.55 to 0.89 per cent after 28 days and the enhancement in the level of acidity was more evident in the case of storage at an ambient temperature wherein the acidity reached 0.89 per cent after 120 h of storage.

Shinde *et al.* (2015) analysed *Lassi* preparation (0 day), titratable acidity percentage was decreased with the increase in the addition of sapota pulp i.e. 0.76, 0.73, 0.70, 0.68 and 0.65 for *Lassi* prepared under treatments T₁, T₂, T₃, T₄ and T₅, respectively; on the 8th day of storage mean titratable acidity percentage was recorded as 0.96, 0.94, 0.93, 0.92 and 0.91 per cent for *Lassi* prepared under five treatments, respectively.

Manuka *et al.* (2019) studied control *Lassi* and *Lassi* was prepared from LS₂ amalgamation having an acidity of 0.51 per cent lactic acid on 0 day and 0.71, 0.72 per cent lactic acid on the end of shelf life, i.e. on the 12th day. The acidity of LS₁ combination *Lassi* was 0.51 on 0 day and increased to 0.7 per cent LA at the end of the shelf life and the acidity of LS₃ combination *Lassi* was 0.51 and 0.7 per cent, respectively on and end of the shelf life.

4.7 MICROBIAL VIABILITY OF PROBIOTIC LASSI DURING STORAGE:

Through microbial activity, approximately one-fourth of the global food supply is lost (Jespersen *et al.* 1996). Milk is a favourably nourishing food that suits the growth medium for many microorganisms (Rajagopal *et al.* 2015). The definition of probiotics acknowledges the capacity of live microorganisms to exert a beneficial effect on the health, so they are incorporated into a food, and the microbiological control of their cell viability assessment is routinely used to monitor their functionality. For this reason, the scientific criteria (Champagne *et al.* 1997; Ross *et al.* 2005), appear necessary to know, manage and control the microbiological variables that may modify the functionality of a strain in a particular food matrix without having an impact on the number of viable cells. Survival of LAB, development of off-flavour and yeast and mould growth are the essential parameters to estimate keeping quality of fermented milk products (Robinson 2002, Patel and Prajapati, 2010).

4.7.1 Lactobacilli count (log CFU/ml) of *Lassi* during storage:

The data in the Table 4.24 portrayed the log lactobacilli count (CFU/ml) range for LS₀, LS₁, LS₂ and LS₃ as 7.73-7.97, 7.62-7.93, 7.63-7.96 and 7.65-7.96, respectively. On the day of preparation highest score was LS₀ (7.97) and the lowest score was LS₁ (7.93), whereas, on the 11th day of storage, the highest score was observed in LS₀ (7.73) and lowest score LS₁ (7.62). The lactobacilli count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. A non-significant difference was found between the *Lassi* sample during the storage duration. Declined trends in the number of bacteria have seen in various studies. According to Oktavia *et al.* (2016) was reported that low temperature which can diminish the number of microbes that live in food and cause denaturation of cell proteins.

Suliman (1982) reported that the mean count of *Lactobacillus spp.* for yoghurt was $1.29 \times 10^9 \pm 5.4 \times 10^8$ CFU/ml. Karagul-Yuceer *et al.* (2001), studied that the yogurt was stored at 4°C for sensory evaluation by an expert panel on d 7, 21, and 45. A consumer panel evaluated carbonated and noncarbonated yogurts on 21 day. The lactobacilli count non-significantly decreased from 7.84 to 7.55 for the first 21 days of storage.

Shah (2011) prepared synbiotic *Lassi* incorporated with honey and whey drink with inulin and orange juice by using *L. helveticus* MTCC 5463 as probiotic culture in both products and encountered that the count of *L. helveticus* MTCC 5463 remained more than 8 log CFU/ml at the end of storage span of 28 days at refrigerated storage.

Hussain *et al.* (2014) reported that there was no significant decline in the probiotic count of APL during the 12 days of its storage. The viability of *L. paracasei ssp. paracasei* in APL varied from 8.4 ± 0.06 log CFU/ml on the day of its manufacture to 8.0 ± 0.12 CFU/mL on the 12th day of cold storage.

Sudheendra *et al.* (2018) studied the lactobacilli count of both the products and reported the lactobacilli count more than 8.5 log CFU/ g in the fresh products and throughout the storage period (28 days). The initial count was 33 and 43×10^7 (8.52 and 8.64 log CFU / g) in plain probiotic *Lassi* (A) and synbiotic *Lassi* which prepared with carrot juice (B), respectively, was statistically non-significant.

Kaur *et al.* (2019) reported that the mean Lactic Acid Bacteria count (LAB) ranged from 8.53 to 8.68 in fortification of *Lassi* with carrot pulp.

Momin and Prajapati (2019) studied the viable count of lactic acid bacteria after 8 hours of milk fermentation in the presence of medicinal herbs and revealed the direct influence of herbs on lactic cultures. The data revealed an increase in the viable count from the commencement of fermentation until 8 hours of incubation. The log viable count of *Lactobacillus helveticus* MTCC 5463 varies from 7.66 to 8.42 (log CFU/ml).

Table 4.24 Lactobacilli count (log CFU/ml) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S. Em. ±	C.D. (P= 0.05)	C. V. (%)
LS ₀ (C)	7.97 ±0.22	7.90 ± 0.17	7.89 ±0.22	7.81 ±0.22	7.78 ± 0.22	7.73 ± 0.14	7.86	0.17	NS	4.27
LS ₁ (P+SE)	7.93 ±0.19	7.84 ±0.18	7.82 ±0.16	7.79± 0.23	7.71 ±0.21	7.62 ±0.21	7.79	0.21	NS	5.02
LS ₂ (KE+SE)	7.96 ±0.21	7.82 ±0.22	7.80 ±0.18	7.73 ±0.20	7.70 ±0.19	7.63 ±0.18	7.77	0.29	NS	4.18
LS ₃ (R+SE)	7.96 ±0.20	7.83 ±0.20	7.80±0.10	7.77±0.20	7.71 ±0.18	7.65 ±0.17	7.79	0.12	NS	6.92
Mean	7.95	7.85	7.82	7.77	7.72	7.65	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- Kesar Elachi Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, Each value is the average of three replications.			
S.Em. ±	0.21	0.20	0.17	0.22	0.21	0.19				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	5.82	5.64	4.97	6.21	6.09	5.42				

4.7.2 Streptococci count (log CFU/ml) of probiotic *Lassi* during storage:

The data in the Table 4.25 portrayed the log Streptococci count (CFU/ml) range for LS₀, LS₁, LS₂ and LS₃ as 8.73-8.90, 8.68-8.83, 8.67-8.83 and 8.68-8.85 log CFU/ml, respectively. On the day of preparation highest score was LS₀ (8.90 log CFU/ml) and the lowest score was LS₁ and LS₂ (8.83 log CFU/ml), whereas, on the 11th day of storage, the highest score was observed in LS₀ (7.73 log CFU/ml) and

lowest score LS₂ (8.67 log CFU/ml). The Streptococci count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. A non-significant difference was found between the *Lassi* samples during the storage duration. Oktavia *et al.* (2016) reported that low temperature can diminish the number of microbes that live in food and cause denaturation of cell proteins.

Shah and Prajapati (2014) reported that the initial viable count of streptococci (8.94 log CFU/ml) was declined to 8.87 log CFU/ml at the end of 28 days which was non-significant between 0 to 7 and 14 to 21 days but significant (P<0.05) on 7th and 28th days, respectively.

The results are in harmony with Karagul-Yuceer *et al.* (2001), who showed the gradual decrease in viable count of streptococci from 8.65 to 8.58 log CFU/g during the storage period of 21 days.

Table 4.25 Streptococci count (log CFU/ml) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S. Em. ±	C.D. (P= 0.05)	C. V. (%)
LS ₀ (C)	8.90 ±0.11	8.87 ±0.06	8.85 ±0.09	8.80 ±0.07	8.77 ±0.05	8.73 ±0.03	8.82	0.14	NS	6.74
LS ₁ (P+SE)	8.83 ±0.17	8.81 ±0.12	8.76 ±0.11	8.73 ±0.06	8.70 ±0.14	8.68 ±0.03	8.75	0.12	NS	9.87
LS ₂ (KE+SE)	8.83 ±0.22	8.78 ±0.05	8.74 ±0.10	8.70 ±0.10	8.69 ±0.04	8.67 ±0.03	8.74	0.14	NS	8.17
LS ₃ (R+SE)	8.85±0.17	8.83±0.09	8.80±0.16	8.74±0.12	8.70 ±0.07	8.68 ±0.09	8.77	0.19	NS	7.17
Mean	8.85	8.82	8.78	8.74	8.71	8.69	Note: LS: <i>Lassi</i> Sample, C-Control (Without flavour and stevia extract), P-Pineapple Flavour, KE-Kesar Elachi Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, Each value is the average of three replications.			
S.Em. ±	0.13	0.09	0.10	0.08	0.09	0.03				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	3.38	1.65	1.57	1.23	1.24	0.70				

4.7.3 Yeast and Mould count and coliform count (log CFU/ml) of probiotic *Lassi* during storage:

4.7.3.1 Yeast and Mould count:

The data in the Table 4.26 portrayed the log yeast and mould count (CFU/ml) range for LS₀, LS₁, LS₂ and LS₃ as 0.63-1.16, 0.64-1.19, 0.63-1.21 and 0.64-1.18 log CFU/ml, respectively. On the day of preparation highest score was LS₂ and LS₄ (0.64 log CFU/ml) and the lowest score was LS₀ (0.63 log CFU/ml), whereas, on the 11th day of storage, the highest score was observed in LS₂ (1.21 log CFU/ml) and lowest score LS₀ (1.16 log CFU/ml). The yeast and mould count gradually increased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. A non-significant ($p < 0.05$) difference was found between the *Lassi* samples during the storage duration.

Table 4.26 Yeast and Mould count (log CFU/ml) of probiotic *Lassi* during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P=0.05)	C.V. (%)
LS ₀ (C)	0.63 ±0.02	0.74 ±0.01	0.81 ±0.02	0.94 ±0.05	1.07 ±0.02	1.16 ±0.01	0.89	0.04	NS	6.74
LS ₁ (P+SE)	0.64 ±0.01	0.73 ±0.01	0.84 ±0.03	0.95 ±0.02	1.11 ±0.01	1.19 ±0.04	0.91	0.03	NS	9.87
LS ₂ (KE+SE)	0.63 ±0.00	0.76 ±0.02	0.84 ±0.03	1.01 ±0.05	1.10 ±0.01	1.21 ±0.02	0.93	0.04	NS	8.17
LS ₃ (R+SE)	0.64 ±0.01	0.77 ±0.02	0.82 ±0.02	0.98 ±0.01	1.13 ±0.02	1.18 ±0.01	0.92	0.02	NS	7.17
Mean	0.63	0.75	0.82	0.97	1.10	1.18	Note: LS: <i>Lassi</i> Sample, C- Control (Without flavour and stevia extract), P- Pineapple Flavour, KE- Kesar Elachi Flavour, R – Rose Flavour, SE- Stevia Extract (NS)- Non Significant, Each value is the average of three replications.			
S.Em. ±	0.01	0.02	0.02	0.02	0.02	0.01				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	4.15	5.68	4.90	4.29	4.30	2.30				

4.7.3.2 Coliform count:

The coliform count is simple and easy to conduct; hence, it can be used as a hygienic indicator to reflect the general microbiological quality in a routine test. In the present study coliform count was absent. As coliforms can be easily killed by heat, these bacteria can also be used as an indicator of heat treatment failure as well as post-heat treatment contamination (Martin *et al.* 1999).

Momin (2009) also found that the mean yeast and mould count was non-significant in the control and herbal probiotic *Lassi* throughout the storage period of 21 days.

Sonali *et al.* (2016) also observed that the cultured buttermilk prepared by blending *Dahi* and fermented whey in 60:40 was free from coliform during storage at $7\pm 2^{\circ}\text{C}$.

Kadam *et al.* (2018) in the present study *E. coli* was found absent in all fresh as well as stored samples during storage at $9 \pm 1^{\circ}\text{C}$. This indicates that proper hygienic precautions were taken during the production and storage of probiotic *Lassi*.

4.8 SENSORY PROFILE OF PROBIOTIC YOGURT DURING STORAGE:

Based on the acceptability control and stevia extract incorporated in different flavoured yogurt was selected for storage studies. Along with their control samples, yogurt samples were stored at refrigeration temperature ($4\pm 2^{\circ}\text{C}$) in the glass jars. They were analyzed for physio-chemical parameters, sensory scores (i.e. Colour and appearance, Flavour, Consistency, Mouth feel and Overall acceptability) and microbiological (i.e. Lactobacilli count, Streptococci count, yeast and mould count, coliform count). The analysis of selected yogurt samples was stopped when it was found to be unacceptable by the sensory panel.

4.8.1 Colour and appearance score of yogurt during storage:

The data in the Table 4.27 showed colour and appearance score range for YS_0 , YS_1 , YS_2 and YS_3 was 7.33-8.13, 7.53-8.27, 7.60-8.40 and 7.50-8.20, respectively. On the day of preparation highest score was YS_2 (8.40) which was “liked very much”, and the lowest score was YS_0 (8.13), which was “liked very much” on the 11th day of storage. The highest score was observed in YS_2 (7.60) and the lowest score was YS_0

(7.33), which was “liked moderately”. The colour and appearance of all the flavoured probiotic yogurt and control non-significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage.

Table 4.27 Colour and appearance score of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	8.13 ±0.19	8.07 ±0.20	8.00 ±0.21	8.00 ±0.20	7.93 ±0.20	7.33 ±0.21	7.91	0.20	NS	9.77
YS ₁ (K+SE)	8.27 ±0.18	8.13 ±0.19	8.13 ±0.19	8.00 ±0.19	7.87 ±0.21	7.53 ±0.22	7.98	0.20	NS	9.66
YS ₂ (KP+SE)	8.40 ±0.15	8.20 ±0.14	8.07 ±0.15	7.87 ±0.16	7.67 ±0.18	7.60 ±0.19	7.96	0.17	NS	8.14
YS ₃ (R+SE)	8.20 ±0.18	8.00 ±0.16	8.00 ±0.13	7.93 ±0.20	7.73 ±0.15	7.50 ±0.28	7.89	0.18	NS	8.79
Mean	8.25	8.10	8.05	7.95	7.80	7.49	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (Mean ±S.Em), (NS)- Non Significant.			
S.Em. ±	0.18	0.18	0.17	0.19	0.19	0.23				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	8.49	8.37	8.38	9.31	9.53	11.77				

Yadav *et al.* (2007) evaluated the storage changes in probiotic *Dahi* stored at seven °C for up to eight days. On the preparation day, it was 45 out of 50; on day eight, they monitored little changes in appearance, and the score was 42 out of 50.

Askary and Bolandi (2013) studied the fortification of yogurt with iron and reported that the score of colour for all yoghurt specimens also decreased along the storage period on 5 points scale.

4.8.2 Flavour score of probiotic yogurt during storage:

The data in the Table 4.28 showed that the flavour score range for YS₀, YS₁, YS₂ and YS₃ was 5.00-6.87, 5.80-7.80, 5.93-8.20 and 5.67-7.93, respectively. On the

day of preparation highest score was YS₂ (8.33) which was “liked very much”, and the lowest flavour score was YS₀ and YS₁ (8.00), which was “liked very much” on the 11th day of storage, the highest flavour score was observed in YS₂ (7.80) and lowest score YS₀ (6.60) which was “liked slightly”. The flavour score of all the flavoured probiotic yogurt and control significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage.

Table 4.28 Flavour score of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	6.87 ±0.13	6.80 ±0.11	6.73 ±0.15	6.67 ±0.12	6.47 ±0.16	5.00 ±0.11	6.42	0.14	0.38*	7.97
YS ₁ (K+SE)	7.80 ±0.22	7.13 ±0.16	7.00 ±0.19	7.00 ±0.09	6.93 ±0.11	5.80 ±0.17	6.94	0.17	0.47*	9.14
YS ₂ (KP+SE)	8.20 ±0.26	7.40 ±0.16	7.33 ±0.13	7.07 ±0.11	7.00 ±0.14	5.93 ±0.15	7.15	0.17	0.46*	8.84
YS ₃ (R+SE)	7.93 ±0.22	7.20 ±0.10	7.27 ±0.12	7.20 ±0.10	6.87 ±0.13	5.67 ±0.23	7.02	0.16	0.46*	8.82
Mean	7.70	7.13	7.08	6.98	6.81	5.60	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance.			
S.Em. ±	0.22	0.14	0.15	0.11	0.14	0.17				
C.D. (P = 0.05)	0.61*	0.39*	0.43*	0.32*	0.40*	0.49*				
C.V. %	10.90	7.52	8.27	6.25	7.94	10.27				

Yadav *et al.* (2007) evaluated the storage changes in probiotic *Dahi* stored at 7°C for up to 8 days. On the fresh day, the flavour score was 21.6 out of 25. On the eighth day, they glimpsed a slight deterioration of flavour and the score was 19.6 out of 25. However, after eight days of storage, the panel disliked the samples, which notified slight bitterness, which may have been due to proteolysis. They justified the findings by reasoning that during storage, the degradation of β -casein because of the

action of chymosin and plasmin results in the formation of smaller peptides that may be responsible for bitterness in *Dahi*. The reason for the sharp decrease in flavour score of the cultured buttermilk with fermented *Paneer* whey was attributed to the lower intensity of desirable flavour and higher acidity.

4.8.3 Consistency score of probiotic yogurt during storage:

The data in the Table 4.29 showed the consistency score range for YS₀, YS₁, YS₂ and YS₃ was 6.13-8.00, 6.67-8.40, 6.87-8.20 and 6.80-8.13, respectively. On the day of preparation, the highest consistency score was YS₂ (8.40) which was “liked very much” and the lowest consistency score was YS₀ (8.00), which was “liked very much” on the 11th day of storage. The highest score was observed in YS₂ (6.87) and the lowest score was YS₀ (6.13), which was “liked slightly”. The consistency score of all the flavoured probiotic yogurt and control non-significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage.

4.8.4 Mouth feel score of probiotic yogurt during storage:

The data in the Table 4.30 showed mouth feel score range for YS₀, YS₁, YS₂ and YS₃ was 4.83-6.87, 5.00-7.80, 5.87-8.00 and 5.80-7.93 respectively. On the day of preparation highest score was YS₂ (8.00) which was “liked very much” and the lowest score was YS₀ (6.87), which was “liked slightly” on the 11th day of storage. The highest score was observed in YS₂ (5.87) which was “Neither like nor disliked”, and the lowest score was YS₀ (4.83) which was “disliked slightly”. The mouth feel score of all the flavoured probiotic yogurt and control significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage.

Table 4.29 Consistency score of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	8.00 ±0.23	7.87 ±0.21	7.67 ±0.23	7.53 ±0.25	7.27 ±0.20	6.13 ±0.27	7.41	0.21	NS	11.17
YS ₁ (K+SE)	8.40 ±0.16	8.07 ±0.21	7.93 ±0.20	7.87 ±0.19	7.33 ±0.23	6.67 ±0.13	7.71	0.20	NS	10.50
YS ₂ (KP+SE)	8.20 ±0.14	8.00 ±0.21	7.53 ±0.25	7.33 ±0.25	7.13 ±0.21	6.87 ±0.27	7.51	0.24	NS	10.40
YS ₃ (R+SE)	8.13 ±0.17	7.87 ±0.19	7.80 ±0.17	7.80 ±0.17	7.40 ±0.13	6.80 ±0.14	7.63	0.25	NS	12.62
Mean	8.18	7.95	7.73	7.63	7.28	6.61	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em).			
S.Em. ±	0.18	0.21	0.21	0.22	0.20	0.22				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. %	8.59	10.42	10.98	11.24	10.64	12.18				

Table 4.30 Mouth feel score of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	6.87 ±0.22	6.80 ±0.15	6.77 ±0.15	6.73 ±0.18	6.47 ±0.19	4.83 ±0.20	6.41	0.15	0.41*	8.53
YS ₁ (K+SE)	7.80 ±0.13	7.60 ±0.16	7.60 ±0.19	7.40 ±0.19	7.13 ±0.21	5.00 ±0.17	7.08	0.20	0.57*	10.53
YS ₂ (KP+SE)	8.00 ±0.24	7.70 ±0.22	7.60 ±0.13	7.53 ±0.31	7.40 ±0.29	5.87 ±0.24	7.35	0.24	0.67*	12.37
YS ₃ (R+SE)	7.93 ±0.18	7.93 ±0.15	7.87 ±0.14	7.80 ±0.23	7.60 ±0.27	5.80 ±0.15	7.48	0.23	0.64*	11.39
Mean	7.65	7.50	7.46	7.36	7.15	5.37	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em). *Significant at 5% level of significance.			
S.Em. ±	0.18	0.18	0.15	0.25	0.26	0.20				
C.D. (P = 0.05)	0.51*	0.52*	0.43*	0.70*	0.72*	0.57*				
C.V. %	9.10	9.38	7.88	12.91	13.82	11.45				

4.8.5 Overall acceptability score of probiotic yogurt during storage:

The data in the Table 4.31 showed that the overall acceptability score range for YS₀, YS₁, YS₂ and YS₃ was 5.00-7.00, 5.20-8.00, 5.60-8.12 and 5.10-8.07, respectively. On the day of preparation highest score was YS₂ (8.12) which was “liked very much”, and the lowest score was YS₀ (7.00), which was “liked very much”, at the 11th day of storage, the highest score was observed in YS₂ (5.60) and lowest score YS₀ (5.00) which was “liked slightly”. The overall acceptability score of all the flavoured probiotic yogurt and control significantly ($p < 0.05$) decreased throughout the storage period from the day of preparation to the 11th day of storage in all yogurt samples. The satiety value of yogurts was directly correlated to sweetness level. Serving an equal, fixed portion of yogurts was, in many studies, an identical amount a panellist would not choose to eat compared with other sweet foods (Johnson and Vickers, 1993; Vande Water and Vickers, 1996). Liking for chocolate bars and peaches dropped after panellists were served yogurt with a high sweetness level. The reason behind this decrease in liking was the overwhelming sweetness that masked the characteristic yogurt sour taste.

Shah and Prajapati (2014) prepare a carbonated fermented drink and sensory analysis was done, the score was higher for the product with 15 per cent sugar (S₃) followed by 12 per cent (S₂) and 10 per cent (S₁) and mean value of overall acceptability scores were 8.22, 7.52, and 7.11 for S₃, S₂ and S₁, respectively.

Askary and Bolandi (2013) reported that the total score of yogurt fortified with iron was stable up to 14 days, then steadily declined along the storage period. Lutfiye *et al.* (2017) assessed the overall acceptability score ranging from 7.25 ± 1.40 to 8.81 ± 0.37 for buffalo yogurt and from 6.55 ± 0.82 to 8.85 ± 0.21 for mixed milk yogurt.

Table 4.31 Overall acceptability score of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	7.00 ±0.23	6.87 ±0.21	6.80 ±0.22	6.60 ±0.16	6.00 ±0.13	5.00 ±0.16	6.37	0.16	0.45*	9.21
YS ₁ (K+SE)	8.00 ±0.17	7.67 ±0.20	7.53 ±0.21	7.40 ±0.19	6.33 ±0.27	5.20 ±0.16	7.02	0.21	0.58*	10.76
YS ₂ (KP+SE)	8.12 ±0.18	7.87 ±0.21	7.80 ±0.24	7.60 ±0.23	6.33 ±0.31	5.60 ±0.22	7.22	0.19	0.55*	9.25
YS ₃ (R+SE)	8.07 ±0.18	7.93 ±0.20	7.80 ±0.20	7.53 ±0.19	6.13 ±0.21	5.10 ±0.20	7.09	0.21	0.60*	10.82
Mean	7.79	7.58	7.48	7.28	6.19	5.22	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em). *Significant at 5% level of significance.			
S.Em. ±	0.22	0.18	0.22	0.23	0.24	0.15				
C.D. (P=0.05)	0.62*	0.51*	0.63*	0.65*	0.69*	0.43*				
C.V. %	10.98	9.28	11.42	12.24	13.39	8.59				

4.9 PHYSIO-CHEMICAL PROPERTIES OF PROBIOTIC YOGURT DURING STORAGE:

4.9.1 Moisture content of probiotic yogurt during storage:

Table 4.32 showed that the moisture content varies for YS₀, YS₁, YS₂ and YS₃ was 84.04-85.98, 84.46-86.52, 85.24-86.95 and 85.21-86.68 per cent, respectively. On the day of preparation highest score was LS₂ (86.95%) and the lowest score was YS₀ (85.98%), whereas, on the 11th day of storage, the highest score was observed in YS₂ (85.24%) and the lowest score was in YS₀ (84.04%). The data in the Table 4.32 showed that the moisture content of all the samples decreased throughout the storage period from fresh yogurt to the 11th day of storage in all yogurt samples. Similar effects have been observed by Qureshi *et al.* (2011) was reported that decrease in the moisture content of the yogurt from 84.78 to 84.65 per cent during cold storage.

Hamdan *et al.* (1971) and Bills *et al.* (1972) also determined that the moisture contents dropped from 86.90% to 84.95% during refrigerated storage.

Table 4.32 Moisture content (%) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P=0.05)	C.V. (%)
YS ₀ (C)	85.98 ± 0.95	85.56 ±0.72	85.22 ±0.51	85.21 ±0.51	84.80 ±0.17	84.04 ±0.09	85.13	0.51	NS	1.32
YS ₁ (K+SE)	86.52 ± 0.62	86.14 ±0.29	85.81 ±0.33	85.06 ±0.35	85.05 ±0.04	84.46 ±0.17	85.50	0.45	NS	1.18
YS ₂ (KP+SE)	86.95 ±0.63	86.81 ±0.58	86.61 ±0.46	85.53 ±0.38	85.49 ±0.25	85.24 ±0.18	86.10	0.46	NS	1.20
YS ₃ (R+SE)	86.68 ±0.23	85.97 ±0.23	85.64 ±0.18	85.55 ±0.52	85.38 ±0.22	85.21 ±0.24	85.73	0.32	NS	0.82
Mean	86.53	86.12	85.82	85.33	85.18	84.73	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.66	0.50	0.39	0.45	0.19	0.18				
C.V. (%)	1.71	1.30	1.02	1.17	2.50	1.47				

4.9.2 Total Solids content of probiotic yogurt during storage:

The data in Table 4.33 portray that the total solids content varies for YS₀, YS₁, YS₂ and YS₃ was 13.97-15.20, 13.47-14.98, 13.04-14.74 and 13.31-14.74 per cent, respectively. On the day of preparation highest score was LS₀ (13.97%) and the lowest score was YS₁ (13.04%), whereas, on the 11th day of storage, the highest score was observed in YS₀ (15.20%) and the lowest score YS₂ and YS₃ (14.74%). Throughout the storage period, total solids content has been increased non-significantly (p<0.05) in all the samples of flavoured probiotic yogurt along with

control. Total solids content has been increased because of the declining level of moisture in the yogurt samples.

Pagthinathan *et al.* (2018) analyzed the total solids content of probiotic yogurt ranging from 12.80 to 21.87 per cent during four weeks of storage. Muhammad *et al.* (2005) estimated the highest range of total solids in yogurt as 17.10 per cent. The average total solids content of probiotic yogurt was 17.75 per cent, and that of natural yogurt was 19.20 per cent which is more than the present study (Hussain *et al.* 2009).

Table 4.33 Total solids (%) content of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P =0.05)	C.V. (%)
YS ₀ (C)	13.97 ±0.45	14.43 ±0.19	14.77 ±0.16	14.78 ±0.47	15.19 ±0.34	15.20 ±0.07	14.72	0.32	NS	4.48
YS ₁ (K+SE)	13.47 ±0.20	13.85 ±0.49	14.17 ±0.28	14.93 ±0.44	14.94 ±0.27	14.98 ±0.25	14.39	0.35	NS	5.52
YS ₂ (KP+SE)	13.04 ±1.01	13.19 ±0.26	13.38 ±0.55	14.46 ±0.43	14.50 ±0.19	14.74 ±0.02	13.88	0.48	NS	7.75
YS ₃ (R+SE)	13.31 ±0.47	13.72 ±0.44	14.35 ±0.40	14.44 ±0.55	14.60 ±0.08	14.74 ±0.11	14.19	0.38	NS	6.04
Mean	13.44	13.79	14.17	14.65	14.81	14.91	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.61	0.36	0.38	0.47	0.24	0.14				
C.V. (%)	10.17	5.86	5.98	7.23	3.65	2.08				

4.9.3 Protein content of probiotic yogurt during storage:

The data in Table 4.34 showed that the protein content varies for YS₀, YS₁, YS₂ and YS₃ 3.98-4.20, 3.76-4.15, 3.65-4.12 and 3.75-4.10 per cent, respectively. On the day of preparation highest score was LS₀ (3.98%) and the lowest score was YS₂

(3.65%), whereas, on the 11th day of storage, the highest score was observed in YS₀ (4.20%) and the lowest score, YS₃ (4.10%). Protein content was increased non-significantly ($p < 0.05$) during the storage period in all the samples of flavoured probiotic yogurt along with control sample.

Table 4.34 Protein content (%) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P=0.05)	C.V. (%)
YS ₀ (C)	3.98 ±0.20	3.99 ±0.10	4.04 ±0.19	4.05 ±0.20	4.09 ±0.05	4.20 ±0.25	4.05	0.18	NS	8.55
YS ₁ (K+SE)	3.76 ±0.06	3.88 ±0.40	3.96 ±0.15	3.99 ±0.20	4.04 ±0.26	4.15 ±0.17	3.96	0.23	NS	11.45
YS ₂ (KP+SE)	3.65 ±0.03	3.79 ±0.24	4.00 ±0.35	4.05 ±0.19	4.09 ±0.24	4.12 ±0.12	3.98	0.22	NS	11.24
YS ₃ (R+SE)	3.75 ±0.21	3.80 ±0.20	3.98 ±0.20	4.01 ±0.23	4.07 ±0.21	4.10 ±0.21	3.95	0.21	NS	10.31
Mean	3.78	3.86	3.99	4.02	4.07	4.14	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.15	0.26	0.23	0.21	0.21	0.20				
C.V. (%)	8.69	13.73	11.67	9.66	9.72	8.54				

The proteolytic activity of Lactic acid bacteria, which hydrolyses proteins into peptides and amino acids, affects the level of protein. Lactic acid bacteria require a wide range of amino acids for growth and their proteolytic enzyme complement is able to split most types of peptide bonds (Thomas and Mills, 1981). The free amino acid present in yogurt probably resulted from the hydrolysis of protein under the influence of proteolytic enzymes. During the storage period, these free amino acids again unite to form the peptide bonds that transform into protein. Hence the protein contents of yogurt increased during storage.

Ali and Kifah (2021), from the results of the statistical analysis, showed that there were no significant differences ($P \leq 0.05$) in the percentage of protein immediately after manufacturing between all treatments. During storage, the percentage of protein was remarkable higher than before for all treatments after 21 days of manufacturing. The tyrosine value of the product significantly ($P < 0.05$) increased from 2.08 to 2.89 mg/100 g during storage of 28 days. This could be because of the commencement of proteolysis by starters themselves or mediated by enzymatic hydrolysis of specific metabolites (Shah and Prajapati, 2004).

4.9.4 Fat content of probiotic yogurt during storage:

The data in Table 4.35 showed that the fat content varies for YS_0 , YS_1 , YS_2 and YS_3 was 3.70-3.99, 3.92-4.18, 4.03-4.20 and 3.94-4.07 per cent, respectively. On the day of preparation highest score was LS_3 (3.94%) and the lowest score was YS_0 (3.70%), whereas, on the 11th day of storage, the highest score was observed in YS_3 (4.18%) and lowest score YS_0 (4.07%). Fat content was slightly increased during the storage period. There was no significant ($p < 0.05$) difference due to the addition of different artificial flavours and duration of storage. The increase in fat percentage is due to the decrease in the moisture content, which led to an increase in the percentage of total solids, including fat. Mutlu *et al.* (2005) stated that the fat content of bio yogurt made from goat milk varies from 3.1 to 4.5 per cent during storage.

4.9.5 Ash content of probiotic yogurt during storage:

The data in Table 4.36 showed that the ash content varies for YS_0 , YS_1 , YS_2 and YS_3 was 0.84-0.95, 0.83-0.93, 0.80-0.91 and 0.82-0.90 per cent, respectively. On the day of preparation highest score was LS_0 (0.84%) and the lowest score was YS_3 (0.80%), whereas, on the 11th day of storage, the highest score was observed in YS_0 (0.95%) and the lowest score was YS_1 (0.90%). The ash content of flavoured probiotic yogurt increased non-significantly ($p < 0.05$) with the progression of the storage period. This result coincides with the results of Owni and Hamid (2008), who reported increasing ash content during the storage period. The increase in ash content could be attributed to a decrease in moisture (Abdalla and Abdel, 1997).

Table 4.35 Fat content (%) of probiotic Yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	3.70 ±0.10	3.82 ±0.09	3.87 ±0.12	3.87 ±0.13	3.88 ±0.13	3.99 ±0.03	3.87	0.11	NS	6.05
YS ₁ (K+SE)	3.92 ±0.04	3.95 ±0.05	4.01 ±0.26	4.04 ±0.11	4.05 ±0.03	4.18 ±0.02	4.02	0.12	NS	6.51
YS ₂ (KP+SE)	4.03 ±0.05	4.05 ±0.07	4.07 ±0.08	4.13 ±0.07	4.17 ±0.02	4.20 ±0.04	4.11	0.6	NS	3.25
YS ₃ (R+SE)	3.94 ±0.07	3.94 ±0.20	3.97 ±0.11	3.98 ±0.11	4.03 ±0.05	4.07 ±0.04	3.98	0.11	NS	6.07
Mean	3.93	3.94	3.98	4.00	4.03	4.11	Note: YS: Yogurt Sample, C-Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.07	0.12	0.16	0.11	0.07	0.02				
C.V. (%)	3.82	6.80	8.84	6.00	3.70	1.12				

Table 4.36 Ash content (%) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	0.84 ±0.02	0.87 ±0.02	0.89 ±0.04	0.92 ±0.04	0.94 ±0.02	0.95 ±0.01	0.90	0.02	NS	3.39
YS ₁ (K+SE)	0.83 ±0.03	0.85 ±0.11	0.88 ±0.27	0.91 ±0.31	0.92 ±0.02	0.93 ±0.06	0.88	0.02	NS	4.75
YS ₂ (KP+SE)	0.80 ±0.03	0.83 ±0.04	0.85 ±0.01	0.88 ±0.21	0.90 ±0.08	0.91 ±0.12	0.86	0.03	NS	4.55
YS ₃ (R+SE)	0.82 ±0.05	0.83 ±0.01	0.86 ±0.02	0.86 ±0.03	0.89 ±0.05	0.90 ±0.04	0.86	0.01	NS	3.44
Mean	0.82	0.84	0.87	0.89	0.91	0.92	Note: YS: Yogurt Sample, C-Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.02	0.01	0.02	0.02	0.01	0.01				
C.V. (%)	5.97	3.23	4.64	5.11	2.22	2.15				

4.9.6 Carbohydrate content of probiotic yogurt during storage:

Table 4.37 showed that the carbohydrate content varies for YS₀, YS₁, YS₂ and YS₃ was 5.19-5.37, 4.61-4.97, 4.23-4.55 and 4.53-4.79 per cent, respectively. On the day of preparation highest score was LS₀ (5.37%) and the lowest score was YS₂ (4.55%), whereas, on the 11th day of storage, the highest score was observed in YS₀ (5.19%) and the lowest score in YS₂ (4.23%). Carbohydrate content was slightly decreased during the storage period. There was no significant (p<0.05) difference due to the addition of different flavours and storage duration was found.

Tamime and Robinson (2007) observed that lactose contents were significantly (p<0.05) decreased during storage due to the fermentation of lactose. Yilmaz-Ersan *et al.* (2014) analyses and stated a decrease in the proportion of carbohydrates in therapeutic yogurt from 4.42 to 4.07 per cent during the storage period of 25 days, and also agreed with Sadiq (2019) regarding a decrease in the percentage of carbohydrates in yogurt fortified with iron coated when refrigerated storage for 21 days.

Table 4.37 Carbohydrates content (%) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	5.37 ±0.23	5.35 ±0.17	5.31 ±0.40	5.25 ±0.13	5.24 ±0.07	5.19 ±0.29	5.28	0.24	NS	10.08
YS ₁ (K+SE)	4.97 ±0.14	4.82 ±0.27	4.73 ±0.18	4.70 ±0.19	4.68 ±0.17	4.61 ±0.14	4.75	0.19	NS	8.82
YS ₂ (KP+SE)	4.55 ±0.04	4.50 ±0.20	4.46 ±0.21	4.41 ±0.18	4.40 ±0.19	4.23 ±0.12	4.42	0.17	NS	8.48
YS ₃ (R+SE)	4.79 ±0.32	4.74 ±0.16	4.71 ±0.21	4.68 ±0.20	4.67 ±0.18	4.53 ±0.19	4.71	0.22	NS	10.34
Mean	4.96	4.85	4.79	4.76	4.75	4.64	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), Each value is the average of five replications.			
S.Em. ±	0.21	0.20	0.27	0.18	0.16	0.19				
C.V. (%)	9.49	9.32	12.43	8.32	7.48	9.37				

4.9.7 pH level of probiotic yogurt during storage:

Table 4.38 showed that the pH levels varied for YS₀, YS₁, YS₂ and YS₃ were 4.03-4.36, 4.00-4.38, 4.01-4.37 and 3.99-4.37, respectively. On the day of preparation highest score was YS₁ (4.38) and the lowest score was YS₀ (4.36), whereas, on the 11th day of storage, the highest score was observed in YS₀ (4.03) and the lowest score was YS₃ (3.99). There was significant effect (p<0.05) of storage has been observed.

Paz-Díaz *et al.* (2021) reported the similar behaviour of the samples after seven days of storage under refrigeration conditions. The reduction in pH and the increase in the total acidity of yogurts during storage could possibly be explained due to the increased consumption of residual lactose by lactic acid bacteria (Curti *et al.* 2017).

Table 4.38 pH levels of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.E m. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	4.36 ±0.02	4.31 ±0.01	4.29 ±0.01	4.28 ±0.04	4.19 ±0.08	4.03 ±0.03	4.24	0.05	0.15*	2.59
YS ₁ (K+SE)	4.38 ±0.03	4.33 ±0.04	4.30 ±0.09	4.25 ±0.01	4.18 ±0.02	4.00 ±0.01	4.24	0.07	0.20*	3.79
YS ₂ (KP+SE)	4.37 ±0.02	4.30 ±0.02	4.28 ±0.08	4.24 ±0.06	4.17 ±0.02	4.01 ±0.02	4.22	0.04	0.11*	2.17
YS ₃ (R+SE)	4.37 ±0.03	4.31 ±0.02	4.29 ±0.05	4.27 ±0.04	4.16 ±0.06	3.99 ±0.03	4.23	0.04	0.11*	1.94
Mean	4.37	4.31	4.29	4.26	4.17	4.00	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance. Each value is the average of five replications.			
S.Em. ±	0.02	0.04	0.05	0.05	0.04	0.02				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. (%)	1.26	0.95	1.12	1.23	1.09	1.27				

Other studies reported that the pH of yogurt decreased during storage under refrigeration conditions between 3.8 and 4.5 (Olson and Aryana, 2008).

Kaur and Riar, (2020) reported that during the first day of storage, pH of YBG₄ yoghurt sample obtained was 4.0, which further decreased to minimum value of 3.7 after 14 days of storage. The decline in pH during storage might be due to the utilization of residual carbohydrates by viable microorganisms and production of lactic acid as well as due to small amounts of CO₂ and formic acid from lactose (Nikoofar *et al.* 2013). The decrease in pH is due to the microorganism's activity, whereas some researchers demonstrated that the decline in pH during storage period was the result of residual enzymes produced by starters during fermentation (Christopher *et al.* 2009)

4.9.8 Titratable acidity level of probiotic yogurt during storage:

Table 4.39 showed that the titratable acidity varies for YS₀, YS₁, YS₂ and YS₃ was 0.86-1.20, 0.85-1.23, 0.85-1.21 and 0.84-1.26 per cent, respectively. On the day of preparation highest score was LS₀ (0.86%) and the lowest score was YS₃ (0.84%), whereas, on the 11th day of storage, the highest score was observed in YS₃ (1.26%) and the lowest score was found in YS₀ (1.20%). There was significant effect ($p < 0.05$) of storage has been observed on titratable acidity of probiotic yogurt samples.

Pagthinathan *et al.* (2018) reported average acidity of without probiotic yoghurt was 0.65 per cent and 0.4 per cent probiotic added yogurt was 0.67 per cent. The results showed that acidity be inclined to increase in all types of yogurt during storage period.

Kaur and Riar, (2020) whom indicated an increase in the acidity rate in the yogurt treatments from 1.22 per cent for the control treatment to 1.41 per cent for the yogurt treatment containing 2 per cent beta-glucan and there were no significant differences ($P \leq 0.05$).

Table 4.39 Titratable acidity (%) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S.Em. ±	C.D. (P = 0.05)	C.V. (%)
YS ₀ (C)	0.86	0.87	1.00	1.10	1.13	1.20	1.02	0.07	0.20*	3.35
YS ₁ (K+SE)	0.85	0.88	0.98	1.12	1.18	1.23	1.04	0.06	0.16*	4.87
YS ₂ (KP+SE)	0.85	0.90	1.03	1.07	1.19	1.21	1.05	0.04	0.10*	2.94
YS ₃ (R+SE)	0.84	0.89	1.04	1.04	1.17	1.26	1.04	0.05	0.16*	5.32
Mean	0.85	0.88	1.01	1.08	1.16	1.21	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, (Mean ±S.Em), *Significant at 5% level of significance. Each value is the average of five replications.			
S.Em. ±	0.01	0.02	0.03	0.01	0.02	0.01				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	0.03*				
C.V. (%)	3.63	4.20	5.62	2.72	3.02	1.84				

4.10 MICROBIAL VIABILITY OF PROBIOTIC YOGURT DURING STORAGE:

Through microbial activity, approximately one-fourth of the global food supply is lost (Jespersen *et al.* 1996). Milk is a favourably nourishing food that suits the growth medium for many microorganisms (Rajagopal *et al.* 2015). The definition of probiotics acknowledges the capacity of live microorganisms to exert a beneficial effect on the health, so they are incorporated into a food, and the microbiological control of their cell viability assessment is routinely used to monitor their functionality. For this reason, the scientific criteria (Champagne *et al.* 1997; Ross *et al.* 2005), appear necessary to know, manage and control the microbiological variables that may modify the functionality of a strain in a particular food matrix

without having an impact on the number of viable cells. Survival of LAB, development of off-flavour and yeast and mould growth are the essential parameters to estimate keeping quality of fermented milk products (Robinson 2002; Patel and Prajapati 2010).

4.10.1 Lactobacilli count (log CFU/ml) of probiotic yogurt during storage:

The data in the Table 4.40 portrayed the log lactobacilli count (CFU/ml) range for YS₀, YS₁, YS₂ and YS₃ as 8.67-8.49, 8.62-8.39, 8.60-8.40 and 8.62-8.44 log CFU/ml, respectively. On the day of preparation highest score was YS₀ (8.67 log CFU/ml) and the lowest score was YS₂ (8.60 log CFU/ml), whereas, on the 11th day of storage, the highest score was observed in YS₀ (8.49 log CFU/ml) and lowest score YS₁ (8.39 log CFU/ml). The lactobacilli count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all yogurt samples. A non-significant difference was found between the yogurt sample during the storage duration. Oktavia *et al.* (2016) reported that low temperature can diminish the number of microbes that live in food and cause denaturation of cell proteins. That because the growth rate of bacteria depends on the amount of lactic acid produced, and thus on the pH. Therefore, at 28 days of storage, the amount of lactic acid increased and pH decreased that affected to the decrease of total bacteria in yogurt product (Sabbah *et al.* 2009). Sulieman (1982) observed that the average count of *Lactobacillus spp.* for yoghurt was $1.29 \times 10^9 \pm 5.4 \times 10^8$ CFU/ml. Kaur *et al.* (2019) also reported that the mean Lactic Acid Bacteria count (LAB) varied from 8.53 to 8.68 in Lassi incorporated with carrot pulp. Karagul-Yuceer *et al.* (2001) also reported the similar trends that the lactobacilli count non-significantly decreased from 7.84 to 7.55 for the first 21 days of storage.

Momin and Prajapati (2019) studied the viable count of lactic acid bacteria after 8 hours of milk fermentation with the incorporation of medicinal herbs and revealed the direct impacts of herbs on lactic cultures. The log viable count of *Lactobacillus helveticus* MTCC 5463 ranged from 7.66 to 8.42 log CFU/ml.

Shah (2011) developed synbiotic *Lassi* with honey and whey drink with inulin and orange juice using *L. helveticus* MTCC 5463 as probiotic culture in both products and encountered that the count of *L. helveticus* MTCC 5463 stable at a level of more than 8 log CFU/ml at the end of storage span of 4 weeks at refrigerated temperature.

Table 4.40 Lactobacilli count (log CFU/ml) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S. Em. ±	C.D. (P=0.05)	C.V. (%)
YS ₀ (C)	8.67 ±0.02	8.63 ±0.03	8.60 ±0.04	8.55 ±0.02	8.50 ±0.03	8.49 ±0.06	8.57	0.05	NS	5.13
YS ₁ (K+SE)	8.62 ±0.04	8.58 ±0.02	8.54 ±0.03	8.49 ±0.03	8.41 ±0.09	8.39 ±0.03	8.50	0.03	NS	3.13
YS ₂ (KP+SE)	8.60 ±0.04	8.55 ±0.03	8.53 ±0.04	8.48 ±0.04	8.45 ±0.07	8.40 ±0.02	8.50	0.05	NS	2.11
YS ₃ (R+SE)	8.62 ±0.02	8.60 ±0.04	8.56 ±0.08	8.51 ±0.04	8.50 ±0.09	8.44 ±0.05	8.54	0.06	NS	3.54
Mean	8.63	8.59	8.55	8.50	8.46	8.43	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- Kesar Pista Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, Each value is the average of three replications..			
S.Em. ±	0.04	0.03	0.05	0.05	0.08	0.05				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. (%)	1.16	0.83	1.43	1.40	2.08	1.43				

Hussain *et al.* (2014) in the present study, there was no significant decrease in the probiotic viable count of APL during the 12 days of storage duration. The viability of *L. paracasei* ssp. *paracasei* in APL ranged from 8.4 log CFU/ml on the initial day of its manufacture to 8.0 CFU/ml on the 12th day of storage.

Sudheendra *et al.* (2018) studied the lactobacilli count of both the products and reported the lactobacilli count above 8.5 log CFU/ g in the fresh products and throughout the refrigerated storage period (28 days). The initial count was 33 and 43 x 10⁷ (8.52 and 8.64 log CFU / g) in plain probiotic *Lassi* (A) and synbiotic *Lassi* with carrot juice (B), respectively, which is statistically non-significant.

Vassilios *et al.* (2020) the viability of lactic acid bacteria was at acceptable levels after fermentation and remained stable over the 21 days of storage period regardless of fortification.

Previous research has indicated that incorporation of yogurt with chickpea flour increases the growth of probiotic bacteria and maintained higher counts than the control over the three weeks refrigerated storage (Chen *et al.* 2012).

4.10.2 Streptococci count (log CFU/ml) of yogurt during storage:

The data in the Table 4.41 portrayed the log streptococci count (log CFU/ml) range for YS₀, YS₁, YS₂ and YS₃ as 8.70-9.34, 8.66-9.28, 8.62-9.27 and 8.64-9.32 log CFU/ml, respectively. On the day of preparation highest score was YS₀ (9.34 log CFU/ml) and the lowest score was YS₂ (9.27 log CFU/ml), whereas, on the 11th day of storage, the highest score was observed in YS₀ (8.70 log CFU/ml) and lowest score YS₂ (8.62 log CFU/ml). The Streptococci count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all yogurt samples. A non-significant difference was found between the probiotic yogurt samples during the storage duration.

Oktavia *et al.* (2016) reported that low temperature can diminish the number of microbes that live in food and cause denaturation of cell proteins.

Shah and Prajapati (2014) The initial viable count of streptococci (8.94 log CFU/ml) was declined to 8.87 log CFU/ml at the end of 28 days which was non-significant between 0 to 7 and 14 to 21 days but significant (P<0.05) on 7th and 28th days, respectively.

Table 4.41 Streptococci count (log CFU/ml) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S. Em. ±	C.D. (P= 0.05)	C.V. (%)
YS ₀ (C)	9.34 ±0.12	9.13 ±0.20	9.07 ±0.09	8.87 ±0.04	8.77 ±0.01	8.70 ±0.05	8.98	0.09	NS	5.47
YS ₁ (K+SE)	9.28 ±0.17	9.20 ±0.26	9.02 ±0.11	8.82 ±0.5	8.70 ±0.08	8.66 ±0.03	8.95	0.13	NS	3.15
YS ₂ (KP+SE)	9.27 ±0.18	9.21 ±0.19	8.79 ±0.12	8.79 ±0.01	8.69 ±0.04	8.62 ±0.05	8.89	0.11	NS	3.99
YS ₃ (R+SE)	9.32 ±0.31	9.28 ±0.24	8.80 ±0.18	8.80 ±0.02	8.70 ±0.01	8.64 ±0.09	8.92	0.18	NS	4.19
Mean	9.30	9.20	8.92	8.83	8.71	8.65	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, Each value is the average of three replications..			
S.Em. ±	0.25	0.24	0.15	0.03	0.02	0.07				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. (%)	6.03	5.92	3.74	0.69	0.61	1.92				

The results are in harmony with Karagul-Yuceer *et al.* (2001), who showed the gradual decrease in viable count of streptococci from 8.65 to 8.58 log CFU/g during the storage period of 21 days.

Ashraf *et al.* (2011) studied the streptococcus spp. count was found to be high in mixed set yoghurt ($2.74 \times 10^9 \pm 15.13 \times 10^9$ CFU/ml) compared to whole set yoghurt ($1.50 \times 10^9 \pm 52.87 \times 10^8$ CFU/ml). Likewise Lactobacillus spp. count was observed to be high in mixed set yoghurt ($3.35 \times 10^9 \pm 16.15 \times 10^9$ CFU/ml) compared to whole set yoghurt ($1.08 \times 10^9 \pm 31.55 \times 10^8$ CFU/ml). Moreover Streptococcus spp., showed more growth compared with Lactobacillus spp throughout the storage period.

4.10.3 Yeast and Mould count and coliform (log CFU/ml) of yogurt during storage:

4.10. 3.1 Yeast and Mould count:

Table 4.42 Yeast and Mould count (log CFU/ml) of probiotic yogurt during storage

Sample No.	Fresh	3 rd day	5 th day	7 th day	9 th day	11 th day	Mean	S. Em. ±	C.D. (P= 0.05)	C.V. (%)
YS ₀ (C)	0.68 ±0.02	0.75 ±0.03	0.86 ±0.00	0.95 ±0.02	1.12 ±0.01	1.18 ±0.02	0.92	0.05	NS	5.13
YS ₁ (K+SE)	0.70 ±0.02	0.78 ±0.01	0.87 ±0.01	0.99 ±0.01	1.15 ±0.03	1.25 ±0.01	0.95	0.03	NS	3.13
YS ₂ (KP+SE)	0.67 ±0.03	0.77 ±0.04	0.88 ±0.01	1.01 ±0.05	1.17 ±0.01	1.27 ±0.01	0.96	0.05	NS	2.11
YS ₃ (R+SE)	0.69 ±0.04	0.78 ±0.02	0.89 ±0.01	1.04 ±0.01	1.18 ±0.01	1.23 ±0.04	0.96	0.06	NS	3.54
Mean	0.68	0.77	0.87	0.99	1.15	1.23	Note: YS: Yogurt Sample, C- Control (Without flavour and stevia extract), K – Kevda Flavour, KP- <i>Kesar Pista</i> Flavour, R- Raspberry Flavour, SE- Stevia Extract, (NS)- Non Significant, Each value is the average of three replications..			
S.Em. ±	0.03	0.02	0.01	0.02	0.02	0.02				
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS				
C.V. (%)	8.66	7.16	3.11	4.91	3.52	3.95				

The data in the Table 4.42 portrayed the log yeast and mould count (log CFU/ml) range for YS₀, YS₁, YS₂ and YS₃ as 0.68-1.18, 0.70-1.25, 0.67-1.27 and 0.69-1.23 log CFU/ml, respectively. On the day of preparation highest score was YS₁ (0.70 log CFU/ml) and the lowest score was YS₀ (0.68 log CFU/ml), whereas, on the 11th day of storage, the highest score was observed in YS₂ (1.27) and lowest score YS₀ (1.18). The yeast and mould count gradually increased throughout the storage period from the day of preparation to the 11th day of storage in all probiotic yogurt samples. A non-significant difference was found between the *Lassi* samples during the storage duration.

Similar results found by Momin (2009) that the mean yeast and mold count was non-significant in control and herbal probiotic *Lassi* throughout storage period up to 21 days.

Vahedi *et al.* 2008, who also reported absence of yeast and mold count in the yogurt drink added with apple.

4.10.3.2 Coliform count

Coliform count is simple and easy to conduct; hence, it can be used as a hygienic indicator to reflect the general microbiological quality in routine test. Coliform count was absent in all the probiotic yogurt samples. As coliforms can be easily killed by heat, these bacteria can also be used as an indicator of heat treatment failure as well as post heat treatment contamination.

Parekh *et al.* (2016) observed that the cultured buttermilk prepared by blending *Dahi* and fermented whey in 60:40 was free from coliform during storage at 7±2°C.

Kadam *et al.* (2018) in the present study *E. coli* was found absent in all fresh as well as stored samples during storage at 9 ±1°C. This indicates that proper hygienic precautions were taken during the production and storage of probiotic kokum *Lassi*.

In the present study coliform count was found absent in all fresh as well as stored samples during storage at 4±1°C. This indicates that proper hygienic precautions were taken during the production and storage of probiotic yogurt.

4.11 GENERAL INFORMATION OF PARTICIPANTS:

In this section general information of the participants regarding their age, education level, occupation, family type, marital status, sleep time, food habits. Detail information of these discussed as:

4.11.1 Age of participants:

Table 4.43 shows that in the control group majority of the participants (56.66%) fell in the age range of 41-45 years, followed by the age range 36-40 (30%) and 29-35 years (13.33%). Similarly, in the experiment group majority of the participants (53.33) falling in the age range of 41-45 years, followed by the age range 36-40 (30%) and 29-35 years (16.67%). Data from the table clearly shows that as age increase the risk of diabetes also increases.

**Table 4.43 Age of participants
(Control Group n= 30 and Experiment Group n=30)**

Age Group	Control Group		Experiment Group	
	Frequency	Percent	Frequency	Per cent
29-35 Years	4	13.33	5	16.67
36-40 Years	9	30.00	9	30.00
41-45 Years	17	56.66	16	53.33

There is an inverse relationship between potential destruction from diabetes and age at diagnosis.

At younger age, when it happens, the more significant the possible harm. The national urban diabetes survey in India showed that more than 50% of diabetic patients had onset at less than 50 years of age (WHO, 2014). The results shown by Bamji (2003) and Deepashree and Prakash (2007) stated that the prevalence of type II diabetes mellitus rose significantly above the age of 35. The DECODE-DECODA Study Group (2003) reported that Asian Indians develop diabetes at a younger age, at least 10-15 years before than the Caucasian population (Ramachandran, 2004).

4.11.2 Education level of participants:

A glance at the Table 4.44 depicted that in the control group out of the total participants 20 per cent was graduated and 20 per cent got senior secondary education followed by 16.67 per cent got middle level education, 13.33 per cent got secondary level education, 10 per cent got primary education, 10 per cent was literate and among those 6.67 per cent got technical education. Whereas, in the experiment group majority of candidate (33.33%) was graduated followed by (16.67%) secondary level, (13.33%) primary and middle level, (10%) senior secondary education and (6.67%) literate. Among those 3.33 per cent participants was post graduated and got technical education.

Education acts as a catalyst for social up-liftment, improving the returns on investments for almost all aspects of developmental efforts and health. Study conducted by Talukder and Hossain (2020) reported a statistical association of education among the participants with diabetes, with 62 per cent more prevalence of type II diabetes mellitus for highly educated participants, 63 per cent higher for individuals with hypertension, 42 per cent higher for wealthy people, and double the incidental of developing diabetes among overweight participants has been reported. However, the individuals engaged in physical labour had less chance of having type II diabetes mellitus.

**Table 4.44 Education level of participants
(Control Group n= 30 and Experiment Group n=30)**

Education level	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Literate	3	10.00	2	6.67
Primary Education	3	10.00	4	13.33
Middle Education	5	16.67	4	13.33
Secondary Education	4	13.33	5	16.67
Senior Secondary Education	6	20.00	3	10.00
Graduate	6	20.00	10	33.33
Post Graduate	0	0.00	1	3.33
Technical Degree/ Diploma	2	6.67	1	3.33

4.11.3 Occupation of participants:

It shows from the Table 4.45 that most of participants from the control group (40%) were doing their own business followed by (23.33%) farming, (20.00%) Service (Private) and (16.67%) Service (Government). While in the experiment group majority (33.33%) were business, 26.67 Service (Government), (23.33%) Service (Private) and (16.67%) farming. Most of the participants were working from sedentary to moderate work type. Sedentary behaviour has recently been known as a significant public health issue. Several epidemiologic studies have suggested a relationship between sedentary behaviour and diabetes (Biswas *et al.* 2015 and Owen *et al.* 2010).

In many occupations, people spend approximately half of their workday in a seated position (Jans *et al.* 2007 and Miller *et al.* 2004) and spend hours of leisure time on activities such as watching TV, using computers, participating in screen-based amusement and driving. Although the mechanisms underlying the relationship between sedentary occupations and diabetes remains unclear, the alternate of physical activity by sedentary behaviour, thus declining energy expenditure, may attribute to the positive association between sitting time and diabetes (Mummery *et al.* 2005).

**Table 4.45 Occupation of participants
(Control Group n= 30 and Experiment Group n=30)**

Service Name	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Service (Private)	6	20.00	7	23.33
Service (Government)	5	16.67	8	26.67
Business	12	40.00	10	33.33
Farming	7	23.33	5	16.67

4.11.4 Type of family of participants:

The data shows in Table 4.46 from the present findings that majority of participants from the control group (60%) have nuclear family and (40%) have joint family. Correspondingly, in the experiment group most of the participants (56.67%) have nuclear family and (43.33%) have joint family. The value in the Table 4.11 also

shows the number of family members in the control group and experiments group. Majority of the participants (46.67%) have 6-8 family members followed by (43.33%) have 3-5 family member and remaining (10%) have more than 8 members family. While in the experiment group Majority of the participants (50%) have 6-8 family members followed by (36.67%) have 3-5 family member and remaining (13.33%) have more than eight members family.

Table 4.46 Type of Family of participants
(Control Group n= 30 and Experiment Group n=30)

Type of Family	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Joint	12	40.00	13	43.33
Nuclear	18	60.00	17	56.67
No. of family Member Control	Frequency	Per cent	Frequency	Per cent
3-5 member	13	43.33	11	36.67
6-8 member	14	46.67	15	50.00
more then 8	3	10.00	4	13.33

4.11.5 Marital status of participants:

The values shows in Table 4.47 from the present research that majority of participants from the control group majority (96.67%) of candidate were married and (3.33%) were divorced. However, in the experiment group most of the participants (86.67%) were married followed by (6.67%) was widower and (3.33%) was unmarried and equal per cent was divorced.

Table 4.47 Marital status of participants
(Control Group n= 30 and Experiment Group n=30)

Marital Status	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Unmarried	0	0.00	1	3.33
married	29	96.67	26	86.67
Divorced	1	3.33	1	3.33
Widower	0	0.00	2	6.67

4.11.6 Sleep time of participants:

Data in the Table 4.48 present the sleep time of the both of groups. Among the control group 40 per cent was sleeping for 6-8 hours, 36.67 per cent participants was sleeping for 5-6 hours, 13.33 per cent was sleeping for 8-10 hours and 10 per cent was sleeping for less than 5 hours. Whereas in the experiment group 46.67 per cent was sleeping for 5-6 hours, 30 per cent was sleeping 6-8 hours, 16.67 per cent was sleeping for 8-10 hours and 6.67 per cent was sleeping > 5 hours. Data clearly shows that majority of participant have alarmed sleeping pattern.

Sleep helps to maintain many important functions. One of the most significant of these functions may be to provide cells and tissues with to recover from the wear and tear of daily life. Determining the risks posed by inadequate sleep is complex. However, Taveras (2008) have shown that people who reported sleeping less than five hours per night had a greatly amplified risk of having or developing type II diabetes.

Spiegel *et al.* (1999) reported that sleep deprivation has been shown to be one of leading risk factor for impaired glucose tolerance (IGT). Various studies have done to explore the overall shape of the relationship between sleep duration and risk of type II diabetes (Holliday *et al.* 2013; Boyko *et al.* 2013; Von *et al.* 2012; Kita *et al.* 2012; Xu *et al.* 2010; Tuomilehto *et al.* 2009; Gangwisch *et al.* 2007; Yaggi *et al.* 2003). A U-shaped association was observed, with the lowest risk of type II diabetes at sleep duration of 7-8 hours per day. Nilsson (2004) studies have also found that improved sleep can positively influence blood sugar control and reduce the effects of type 2 diabetes. A number of studies have reported that habitual sleep disturbances are associated with risk of developing type II diabetes (Cappuccio *et al.* 2010).

**Table 4.48 Sleep time of participants
(Control Group n= 30 and Experiment Group n=30)**

Timings of sleep (Hours)	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
less than 5 hours	3	10.00	2	6.67
5-6 hours	11	36.67	14	46.67
6-8 hours	12	40.00	9	30.00
8-10 hours	4	13.33	5	16.67

4.12 ANTHROPOMETRIC MEASUREMENT OF PARTICIPANTS:

4.12.1 Height of participants:

It is evident from the value given in Table 4.49 that height of the control group majority (60.00 per cent) of participants were having height between 160 to 170 cm, (35.33 per cent) followed by (23.33%) were having the height from 150-160 cm, (13.33%) having the height from 171-180 cm rest of participants (3.33%) having the height more than 180 cm. Whereas in the experiment group less than half (40%) of the participants were having the from 160-170 cm, followed by (30%) who have height range from 171-180 cm, (20%) who have height range from 171-180 cm remaining (10%) were having height <180 cm. Data clearly shows that majority of participants have less height than average height proposed by ICMR.

Tall physique is associated with reduction of risk for developing type II diabetes, with each 10 cm increase in height linked with a 41 per cent decreased risk for diabetes among men and a 33 per cent decreased risk among women, according to findings published in *Diabetologia* (<https://www.healthline.com>, <https://diabetologia-journal.org>)

**Table 4.49 Height of participants
(Control Group n= 30 and Experiment Group n=30)**

Height (cm)	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
150-160 cm	7	23.33	6	20
160-170 cm	18	60.00	12	40
171-180 cm	4	13.33	9	30
More than 180 cm	1	03.33	3	10
Mean	165.11 cm		168.27cm	

4.12.2 Weight of participants:

The data given in Table 4.50 revealed that 30 per cent participants were come under the weight category of 60-70 kg and 81-90 kg, 23.33 per cent were come under

the weight category of 71-80 kg, 13.33 per cent were come under the weight category of 91-100 kg and 3.33 per cent were come under the weight category of more than 100 kg, respectively in the control group. However, in the experiment group, majority of the 43.33 per cent come under the weight category of 71-80 kg followed by 26.66 per cent come under the weight category of 60-70 kg, 16.66 per cent, 10 per cent come under the weight category of 91-100 kg 3.33 per cent come under the weight category of more than 100 kg, respectively. Data shows clearly that most of the participants in the both group was overweight to obese.

Obesity has been consistently considered as major risk factor for type II diabetes mellitus, as obesity was hypothesized to induce insulin resistance (IR) and β -cell failure (Eckel *et al.* 2011). A strong association between overweight and the increased prevalence of diabetes was noted (Ramachandran *et al.* 2003) Overweight, in turn, was correlated with a lack of physical activity and with a high socioeconomic background (Ehtisham *et al.* 2000). Obesity leads to enlarged lipid storage in adipose tissues, which at a certain point no longer have the capability to store excess energy intake. At this point, adipose tissues release free fatty acids (FFAs) by increased lipolysis, which can remains in the circulation (Frayn *et al.* 2002) and result in allover raise circulating level of FFAs (Shulman *et al.* 2013, Boden, 2008), which in turn advances muscle and hepatic insulin resistance (IR) as well as impaired insulin secretion from the β -cell in the pancreas (DeFronzo *et al.* 2014, Shulman *et al.* 2013). Eun *et al.* (2018) aimed to investigate the effect of weight alteration on the incidence of type II diabetes in Korea. They concluded that individuals who developed type II diabetes were more likely to be older and male, to have high body mass index (BMI), fasting blood glucose and blood pressure. Compared with the consistently non-obese group, there was a high risk ratio for incident diabetes in subjects becoming obese and remaining obese later than adjustment for confounding factors. Decreased BMI was significantly linked with lower risks for incident diabetes and the trends were more evident in the non-obese group. However, overall there was no significant association of increased BMI with incident diabetes. In conclusion, weight loss was significantly associated with lower risk for diabetes both in non-obese and obese Koreans, but particularly in the non-obese.

Table 4.50 Weight of participants
(Control Group n= 30 and Experiment Group n=30)

Weight (kg)	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
60-70 kg	9	30	8	26.66
71-80 kg	7	23.33	13	43.33
81-90 kg	9	30	5	16.66
91-100 kg	4	13.33	3	10
more than 100 kg	1	3.33	1	3.33
Mean	79.05 kg		77.33 kg	

4.12.3 BMI of participants:

Data in the Table 51 presented the BMI of both groups. Among the control group most of the participants (40%) have BMI <30 kg/m² followed by (36.66%) who have BMI 18.5-24.9 kg/m² and (23.33%) have BMI range from 25-29.9 kg/m². Whereas, among the experiment group (40%) of the candidate have BMI range from 25-29.9 kg/m² and (30%) participants have BMI 18.5-24.9 kg/m² and <30 kg/m² by each group.

Being overweight or obese is strongly linked to diabetes. Despite the global voluntary target to halt the rise in obesity by 2025 (WHO, 2013 and WHO, 2015), being overweight or obese has increased in almost all countries. In 2014, the latest year for which global estimates are available, more than one in three adults aged over 18 years were overweight and more than one in 10 were obese. Mokdad *et al.* (2003) also found statistically significant for Type II diabetes mellitus among overweight adults, with BMI between 30 and 39.9 kg/m² and adults with BMI ≥ 40 kg/m² relative to adults with normal BMI.

Asian Indians generally have a lower BMI than many other ethnic groups, but the association between BMI and glucose intolerance is as strong as in any other population (Ramachandran *et al.* 2001). The risk of diabetes was significant for urban Indian populations with a BMI of >23 kg/m² (Snehalatha *et al.* 2003). This has been confirmed by studies from other parts of India, (Mishra *et al.* 2001) as well as in

studies of migrant Indians (Chandalia *et al.* 1999) and in other Asian populations (WHO, 2000). According to WHO recommendations, a BMI of 18.5-22 kg / m² is considered healthy for Asian populations

**Table 51 BMI of participants
(Control Group n= 30 and Experiment Group n=30)**

kg/m ²	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
less than 18.5	0	0	0	0
18.5-24.9	11	36.66	9	30
25-29.9	7	23.33	12	40
greater then 30	12	40	9	30
Mean	28.93 kg/m²		27.38 kg/m²	

4.13 DAIBETES MELLITUS AND PHYSICAL ACTIVITIES ROUTINE RELATED INFORMATION:

4.13.1 Diabetic History of participants:

Data shows from the Table 4.52 shows the presence of diabetic history among both groups' participants. In the control group majority (56.67%) of the participants have positive diabetes mellitus history; whereas, remaining (43.33%) participants have negative diabetes mellitus history. Similarly, in the experiment group majority (63.33%) of the participants and remaining (36.67%) participants have negative diabetes mellitus history. The same Table 4.13 (I) also shows the duration of diagnosed diabetes mellitus, In the control group less than half (40%) have diabetes from 4-5 years followed by (26.67%) have diabetes from 5-6 years, (23.33%) have diabetes from 2-3 years, (6.67%) have diabetes from 0-1 year remaining (3.33%) have diabetes from <6 years. However, in the experiment group one third of the participants (33.33%) have diabetes from 2-3 year followed by (30%) of participants have diabetes from 4-5 year and 5-6 year by each group and remaining (6.67%) were have diabetes from <6 years.

Family members share genes, behaviors, lifestyles and environments that together may influence their health and their risk of chronic disease. Most of people

have a family health history of some chronic diseases (*e.g.*, cancer, coronary heart disease and diabetes) and health conditions (*e.g.*, high blood pressure and hypercholesterolemia). People who have a close family member with a chronic disease may have a higher risk of developing that disease than those without such a family member.

Type II diabetes has a stronger link to family history and lineage than type I, and studies of twins have shown that genetics play a very strong role in the development of type II diabetes (<https://diabetes.org/diabetes/genetics-diabetes>). These findings also support previous studies demonstrating that family history is a strong and independent risk factor for diabetes. (Harrison *et al.* 2003, O’Rahilly *et al.* 2005, Newell *et al.* 2004, Arslanian *et al.* 2005, Annis *et al.* 2005). Another studies have also shown that a family history of diabetes is associated with an increased risk of developing type II diabetes (InterAct, 2013, Wagner, 2013, Lyssenko, 2008).

MMWR (2004) recent surveys indicate that the majority of United States adults are aware of and can report Type II diabetes mellitus (DM) among first-degree relatives (88%-95%, depending on type of relative) and, to a lesser extent, second-degree relatives (70%-77%, depending on type of relative).

Lundgren *et al.* (2010) a positive family history of type I diabetes increases the risk to develop type II diabetes, and is associated with earlier onset of type II diabetes.

Pozzilli *et al.* (1991) patients with newly diagnosed type I diabetes have been observed to enter clinical remission more often if they have relatives with type II diabetes compared with those without type II diabetes in the family.

Ali (2013) Family and twin studies have indicated 20–80 per cent of inheritability of diabetes. First-degree relatives of individuals with Type II diabetes mellitus (DM) were three times more likely to develop the disease than individuals without a positive family history.

Anand *et al.* (2020) reported that family history of diabetes is a important risk factor for the development of type II diabetes. In study 41 out of 100 patients have positive hereditary diabetes.

Raikar *et al.* (2018) in urban slums of Mumbai in 27.40 per cent were having positive family history.

**Table 4.52 Diabetes mellitus history of participants
(Control Group n= 30 and Experiment Group n=30)**

Diabetes mellitus History	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Positive	17	56.67	19	63.33
Negative	13	43.33	11	36.67
Duration of diagnosed diabetes mellitus				
0-1 year	2	6.67	0	0.00
2-3 year	7	23.33	10	33.33
4-5 year	12	40.00	9	30.00
5-6 year	8	26.67	9	30.00
More than 6 year	1	3.33	2	6.67

4.13.2 Symptom of diabetes mellitus:

Table 4.53 depicted the symptoms of diabetes mellitus among the both group. In the control majority of the participants have (86.66%) polyphagia, followed by (76.66%) have polyuria, (73%) have dry mouth, (63%) suffering from general weakness, (60%) have polydypsia and pain in legs, (40%) observed for weakness of eyes, (37%) suffering from delayed wound healing and (30%) were reported having weight loss. However, in the experiment group majority of the participants (73.33%) have polyphagia followed by (70%) suffering from pain in legs and general weakness, (63%) reported for dry mouth, Polyuria and polydypsia, (47%) suffering from weakness of eyes, (30%) suffering from delayed wound healing and merely (16.67%) was observed for losing weight.

According to the patients' perception, majority of the respondents (Type II DM) were suffering from visual impairment (66%) followed by joint pains (54%), fatigue (50%), increased frequency of urination (47%), polyphagia (29%), and loss of appetite (24%) despite being on treatment. (Senadheera *et al.* 2016).

**Table 4.53 Symptoms of diabetes mellitus of participants
(Control Group n= 30 and Experiment Group n=30)**

Symptoms	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Polyuria				
Present	23	76.66	19	63
Absent	07	23.33	11	37
Polyphagia				
Present	26	86.66	22	73.33
Absent	4	13.33	8	26.67
Polydypsia				
Present	18	60	19	63
Absent	12	40	11	37
General weakness				
Present	19	63	21	70
Absent	11	37	9	30
Weakness of eyes				
Present	12	40	14	47
Absent	18	60	16	53
Delayed wound healing				
Present	11	37	9	30
Absent	19	63	21	70
Weight Loss				
Present	9	30	5	16.67
Absent	21	70	25	83.33
Pain in legs				
Present	18	60	21	70
Absent	12	40	9	30
Dry mouth				
Present	22	73	19	63
Absent	12	40	11	37

Ramachandran (2014) reported the classic symptoms of diabetes such as polydipsia, polyuria and polyphagia occur frequently in type I diabetes, which has a hasty development of severe hyperglycaemia and also in type II diabetes with very high levels of hyperglycaemia. Severe weight loss is general only in type I diabetes or if type II diabetes remains undetected for an extensive duration. Unexplained weight reduction, restlessness, weakness and body pain are also common signs of undetected diabetes. Symptoms that are mild or have steady development could also remain unnoticed.

Anand *et al.* (2020) reported in the study most common complications were ophthalmic 48 per cent like retinopathy, cataract and blurry vision and 22 per cent have heart related complications and 22 per cent have other complications like tingling in hand and feet.

4.13.3 Exercise timings and frequency followed by participants:

The data in the Table 4.54 shows that in the control group only 30 per cent were doing exercise whereas 70 per cent were not involved in the physical activities. Participants who were doing exercise majority of them (16.67%) were walking followed by (3.33%) jogging, cycling, sports and gym. The Table 4.54 also shows frequency of exercise performed by participants. Among the participants of control group (10%) were doing daily and on weekend followed by (6.67%) were doing exercise twice a week and (3.33%) were doing exercise thrice a week. However, in the experiment group 37 per cent were doing exercise while 63 per cent were not involved in the physical activities. Participants who were involved in the physical activities (20%) were doing walking, (6.67%) were doing jogging, and (3.33%) were doing cycling, climbing stairs and gym. The table shows frequency of exercise performed by participants of experiment group also. Among all the participants (13.33%) were doing daily followed by (10%) were doing exercise four times a week, (6.67%) were doing exercise twice a week and on weekends and (3.33%) were doing exercise thrice a week. Data from the table evidently illustrate that a small number of participants were involved in the physical activities.

Various studies reported that participation in regular physical activity improves blood glucose level and can prevent or deferment onset of type II diabetes (Duncan *et al.* 2003; Hamman *et al.* 2006; Knowler *et al.* 2002; Laaksonen *et al.* 2005; Li *et al.* 2008; Tuomilehto *et al.* 2001).

U.S. Department of Health and Human Services Centers for Disease Control and Prevention (2000) reported that patient with type II diabetes can achieve blood glucose manage by following a nutritious meal plan and exercise program, losing excess weight, and taking oral medications, although others may required supplementation of insulin.

A study by Hahn *et al.* (2009) on subjects aged 55-74 years found that regular exercise at least ≤ 1 hour per week reduced the risk of metabolic syndrome. Sports activities > 2 hours per week would be effective in lowering the risk of metabolic syndrome.

**Table 4.54 Exercise timings and frequency followed by participants
(Control Group n= 30 and Experiment Group n=30)**

Exercise	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Yes	9	30	11	37
No	21	70	19	63
Types of Exercise				
Walking	5	16.67	6	20
Jogging	1	3.33	2	6.67
Cycling	1	3.33	1	3.33
Climbing stairs	0		1	3.33
Sports	1	3.33	0	0
Gym	1	3.33	1	3.33
Frequency of exercise				
daily	3	10.0	4	13.33
twice a week	2	6.67	2	6.67
Thrice a week	1	3.3	1	3.33
Four times a week	0	0	3	10.00
on weekends	3	10	1	3.33

Yates *et al.* (2012) Physical activity interventions can improve glucose tolerance and reduce the risk of T2DM, because it simply helps achieve weight loss (Telford, 2007). Regular exercise may prevent or delay type II diabetes developments (Schellenberg *et al.* 2013). Yardley *et al.* (2014) Regular exercise also has

considerable health benefits for people with type I diabetes (e.g., improved cardiovascular fitness, muscle strength and insulin sensitivity).

4.13.4 Treatment taken for Diabetes mellitus by participants:

Table 4.55 illustrate that treatment taken for diabetes mellitus by participants, In the control group, majority of the candidate (93.33%) were taking treatment through oral drugs with dietary modification whereas in the experiment group all of the candidate (100%) were taking treatment through oral drugs with dietary modification.

**Table 4.55 Treatment taken for diabetes mellitus by participants
(Control Group n= 30 and Experiment Group n=30)**

Treatment taken	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Dietary modification only	2	6.67	0	0.0
Oral drugs with Dietary Modification	28	93.33	30	100.0
Insulin	0	0.00	0	0.0
Type of Medication				
Allopathic only	26	86.67	28	93.33
Homeopathic only	0	0.00	0	0.00
Ayurvedic only	0	0.00	0	0.00
Allopathic + Homeopathic	4	13.33	2	6.67

Ali *et al.* (2013) reported that lifestyle modification such as dietary modification and increased physical activity can be very efficient in improving glycemic control, over the long-term most patients with Type II diabetes (T2DM) will require medications to attain and keep up glycemic control. A meta-analysis thirty five trials by Hirst *et al.* (2012) recommended substantiation in support of efficiency of metformin therapy in lowering of HbA1c when used as monotherapy or in combination.

4.14 DIETARY SURVEY OF PARTICIPANTS:

4.14.1 Food habits of participants:

The values in the Table 56 depicted that more than one third (36.67%) participants from the control group was ovo-vegetarian followed by (33.33%) were vegetarian and (30%) non vegetarian whereas in the experiment group majority of the participants (53.33%) were non vegetarian followed by (26.67%) were vegetarian and (20%) ovo-vegetarian. None of the participants among the both group were vegan. Data illustrated clearly that greater part of the participants were non vegetarian and ovo-vegetarian.

Various data have also directed toward the regular consumption of meat is a menace factor for type II diabetes (Barnard *et al.* 2014 and Pan *et al.* 2011). Slavíček *et al.* (2008) reported in a cross-sectional study that those who adhered to a lacto-ovo-vegetarian diet had significantly decreased CVD risk factors, specifically blood pressure, high serum cholesterol levels and blood glucose levels than those adhering to an omnivorous diet.

**Table 4.56 Food habits of participants
(Control Group n= 30 and Experiment Group n=30)**

Food Habits	Control Group		Experiment Group	
	Frequency	Per cent	Frequency	Per cent
Vegetarian	10	33.33	8	26.67
Non vegetarian	9	30.00	16	53.33
Ovo-vegetarian	11	36.67	6	20.00
Vegan	0	0.00	0	0.00

In the various studies researchers observed that a diet that differ from typical western foods is a vegetarian diet, and vegetarians in the U.S. have a lesser prevalence of diabetes than omnivores. (Consuming both plant and animal foods) (Snowdon *et al.* 1985, Vang *et al.* 2008, Fung *et al.* 2004).

Chiu *et al.* (2018) reported that pesco-vegetarians and semi-vegetarians have reported to have less protection against diabetes than those on more non vegetarian diet compare with restrictive vegetarian diets. Semi-vegetarians who consume meat

minimally but regularly consume saturated fats, which declines insulin secretion and can prompt beta-cell apoptosis.

Olfert and Wattick (2018) reported that the peculiarity in risk by type of vegetarian diet highlights a key aspect in generating the health advantages of vegetarian diets. Vegetarian diets are more fetching all over the world, but not all vegetarian diets remained to healthy concepts, such as consuming whole grains, vegetables, fruits, and legumes. To evaluate the type II diabetes risk between vegetarians who consume a healthy diet (high in fruits and vegetables, low in refined grains and starchy foods, and added sugars) and those who don't consume (characterized by whole grains, fruits, vegetables and legumes). A healthful Plant-Based Diet Index (hPDI) and Unhealthy Plant-Based Diet Index (uPDI) that illustrious between healthy and unhealthy plant foods were also formed. They stated that the PDI and hPDI were inversely linked with Type II diabetes mellitus, and the uPDI was positively associated with Type II diabetes mellitus. This research reported the benefit of consuming a vegetarian diet that is high in whole grains, vegetables, fruits, nuts, and legumes in preventing Type II diabetes mellitus.

4.14.2 Dietary assessment of participants:

Nutrition is an input to the basis for wellbeing and development. Better nutrition is a major entry point to attain better quality of life. Dietary survey is study of nutritional status of individuals or groups. It is providing essential information on food habits and inadequate nutrient intake. In the National Family Health Survey of India in 2005-06, among participants of both genders between ages from 20-49 years reported relation between food intake and prevalence of diabetes (Agrawal *et al.* 2015).

4.14.2.1 Energy:

Table 4.57 shows that mean value for energy in the control group 1970 Kcal which was slightly less than the recommended values whereas mean value for energy in the experiment group 2039 Kcal which was slightly less than recommended value.

Angela *et al.* (2014) reported that the links between energy intake and type II diabetes risk markers, which illustrate a clear graded relationship.

**Table 4.57 Dietary assessment of participants
(Control Group n= 30 and Experiment Group n=30)**

S.No.	Nutrient	RDA (2020)	Control Group (±S.D.)	Experimental Group (±S.D.)
1	Energy	2110 Kcal	1970±337.52 (Kcal)	2039±314.49 (Kcal)
2	Protein	54.0 g/d	54.88±11.76 (g)	64.06±12.06 (g)
3	Fat (Visible and Invisible)	30g/d (Visible)	73.18±34.87 (g)	67.19±18.59 (g)
4	Carbohydrates	NA	273.45±46.66 (g)	294.81±54.23 (g)
5	Total Dietary Fiber	41g/d	38.84±13.28(g)	42.98±13.64 (g)
6	Retinol	840 µg/d	449±119	432.52±188.04
7	Thiamine	1.8 mg /day	1.60±0.41	1.72±0.27
8	Riboflavin	2.5 mg/day	1.02±0.22	1.19±0.24
9	Niacin	18mg/d	10.53±4.16	12.81±5.31
10	Vitamin B6	2.4 mg/d	0.74±0.28	0.91±0.31
11	Calcium ((mg)	1000 mg/d	605±135.00	730±197
12	Iron (mg)	19 mg/d	18.16±4.79	21.28±6.55

4.14.2.2 Protein:

Data from Table 4.57 depicted that protein content in the both groups was to some extent higher than the recommended value 54.88 g and 64.06 g, respectively. Similar study was conducted by Mittal (2014) and reported that the protein consumption was also normal in most of the respondents. Only a few numbers were consumed less amount of protein. 10 per cent consumed proteins ranging from 30-40 g, 24 per cent from 40-50 g, 52 per cent from 50-60 g and 14 per cent were consuming from 60-70 g.

4.14.2.3 Fat:

Value in the Table 4.57 shows that the mean value of fat consumption was in the control group (73.18g) as well as in the experiment group (67.19g) was higher than the recommended values.

Similar study was conducted by Mittal (2014) reported that most of respondents were consuming fat more than the normal amount. A small number of numbers of respondents were consuming under the normal range. 14 per cent were consuming from the range from 20-30 g, 42 per cent from 30-40 g, 34 per cent from 40-50 g and 10 per cent from 50-60 g.

Over recent decades surveys have reported children and young people with diabetes consume fat and in that fat consumption saturated fat also above than dietary recommendations (Virtanen *et al.* 2001) and this situation has not changed (Helgeson *et al.* 2006; Mayer-Davies *et al.* 2006; Overby *et al.* 2007). The intake of total fat, saturated fat, and trans-fatty acids should be decreased (Franz 2002) while Monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) can be used as replacement to keep lipid intake with in recommended ranges or to increase the lipid profile (Franz 2002).

4.14.2.4 Carbohydrates:

Data from the Table 4.57 shows the consumption of carbohydrates by the participants. In the control group mean consumption of carbohydrates was 273.45 g while experiment group was consuming 294.71 g of carbohydrates.

Similar results shown by Mittal (2014) reported that mostly people consumed average recommended amount of carbohydrates. But some even consumed the amount more than the normal range i.e. 24 per cent consumed from range 250-300 g, 36 per cent from 300- 350 g, 32 per cent from 350-400 g and 08 per cent from 400-450 g. According to ICMR evidence is inconclusive for an ideal amount of carbohydrate intake for people with diabetes. Therefore, collaborative goals should be developed for individuals with diabetes. 55-60 per cent of energy from carbohydrates is an ideal recommendation. Carbohydrates should be complex in nature.

4.14.2.5 Dietary Fiber:

Table 4.57 presented the dietary fiber consumption for both groups. In the control group mean value for dietary fiber consumption was 38.84 g which was slightly lower than the recommended values. While, in the experiment group mean value for dietary fiber consumption was 42.98 g that was slightly higher than the recommended values.

Dietary fiber can decrease the risk of type II diabetes by several mechanisms. Dietary fiber can decrease glucose absorption from the intestine, which reduce the glycemic index of carbohydrates (Livesey and Tagami, 2009). Some types of dietary fiber have been directed towards to augmentation of bile acid excretion and increase pool size of bile acids (Story and Kritchevsky, 1978). Those increased bile acid increase secretion of glucagon-like peptide 1 via activation of TGR5 and improve glycemic control (Slavin 2008, Mansour *et al.* 2013, Harach *et al.* 2012, Watanabe *et al.* 2012)

In addition, increased synthesis of short-chain fatty acids by bacterial fermentation of fiber is also notion to have beneficial effects on glucose levels and energy homeostasis by way of regulation the intestinal gluconeogenesis (Weickert and Pfeiffer, 2000, Jenkins *et al.* 2000, De Vadder *et al.* 2014)

4.14.2.6 Vitamins:

Tables 4.57 illustrate the mean value of vitamin consumption in the both group. The average consumption of retinol was 449 µg and 432.52 µg by the control group and experiment group. In the control group average consumption of thiamine, riboflavin, niacin and vitamin B6 was 1.60 mg, 1.02 mg, 10.53 mg and 0.74 mg, respectively. While in the experiment group average consumption of thiamine, riboflavin, niacin and vitamin B6 was 1.72 mg, 1.19 mg, 12.8 mg and 0.91mg, respectively. Data clearly shows that that both groups were consuming less than recommended amount of vitamins.

4.14.2.7 Minerals:

Tables 4.57 present the mean value of minerals consumption in the both group. In the control group average consumption of calcium and iron 605 mg and 18.16 mg that is less than recommended amount. While, in the experiment group average consumption of calcium and iron 730 mg and 21.28 mg, Data shows that consumption of calcium less than recommended amount while iron was slightly more than the recommended quantity.

4.15 FASTING BLOOD GLUCOSE LEVEL OF PARTICIPANTS

Table 4.58 shows in the control group the mean value of fasting blood glucose level at the initial phase was 158.91 mg/dl and after thirty days mean value was blood

glucose level 155.26 mg/dl. There was no significant difference in fasting blood glucose level in the control group. While, in the experiment group the mean blood glucose level was 157.04 mg/dl in the initial phase and after the administration of flavoured probiotic *Lassi* (300 ml) for thirty days, the mean blood glucose level was 132.14 mg/dl (Without control the lifestyle and diet) was reported. There was significant difference in fasting blood glucose level after the intervention.

Present study supported by Moroti *et al.* (2012) reported that the feeding of *L. acidophilus*, *B. bifidum*, fructooligosaccharides to twenty participants ages 50-60 years was double-blind, for 30 days. They reported that significant increase in HDL and a significant reduction in glycaemia.

**Table 4.58 Fasting blood glucose level of participant
(Control Group n= 30 and Experiment Group n=30)**

Particulars	Control Group		Experiment Group	
	Initial Phase	After 30 days	Initial Phase	Post feeding Phase
Mean ± S.D.	158.91±15.74	155.26±14.84	157.04±13.31	132.14±14.19
Variance	247.98	220.44	178.03	173.23
df	29	NS: Non Significant	29	* significant at 5% level of significance
t Stat	1.613 ^{NS}		15.033*	

Ejtahed *et al.* (2012) Sixty-four patients with type II diabetes mellitus, 30 to 60 y old, were rationed to two groups in this randomized, double-blind, controlled clinical trial. The patients in the intervention group administrated 300 g/day of probiotic yogurt incorporated with *Lactobacillus acidophilus* La5 and *Bifidobacterium lactis* Bb12 and those in the control group consumed 300 g/day of conventional yogurt for 6 weeks.

In a meta-analysis conducted by Yao *et al.* (2017), probiotic ingestion significantly diminished HbA1c and insulin levels in patients with T2DM.

Raygan *et al.* (2018) studied that supplementation of 8×10^9 CFU/g probiotic, possessing *Lactobacillus acidophilus*, *Bifidobacterium bifidum*, *Lactobacillus*

reuteri and *Lactobacillus fermentum* (each 2×10^9) and 50,000 IU vitamin D every 2 weeks for 12 weeks to Type II DM patients with coronary heart disease (CHD) had worthwhile influences on mental health parameters, total antioxidant capacity, inflammatory markers and glycemic control, although there was no effect on other parameters of metabolic profiles.

Further, several meta-analyses of various studies support that the administration of probiotics to patients with Type II DM probably affects, to a moderate degree, the metabolism of glucose, particularly in ≥ 8 weeks interventions (Ruan *et al.* 2015; Wang *et al.* 2017; Yao *et al.* 2017; Hu *et al.* 2017; Kesika, *et al.* 2019; Zhang *et al.* 2016).

SUMMARY AND CONCLUSION

Probiotics have a wide range of therapeutic benefits. Indigenous strain *L. helveticus* MTCC 5463 has been proven to possess probiotic potentials with their specific favorable role in the gastrointestinal tract and ability to reduce cholesterol and stimulate immunity. Goat milk possesses potential for successful delivery of probiotics though it has goaty flavour. Goat milk possesses some specific characteristics which bestow it different from cow milk like fat globules size which is smaller than cow milk that provide a smoother texture in derived products, lesser amount of alpha1-casein, consequential in softer gel products, a lower viscosity and higher water holding capacity. Changing lifestyle, high demand for new kinds of tastes with the enormous innovation in the food market is expected to drive the growth for the food with different flavour. Expanding health awareness, there has been a huge demand for sugar substitutes that would provide lesser or no calories and possess better sweetening potency. Stevia, which plays an important role as a non-nutritive natural sweetener, emerged as a safe sugar substitute that does not pose any threat to human health. In view of the above resume, combination of the goat milk, probiotic culture and stevia extract was used to make the flavored probiotic *Lassi* and yogurt. The study was focused on the objectives, included preparation of probiotic products, its sensory profile, physico-chemical properties and storage study of products and its effects on diabetic patients.

To obtain data the products (*Lassi* and Yogurt) was prepared with different flavours. *Lassi* was prepared with Pineapple, *Kesar Elachi* and Rose Flavour and yogurt was prepared with Kevda, *Kesar Pista* and Raspberry flavour.

Salient findings of the experiment are given below.

(1) PHYSICO-CHEMICAL PROFILE OF FRESH PRODUCTS:

Physio-chemical profile of probiotic *Lassi*:

The highest average value for moisture content was observed in LS1 (88.10%) followed by LS₂ (87.90%), LS₃, (87.31%) and LS₀ (87.27%). The highest average value for fat content was observed in LS₂ (3.59%), followed by LS₁ (3.58%), LS₃

(3.57%) and LS₀ (3.50%), respectively. The highest mean value for carbohydrates content was observed in LS₀ (4.74%), followed by LS₃ (4.70%), LS₂ (4.21%) and LS₁ (4.13%), respectively. The highest average value for protein content was observed in LS₀ (3.52%), followed by LS₂ (3.41%), LS₃ (3.37%) and LS₁ (3.31%), respectively. The highest average value for ash content was observed in LS₀ (0.80%) followed by LS₃ (0.79%), LS₂ (0.78%) and LS₁ (0.77%) was respectively. The highest average value for energy value was observed in LS₀ (64.54 Kcal) followed by and LS₃ (64.41 Kcal) LS₁ (61.98 Kcal), LS₂ (62.79 Kcal) and LS₃ (64.41 Kcal) was respectively. The highest average value for total solids content was observed in LS₀ (12.70%) followed by LS₃ (12.52%), LS₂ (12.05%) and LS₃ (11.79%), respectively. The highest average value for Solids Non-Fat (SNF) content was observed in LS₀ (9.19%) followed by LS₃ (8.93%), LS₂ (8.22%) and LS₃ (8.37%), respectively. The highest average value for pH level was observed in LS₀ (4.40) followed by LS₁ (4.39), LS₃ (4.39) and LS₂ (4.38), respectively. The highest average value for titrable acidity level was observed in LS₂ (0.84%), followed by LS₁, LS₃ (0.83%) and LS₀ (0.82%), respectively. The highest mean value for viscosity was observed in LS₀ (316.10 cp) followed by LS₁, LS₂ (315.40 cp) and LS₃ (314.70 cp), respectively.

Mineral profile of probiotic *Lassi*:

The highest average value for calcium content was observed in LS₀ (116.40 mg%), followed by LS₃ (115.40 mg%), LS₂ (114.80 mg%) and LS₁ (112.40 mg%), respectively. . The highest average value for phosphorus content was observed in LS₀ (123.91 mg%), followed by LS₃ (122.60 mg%), LS₁ (121.81 mg%) and LS₂ (121.12 mg%), respectively. The highest average value for sodium content was observed in LS₀ (35.87 mg%), followed by LS₃ (35.16 mg%), LS₁ (35.11 mg%) and LS₂ (33.90 mg%), respectively. The highest average value for potassium content was observed in LS₀ (162.21 mg%), followed by LS₃ (162.09 mg%), LS₂ (160.80 mg%) and LS₁ (159.77 mg%), respectively. The highest average value for magnesium content was observed in LS₀ (13.98 mg%), followed by LS₃ (13.95 mg%), LS₂ (13.93 mg%) and LS₁ (13.92 mg%), respectively. The highest average value for copper content was observed in LS₀ (0.058 mg%), followed by LS₃ (0.057 mg%), LS₂ (0.056 mg%) and LS₁ (0.054 mg%), respectively. The highest average value for iron content was observed in LS₀ (0.054 mg%), followed by LS₃ (0.053 mg%), LS₂ (0.051 mg%) and

LS₁ (0.049 mg%), respectively. The highest average value for iron content was observed in LS₀ (0.53 mg%), followed by LS₁, LS₂ and LS₃ (0.52 mg%), respectively.

Physio-chemical profile of probiotic yogurt:

The highest average value for moisture content was observed in YS₂ (86.95%), followed by YS₃ (86.68%), YS₁ (86.52%) and YS₀ (85.98%), respectively. The highest average value for fat content was observed in YS₂ (4.03%) followed by YS₃ (3.94%), YS₁ (3.92%) and YS₀ (3.70%), respectively. The highest average value for carbohydrates content was observed in YS₀ (5.37%) followed by YS₁ (4.97%), YS₃ (4.79%) and LS₂ (4.55%) which was at some level lower than control. The highest average value for protein content was observed in YS₀ (3.98%), followed by YS₁ (3.76%), YS₃ (3.75%) and YS₂ (3.65%) which was slightly less than the control. The highest average value for ash content was observed in YS₀ (0.84%) followed by YS₁, LS₃ (0.83%) and YS₂ (0.80%) which was slightly lower with compare to the control. The highest average value for energy value was observed in YS₀ (70.70 Kcal) followed by YS₁, (70.20 Kcal) YS₃ (69.62 Kcal) and YS₂ (69.07 Kcal), respectively. The highest average value for total solids content was observed in YS₀ (13.97%), followed by YS₁, (13.47%), YS₃ (13.31%) and YS₂ (13.04%) which was slightly lower with compare to the control. The highest average value for Solid Non-Fat content was observed in YS₀ (10.20%), followed by YS₁, (9.55%), YS₃ (9.35%) and YS₂ (8.99%) which was slightly lower with compare to the control. The highest average value for pH level was observed in YS₁ (4.38), followed by YS₂, YS₃ (4.37) YS₁, (4.36), respectively. The highest average value for titrable acidity level was observed in YS₀ (0.86%), followed by YS₁, YS₂ (0.85%) and YS₂ (0.84%), respectively. The highest average value for viscosity was observed in YS₀ (376.20 cp), followed by YS₁, (375.50 cp) YS₃ (375.80 cp) and YS₂ (375.40 cp) which was slightly lower with compare to the control.

Mineral profile of probiotic yogurt:

The highest average value for calcium content was observed in YS₀ (130.89 mg%), followed by YS₁ (130.74 mg%), YS₃ (129.54 mg%) and YS₂ (129.37 mg%), respectively. The highest average value for phosphorus content was observed in YS₀ (139.49 mg%), followed by YS₁ (139.47 mg%), YS₃ (139.84 mg%) and YS₂ (139.01 mg%), respectively. The highest average value for sodium content was observed in

YS₀ (40.28 mg%), followed by YS₃ (40.22 mg%), YS₁ (39.81 mg%) and YS₂ (38.75 mg%), respectively. The highest average value for potassium content was observed in YS₀ (175.55 mg%), followed by YS₁ (175.46 mg%), YS₃ (175.04 mg%) and YS₂ (174.46 mg%), respectively. The highest average value for magnesium content was observed in YS₀ (15.11 mg%), followed by YS₁ (14.91 mg%), YS₃ (14.86 mg%) and YS₂ (14.82 mg%), respectively. The highest average value for copper content was observed in YS₀ (0.072 mg%), followed by YS₁, YS₃ (0.071 mg%) and YS₂ (0.069 mg%), respectively. The highest average value for iron content was observed in YS₀ (0.081 mg%), followed by YS₁, YS₃ (0.079 mg%) and YS₁ (0.077 mg%), respectively. The highest average value for iron content was observed in YS₀ (0.54 mg%), followed by YS₁ (0.53 mg%), YS₃ (0.52 mg%) and YS₃ (0.51 mg%), respectively. There was no wide variation in the mineral content in all yogurt samples. It was slightly less than control. The effect of the addition of flavour and stevia extract on the calcium, phosphorus, sodium, potassium, magnesium, copper, iron and zinc content was non-significant ($p < 0.05$) between all flavoured Probiotic yogurt samples on the day of preparation.

SENSORY PROFILE OF FRESH PROBIOTIC *LASSI*:

The highest average value for colour and appearance was observed in LS₂ (8.33) which was “liked very much”, followed by LS₅ (8.27), LS₃, LS₄ and LS₆ (8.07) lowest score was found in LS₀ and LS₁ (8.00). *Lassi* samples prepared with stevia extract and powder did not differ significantly. The highest average value for flavour was observed in LS₃ (8.67) which was “liked very much”, followed by LS₂ (8.47), LS₁ (8.33), LS₃ and LS₅ (7.60) lowest score was observed in LS₀ (7.40), respectively which was “like moderately”. Flavoured *Lassi* obtained a higher score compared to the control. There was a significant difference ($p < 0.05$) between the flavour score provided by panel members to the samples of *Lassi* prepared with stevia extract and powder. The highest average value for consistency was observed in LS₃ (8.47), followed by LS₁ (8.40), LS₂ and LS₅ (8.20) lowest score determined in LS₀ LS₄, and LS₆ (8.00), respectively. No significant difference ($p < 0.05$) was found between the samples of *Lassi* prepared with stevia extract and powder. The highest average value for mouth feel was observed in LS₃ (8.47) which was “liked very much” followed by LS₂ (8.27), LS₁ (8.07), LS₄, LS₆ (7.07), however, lowest score was determined in LS₅ (7.00) LS₀ (6.93), respectively. There was a significant difference ($p < 0.05$) found

between the samples of *Lassi* prepared with different flavours, stevia extract and powder. The highest average value for overall acceptability was observed in LS3 (8.40) which was “liked very much”, followed by LS₂ (8.27), LS₁ (8.07), LS₄ and LS₆ (7.07), the lowest score was determined in LS₅ (7.00) and LS₀ (6.93) respectively. A significant difference ($p < 0.05$) was found between the overall acceptability score allotted by panel members to the samples of *Lassi* prepared with stevia extract and powder.

SENSORY PROFILE OF FRESH PROBIOTIC YOGURT:

The highest average value for colour and appearance was observed in YS₂ (8.40) which was “liked very much”, followed by YS₁ (8.27), YS₃, YS₄ and YS₅ (8.20) lowest score was found in YS₀ and YS₁ (8.13), respectively which was also obtained score “liked very much” category. Yogurt samples prepared with stevia extract and powder did not differ significantly. The highest average value for flavour was observed in YS₂ (8.00) which was “liked very much” followed by YS₁, YS₃ (7.93), YS₅ (7.87), YS₄ (7.73) and YS₆ (7.60) lowest score was observed in YS₀ (6.87), respectively which was “liked slightly”. Flavoured yogurt obtained more scores compared to control. A significant difference ($p < 0.05$) was found between the samples of yogurt prepared with stevia extract and powder. The highest average value for consistency was observed in YS₂ (8.40) which was “liked very much” followed by YS₁ (8.20), YS₃ (8.13) and YS₄ YS₅ (8.07) lowest score was determined in YS₀ and YS₆ (8.00), respectively which was also found in category “liked very much. No significant difference in consistency score ($p < 0.05$) was found between the samples of yogurt prepared with stevia extract and powder. The highest average value for mouth feel was observed in YS₂ (8.00), which was “liked very much” followed by YS₃ (8.93), YS₁ (7.80), YS₅ (7.20), YS₄ (7.13), YS₆ (7.07) however, the lowest score was determined in YS₀ (6.87), respectively which was “liked slightly”. A significant difference ($p < 0.05$) was found between the samples of yogurt prepared with different flavours, stevia extract and powder. The highest average value for overall acceptability was observed in YS₂ (8.12) which was “liked very much”, followed by YS₃ (8.07), YS₁ (8.00), YS₅ (7.20), YS₄ (7.13) YS₆ (7.07) however lowest score was determined in YS₀ (7.00), respectively which was “moderately liked” by the panel members. There was a significant difference ($p < 0.05$) found between the score of

overall acceptability allotted by panel members to the samples of yogurt prepared with stevia extract and powder.

SHELF LIFE ASSESSMENT OF *LASSI* AND YOGURT:

Sensory profile of probiotic *Lassi*

Colour and appearance score range for LS₀, LS₁, LS₂ and LS₃ was 6.60-8.00, 6.67-8.00, 7.00-8.33 and 6.73-8.07, respectively. The colour and appearance of all the flavoured probiotic *Lassi* and control non-significantly decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. Flavour score range for LS₀, LS₁, LS₂ and LS₃ was 5.00-7.47, 5.53-8.33, 5.20-8.47 and 5.40-8.67, respectively. The flavour of all the flavoured probiotic *Lassi* and control samples were significantly decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. The consistency score range for LS₀, LS₁, LS₂ and LS₃ was 6.80-8.00, 7.00-7.93, 6.87-8.20 and 7.20-8.47, respectively. The consistency score of all the flavoured probiotic *Lassi* and control non-significantly gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. The mouth feel score range for LS₀, LS₁, LS₂ and LS₃ was 4.90-6.93, 5.33-8.07, 5.20-8.07 and 5.27-8.47, respectively. The mouth feel score of all the flavoured probiotic *Lassi* and control significantly gradually decreased throughout the storage period from the day of preparation to the 11th day of storage. The overall acceptability score range for LS₀, LS₁, LS₂ and LS₃ was 5.00-6.93, 5.40-8.27, 5.20-8.33 and 5.80-8.40, respectively. The overall acceptability score of all the flavoured probiotic *Lassi* and control significantly decreased throughout the storage period from the day of preparation to the 11th day of storage.

Physio-chemical properties of probiotic *Lassi* :

The moisture content range for LS₀, LS₁, LS₂ and LS₃ was 87.27-85.94, 88.18-86.10, 87.92-86.12 and 87.31-86.06, respectively. The moisture content of all the samples decreased throughout the storage period from fresh yogurt to the 11th day of storage in all *Lassi* samples. The total solids content range for LS₀, LS₁, LS₂ and LS₃ was 12.70-14.01, 11.79-13.90, 12.05-13.78 and 12.52-13.92, respectively. As moisture content turned down due to that, the total solids content was more than

before. The protein content range for LS₀, LS₁, LS₂ and LS₃ was 3.52-4.82, 3.31-4.78, 3.41-4.80 and 3.37-4.64 per cent, respectively. Protein content was increased during the storage period in all the samples of flavoured probiotic *Lassi* along with control. The fat content range for LS₀, LS₁, LS₂ and LS₃ 3.50-3.89, 3.58-3.97, 3.59-4.00 and 3.57-3.95 per cent, respectively. Fat content was slightly increased during the storage period. The ash content range for LS₀, LS₁, LS₂ and LS₃ 0.80-0.89, 0.79-0.85, 0.78-0.83 and 0.77-0.82 per cent, respectively. The ash content of flavoured probiotic *Lassi* as well as in the control sample was increased with the progression of the storage period. The carbohydrate content was ranges for LS₀, LS₁, LS₂ and LS₃ 4.74-4.17, 4.13-3.89, 4.21- 4.01 and 4.86-4.26 per cent, respectively. Carbohydrate content was slightly decreased during the storage period. The pH level varies for LS₀, LS₁, LS₂ and LS₃ was 4.07-4.40, 4.02-4.39, 4.01-4.38 and 4.05-4.39, respectively. pH content was slightly decreased during the storage period. Titratable acidity level range for LS₀, LS₁, LS₂ and LS₃ 0.82-1.09, 0.83-1.17, 0.84-1.22 and 0.83-1.19 per cent, respectively. Titratable acidity level was increased during the storage period There was a significant difference found due to the storage period.

Microbial viability of probiotic *Lassi*:

The log lactobacilli count (CFU/ml) range for LS₀, LS₁, LS₂ and LS₃ as 7.97-7.73, 7.93-7.62, 7.96-7.63 and 7.96-7.65, respectively. A non-significant difference in Lactobacilli count difference between control and flavoured probiotic *Lassi* samples was found during the storage duration. The log lactobacilli count (CFU/ml) range for LS₀, LS₁, LS₂ and LS₃ as 8.90-8.73, 8.83-8.68, 8.83-8.67 and 8.85-8.68 log CFU/ml, respectively. The Streptococci count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. The log yeast and mould count (CFU/ml) range for LS₀, LS₁, LS₂ and LS₃ as 0.63-1.16, 0.64-1.19, 0.63-1.21 and 0.64-8.1.18 log CFU/ml, respectively. In the present study coliform count was absent.

Sensory profile of probiotic yogurt:

The colour and appearance score range for YS₀, YS₁, YS₂ and YS₃ was 7.33-8.13, 7.53-8.27, 7.60-8.40 and 7.50-8.20, respectively. The colour and appearance of all the flavoured probiotic yogurt and control non-significantly decreased throughout the storage period from the day of preparation to the 11th day. The flavour

score range for YS₀, YS₁, YS₂ and YS₃ was 5.00-6.87, 5.80-7.80, 5.93-8.20 and 5.67-7.93, respectively. The flavour score of all the flavoured probiotic yogurt and control non-significantly decreased throughout the storage period from the day of preparation to the 11th day of storage in all yogurt samples. The consistency score range for YS₀, YS₁, YS₂ and YS₃ was 6.13-8.00, 6.67-8.40, 6.87-8.20 and 6.80-8.13, respectively.” The consistency score of all the flavoured probiotic yogurt and control non-significantly decreased throughout the storage period from the day of preparation to the 11th day of storage. The mouth feel score range for YS₀, YS₁, YS₂ and YS₃ was 4.83-6.87, 5.00-7.80, 5.87-8.00 and 5.80-7.93 respectively. The mouth feel score of all the flavoured probiotic yogurt and control non-significantly decreased throughout the storage period from the day of preparation to the 11th day of storage. The overall acceptability score range for YS₀, YS₁, YS₂ and YS₃ was 5.00-7.00, 5.20-8.00, 5.60-8.12 and 5.10-8.07, respectively. The overall acceptability score of all the flavoured probiotic yogurt and control non-significantly decreased throughout the storage period from the day of preparation to the 11th day of storage.

Physio-chemical properties of probiotic yogurt:

The moisture content varies for YS₀, YS₁, YS₂ and YS₃ was 84.04-85.98, 84.46-86.52, 85.24-86.95 and 85.21-86.68 per cent, respectively. The moisture content of all the samples decreased throughout the storage period from fresh yogurt to the 11th day of storage. The total solids content varies for YS₀, YS₁, YS₂ and YS₃ was 13.97-15.20, 13.47-14.98, 13.04-14.74 and 13.31-14.74 per cent, respectively. The protein content varies for YS₀, YS₁, YS₂ and YS₃ 3.98-4.20, 3.76-4.15, 3.65-4.12 and 3.75-4.10 per cent, respectively. Protein content was increased during the storage period in all the samples of flavoured probiotic yogurt along with control sample. The fat content varies for YS₀, YS₁, YS₂ and YS₃ was 3.70-3.99, 3.92-4.18, 4.03-4.20 and 3.94-4.07 per cent, respectively. The increase in fat percentage is due to the decrease in the moisture content. The ash content varies for YS₀, YS₁, YS₂ and YS₃ was 0.84-0.95, 0.83-0.93, 0.80-0.91 and 0.82-0.90 per cent, respectively. The carbohydrate content varies for YS₀, YS₁, YS₂ and YS₃ was 5.37-5.19, 4.97-4.61, 4.55-4.23 and 4.79-4.53 per cent, respectively. Carbohydrate content was slightly decreased during the storage period. The titratable acidity levels varies for YS₀, YS₁, YS₂ and YS₃ was 0.86-1.20, 0.85-1.23, 0.85-1.21 and 0.84-1.26 per cent, respectively.

Microbial viability of probiotic yogurt:

The log lactobacilli count (CFU/ml) range for YS₀, YS₁, YS₂ and YS₃ as 8.67-8.49, 8.62-8.39, 8.60-8.40 and 8.62-8.44 log CFU/ml, respectively. The lactobacilli count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. The log streptococci count (log CFU/ml) range for YS₀, YS₁, YS₂ and YS₃ as 9.34-8.70, 9.28-8.66, 9.27-8.62 and 9.32-8.64 log CFU/ml, respectively. The Streptococci count gradually decreased throughout the storage period from the day of preparation to the 11th day of storage in all *Lassi* samples. The log yeast and mould count (log CFU/ml) range for YS₀, YS₁, YS₂ and YS₃ as 0.68-1.18, 0.70-1.25, 0.67-1.27 and 0.69-1.23 log CFU/ml, respectively. The coliform count was absent during the entire storage period.

INTERVENTION OF PROBIOTIC LASSI:

General Information:

In the control group and experiment group majority of the participants fell in the age range of 41-45 years. Most of the participants were working from sedentary to moderate work type. In the both group Most of the have nuclear family. This has been observed that majority of participant have alarmed sleeping pattern.

Anthropometric measurement:

That height of the control group majority (60.00 per cent) of participants were having height between 160 to 170 cm, (35.33 per cent) followed by (23.33%) were having the height from 150-160 cm, (13.33%) having the height from 171-180 cm rest of participants (3.33%) having the height more than 180 cm. Whereas in the experiment group less than half (40%) of the participants were having the from 160-170 cm, followed by (30%) who have height range from 171-180 cm, (20%) who have height range from 171-180 cm remaining (10%) were having height <180 cm. It was observed that 30 per cent participants were come under the weight category of 60-70 kg and 81-90 kg, 23.33 per cent were come under the weight category of 71-80 kg, 13.33 per cent were come under the weight category of 91-100 kg and 3.33 per cent were come under the weight category of more than 100 kg, respectively in the control group. However, in the experiment group, majority of the 43.33 per cent come under the weight category of 71-80 kg followed by 26.66 per cent come under the

weight category of 60-70 kg, 16.66 per cent, 10 per cent come under the weight category of 91-100 kg 3.33 per cent come under the weight category of more than 100 kg, respectively. Among the control group most of the participants (40%) have BMI <30 kg/m² followed by (36.66%) who have BMI 18.5-24.9 kg/m² and (23.33%) have BMI range from 25-29.9 kg/m². Whereas, among the experiment group (40%) of the candidate have BMI range from 25-29.9 kg/m² and (30%) participants have BMI 18.5-24.9 kg/m² and <30 kg/m² by each group.

Diabetic History

The presence of diabetic history among both groups' participants. In the control group majority (56.67%) of the participants have positive diabetes mellitus history; whereas, remaining (43.33%) participants have negative diabetes mellitus history. Similarly, in the experiment group majority (63.33%) of the participants and remaining (36.67%) participants have negative diabetes mellitus history. In the control group, majority of the candidate (93.33%) were taking treatment through oral drugs with dietary modification whereas in the experiment group all of the candidate (100%) were taking treatment through oral drugs with dietary modification.

Dietary Survey

In the both groups that greater part of the participants were non vegetarian and ovo-vegetarian. Mean value in the control group 1970 Kcal which was slightly less than the recommended values whereas mean value for energy in the experiment group 2039 Kcal which was slightly lower than the recommended value. Protein content in the both groups was to some extent higher than the recommended value 54.88 g and 64.06 g, respectively. The mean value of fat consumption was in the control group (73.18 g) as well as in the experiment group (67.19 g) was higher than the recommended values. In the control group mean consumption of carbohydrates was 273.45 g while experiment group was consuming 294.71 g of carbohydrates. In the control group mean value for dietary fiber consumption was 38.84 g which was slightly lower than the recommended values. The average consumption of retinol was 449 µg and 432.52 µg by the control group and experiment group. In the control group average consumption of thiamine, riboflavin, niacin and vitamin B6 was 1.60 mg, 1.02 mg, 10.53 mg and 0.74 mg, respectively. While in the experiment group average consumption of thiamine, riboflavin, niacin and vitamin B6 was 1.72 mg,

1.19 mg, 12.8 mg and 0.91mg, respectively. In the control group average consumption of calcium and iron 605 mg and 18.16 mg that is less than recommended amount. While, in the experiment group average consumption of calcium and iron 730 mg and 21.28 mg,

Fasting Blood Glucose level of participants

The control group the mean value of fasting blood glucose level at the initial phase was 158.91 mg/dl and after thirty days mean value was blood glucose level 155.26 mg/dl. There was no significant difference in fasting blood glucose level in the control group. While, in the experiment group the mean blood glucose level was 157.04 mg/dl in the initial phase and after the administration of flavoured probiotic *Lassi* (300 ml) for thirty days, the mean blood glucose level was 132.14 mg/dl. There was significant difference in fasting blood glucose level after the intervention.

CONCLUSION :

The present research acquaints that goat milk can be use as a vehicle to provide probiotics, with the incorporation of stevia extract which lower the calorific value of probiotic *Lassi* and yogurt and provide sweet taste also. High amount of calorie consumption leads to various non-communicable diseases as obesity, diabetes mellitus and cardiovascular diseases. The mean score of overall acceptability of both *Lassi* and yogurt prepared with stevia powder and extract, *Lassi* and yogurt prepared with stevia extract obtained more scores compared with *Lassi* and yogurt prepared with stevia powder; however, control obtained lower scores compared with the both products. Among all the samples highest score was obtained by Rose flavour. The products was stored for 11 days and evaluated at fresh, 3rd, 5th, 7th, 9th and on 11th day. Sensory score was decreased throughout the storage period. The colour and appearance and consistency of all the flavoured probiotic *Lassi* and yogurt along with control non-significantly decreased throughout the storage period. Whereas, flavour, mouth feel and overall acceptability of all the flavoured probiotic *Lassi* and yogurt samples along control samples were significantly decreased throughout the storage period from the day of preparation to the 11th day of storage. Whereas, In human trial, after feeding the 300 ml of Rose flavour *Lassi* the experiment group the mean blood glucose level was shown significant difference in fasting blood glucose level after the 30 days. Therefore, it could be concluded that probiotic *Lassi* was effective in

managing the blood glucose levels and is suitable for diabetic patients to fulfill their cravings regarding sweetness.

RECOMMENDATIONS:

- Other products can be prepared based on goat milk.
- Other base can be used to deliver probiotics and other culture of probiotics can be used.
- Further human trial can be conducted for other non-communicable diseases.

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Ph.D. Thesis (2023)**

Topic: “Effect of Goat Milk Based Probiotic Products on Diabetic Patients”

ABSTRACT

Probiotics considered as a functional food have a wide range of therapeutic benefits. Goat milk can be used as potential source of delivery of probiotics. Along with innovative taste awareness about health and nutrition is also increasing. Today's diabetes is one of the major global health issues because of its burgeoning prevalence. The burgeoning prevalence of diabetes mellitus is estimated to reach 366 million by 2030. People are facing problem regarding consumption of high content of sugar and finding the sugar substitutes that would provide lesser or no calories with better sweetness. Hence, the present study was planned to prepare goat milk based fermented products includes starter culture (*Streptococcus thermophilus* MD2), probiotic culture (*Lactobacillus helveticus* MTCC 5463) and stevia extract and powder with different flavours. *Lassi* was prepared with Pineapple, *Kesar-Elaichi* and Rose flavour and yogurt was prepared with Kevda, Kesar-Pista and Raspberry flavour. Probiotic *Lassi* and yogurt as a functional food and analyse the sensory profile, physico-chemical properties, shelf life and impact on blood glucose level. By observing the mean score of overall acceptability of both *Lassi* and yogurt prepared with stevia powder and extract, *Lassi* and yogurt prepared with stevia extract obtained more scores compared with *Lassi* and yogurt prepared with stevia powder; however, control obtained lower scores compared with the both products. Among all the samples highest score was obtained, i.e. Rose flavour followed by Kesar Elaichi and Pineapple, whereas in yogurt highest score was Kevda, Kesar-Pista and Raspberry which was prepared with stevia extract. So stevia extract incorporated products were selected for further work. Nevertheless, among all the sample of probiotic *Lassi* and yogurt highest mean score (Sensory profile) was obtained by rose flavour. The physicochemical properties of *Lassi* sample prepared with Rose flavour shows moisture content (87.31%), fat content (3.57%), carbohydrates content (4.70%), ash content

(0.79%), low energy value (64.41Kcal), T.S. (12.52%), SNF (8.93%), desired pH (4.39), titratable acidity (0.83%) and viscosity (314.70 cp). Mineral profile shows fair amount of calcium (115.40 mg%), phosphorus (122.60 mg%), sodium (35.16 mg%), (162.09 mg%), magnesium (13.95mg%),copper (0.057mg%), iron (0.053mg%), zinc (0.52 mg%).

To evaluate the shelf life products were stored in glass jars (yogurt) and glass bottles (*Lassi*) for 11 days and evaluated at fresh, 3rd, 5th, 7th, 9th and on 11th day. Sensory score was decreased throughout the storage period. The colour and appearance and consistency of all the flavoured probiotic *Lassi* and yogurt along with control non-significantly decreased throughout the storage period. Whereas, flavour, mouth feel and overall acceptability of all the flavoured probiotic *Lassi* and yogurt samples along control samples were significantly decreased throughout the storage period from the day of preparation to the 11th day of storage.

To determine the impact of probiotic product on the fasting blood glucose levels of diabetic type II patients, Rose flavour *Lassi* was selected. Sixty patients were alienated into two groups: Control group and experiment group. The experiment group was administrated 300 ml (fresh) probiotic *Lassi* in mid meal for 30 days. There was no significant difference in fasting blood glucose level in the control group. While, in the experiment group the mean blood glucose level was shown significant difference in fasting blood glucose level after the intervention of probiotic *Lassi*. Therefore, it could be concluded that probiotic *Lassi* was effective in managing the blood glucose levels and is suitable for diabetic patients to fulfill their cravings regarding sweetness.

Keywords: Probiotics, *Lactobacillus helveticus*, *Lassi*, Sensory profile, *Streptococcus thermophilus*, Yogurt.

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महाराणा प्रताप कृषि एवं प्रौद्योगिकी विश्वविद्यालय
सामुदायिक एवं व्यवहारिक विज्ञान महाविद्यालय, उदयपुर, राजस्थान
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विषय: "मधुमेह रोगियों पर बकरी के दूध आधारित प्रोबायोटिक उत्पादों का प्रभाव"

अनुक्षेपण

वर्तमान परिदृश्य में नवोन्मेषी फ्लेवर के साथ-साथ स्वास्थ्य और पोषण के प्रति जागरूकता भी बढ़ रही है। कार्यात्मक भोजन के रूप में माने जाने वाले प्रोबायोटिक्स के चिकित्सीय लाभों की एक विस्तृत श्रृंखला है। बकरी के दूध का उपयोग प्रोबायोटिक्स के वितरण के संभावित स्रोत के रूप में किया जा सकता है। मधुमेह का बढ़ता प्रसार प्रमुख वैश्विक स्वास्थ्य मुद्दों में से एक है। 2030 तक मधुमेह की बढ़ती व्यापकता 366 मिलियन तक पहुंचने का अनुमान है। लोगों को चीनी की अधिक खपत और बेहतर मिठास के साथ कम कैलोरी प्रदान करने वाले चीनी के विकल्प चुनने में दुविधा का सामना करना पड़ रहा है। इसलिए, बकरी के दूध आधारित किण्वित उत्पादों को तैयार करने के लिए वर्तमान अध्ययन की योजना बनाई गई थी, जिसमें विभिन्न फ्लेवर के साथ प्रोबायोटिक कल्चर लैक्टोबैसिलस हेल्वेटिकस एमटीसीसी 5463 स्टार्टर कल्चर स्ट्रेप्टोकोकस थर्मोफिलस एमडी2 और स्टीविया अर्क और पाउडर शामिल हैं। अनन्नास, केसर-इलाइची और गुलाब के फ्लेवर से लस्सी बनाई गई और केवड़ा, केसर-पिस्ता और रास्पबेरी के फ्लेवर से योगर्ट बनाया गया। प्रोबायोटिक लस्सी और योगर्ट एक कार्यात्मक भोजन के रूप में और संवेदी प्रोफाइल, भौतिक-रासायनिक गुणों, शोल्फ लाइफ और रक्त ग्लूकोज स्तर पर प्रभाव का विश्लेषण करते हैं। स्टीविया पाउडर और अर्क से तैयार लस्सी और योगर्ट दोनों की समग्र स्वीकार्यता के औसत स्कोर को देखकर, स्टीविया पाउडर से तैयार लस्सी और योगर्ट की तुलना में स्टीविया अर्क से तैयार लस्सी और योगर्ट ने अधिक अंक प्राप्त किए; हालांकि, नियंत्रण ने दोनों उत्पादों की तुलना में कम अंक प्राप्त किए। सभी नमूनों में उच्चतम स्कोर प्राप्त किया गया, यानी गुलाब का फ्लेवर और उसके बाद केसर इलायची और अनानास, जबकि योगर्ट में उच्चतम स्कोर केवड़ा, केसर पिस्ता और रसभरी था जिसे स्टीविया के अर्क से तैयार किया गया था। इसलिए स्टीविया के अर्क में शामिल उत्पादों को आगे के काम के लिए चुना गया। फिर भी, प्रोबायोटिक लस्सी और योगर्ट के सभी नमूनों में उच्चतम माध्य स्कोर (संवेदी प्रोफाइल) गुलाब के फ्लेवर द्वारा प्राप्त किया गया था।

गुलाब के फ्लेवर से तैयार लस्सी के नमूने के भौतिक रासायनिक गुणों में नमी की मात्रा (87.31%), वसा की मात्रा (3.57%), कार्बोहाइड्रेट की मात्रा (4.70%), राख की मात्रा (0.79%), कम ऊर्जा मूल्य (64.41Kcal), T.S. (12.52%), एसएनएफ (8.93%), वांछित पीएच (4.39), टिट्रेटेबल एसिडिटी (0.83%) और चिपचिपापन (314.70 सीपी)। मिनरल प्रोफाइल कैल्शियम (115.40 mg%), फॉस्फोरस (122.60 mg%), सोडियम (35.16 mg%), (162.09 mg%), मैग्नीशियम (13.95mg%), कॉपर (0.057mg%), आयरन (0.053) की उचित मात्रा दिखाता है मिलीग्राम%), जस्ता (0.52 मिलीग्राम%) शोल्फ लाइफ का मूल्यांकन

करने के लिए उत्पादों को 11 दिनों के लिए लस्सी को कांच की बोतलों में एवं योगर्ट को कांच के जार में संग्रहित किया गया और ताजा, तीसरे, पांचवें, सातवें, नौवें और ग्यारहवें दिन मूल्यांकन किया गया। भंडारण अवधि के दौरान संवेदी स्कोर कम हो गया था। सभी फ्लेवरयुक्त प्रोबायोटिक लस्सी और योगर्ट का रंग और दिखावट और गाढ़ेपन नियंत्रण के साथ पूरी भंडारण अवधि के दौरान महत्वपूर्ण रूप से कम नहीं हुआ। जबकि नियंत्रण नमूनों के साथ-साथ सभी फ्लेवर्ड प्रोबायोटिक लस्सी और योगर्ट के नमूनों का फ्लेवर, माउथ फील और समग्र स्वीकार्यता तैयारी के दिन से भंडारण के ग्यारहवें दिन तक भंडारण अवधि के दौरान काफी कम हो गई थी। मधुमेह के प्रकार II रोगियों के उपवास रक्त शर्करा के स्तर पर प्रोबायोटिक उत्पाद के प्रभाव को निर्धारित करने के लिए, गुलाब के फ्लेवर वाली लस्सी का चयन किया गया था। साठ रोगियों को दो समूहों में अलग कर दिया गया: नियंत्रण समूह और प्रयोग समूह। प्रयोग समूह को 30 दिनों के लिए मध्य भोजन में 300 मिलीलीटर (ताजा) प्रोबायोटिक लस्सी दी गई। नियंत्रण समूह में तेजी से रक्त शर्करा के स्तर में कोई महत्वपूर्ण अंतर नहीं था। जबकि, प्रयोग समूह में माध्य रक्त शर्करा स्तर प्रोबायोटिक लस्सी के हस्तक्षेप के बाद उपवास रक्त शर्करा स्तर में महत्वपूर्ण अंतर दिखाता है। इसलिए, यह निष्कर्ष निकाला जा सकता है कि प्रोबायोटिक लस्सी रक्त शर्करा के स्तर को प्रबंधित करने में प्रभावी थी और मधुमेह के रोगियों के लिए मिठास के प्रति उनकी लालसा को पूरा करने के लिए उपयुक्त है।

शब्द कुंजी : प्रोबायोटिक्स, योगर्ट, लस्सी, लैक्टोबैसिलस हेल्वेटिकस, संवेदी प्रोफ़ाइल, स्ट्रेप्टोकोकस थर्मोफिलस।

डॉ. सरला लखावत
मुख्य सलाहकार

जसप्रीत कौर
शोधकर्ता

APPENDIX-I
Sensory Profile of Products
College of Community and Applied Science

Name of Panel Member:

Date :

Product Name :

Sample No.	Colour and Appearance	Flavour	Mouth feel	Consistency	Overall acceptability

Remarks :

9 Point Hedonic Rating Scale for Score

Like Extremely	9
Like Very Much	8
Like Moderately	7
Like Slightly	6
Neither Like nor Dislike	5
Dislike Slightly	4
Dislike Moderately	3
Dislike Very Much	2
Dislike Extremely	1

Appendix-II

Preparation of media for microbial analysis

1. Phosphate buffer: Dissolved 38.2 g KH_2PO_4 in 1 L distilled water (a) and 50 g MgSO_4 in 1 L distilled water (b). 1.2 ml of solution (a) and 5 ml of solution (b) was mixed and volume was made upto 1000 ml. From the prepared solution 9 ml each was dispensed in to test tubes and autoclaved at 121°C for 15 min.

2. Total Lactic Agar: 63.5 g of dehydrated powder of lactic agar (Himedia Laboratories Pvt. Ltd., Mumbai, India), was suspended in 1000 ml of distilled water and allow them to stand for few min. The contents were heated with stirring to boiling temperature and held for 2 min. to dissolve the medium completely. It was aseptically distributed in the 100 ml quantities in sterilized flasks of 250 ml capacity and autoclaved at 121°C for 15 min. The medium was allowed to cool at 45 to 46°C and used for plating.

Composition of Lactic Agar

Ingredients g/l

Casein enzymic hydrolysate 20.0

Yeast extract 5.00

Gelatin 2.50

Dextrose 5.00

Lactose 5.00

Sucrose 5.00

Sodium chloride 4.00

Sodium acetate 1.50

Ascorbic acid 0.50

Agar 15.0

Violet Red Bile Agar (VRBA)

41.53 g of dehydrated powder of VRBA (Himedia Laboratories Pvt. Ltd., Mumbai, India), was suspended in 1000 ml of distilled water and allowed them to stand for few min. The contents was heated with stirring to boiling temperature and held for 2 min. to dissolve the medium completely. The pH of the medium was adjusted to 7.4 ± 0.1 . It was aseptically distributed in the 100 ml quantities in sterilized flasks of 250 ml capacity and autoclaved at 121°C for 15 min. The medium was allowed to cool at 45 to 46°C and used for plating (BIS IS: 5401-1969).

Ingredients g/l

Pectic digest of animal tissue 7.00

Yeast extract 3.00

Lactose 10.0

Bile salt mixture 1.50

Sodium chloride 5.00

Neutral red 0.03

Crystal violet 0.02

Agar 1.50

Potato Dextrose Agar

41.0 of dehydrated powder of PDA (Himedia Laboratories Pvt. Ltd., Mumbai, India), was suspended in 1000 ml of distilled water and allowed them to stand for few min. The contents were heated with stirring to boiling temperature and held for 2 min. to dissolve the medium completely. The pH of the medium was adjusted to 7.4 ± 0.1 . It was aseptically distributed in the 100 ml quantities in sterilized flasks of 250 ml capacity. The basal medium was sterilized at 121°C (15psi) for 15 min. At the time of

plating, the pH of the melted agar medium (60°C) was adjusted to 3.5 with 10 % sterilized tartaric acid (BIS IS: 5403-1969).

Ingredients g/l

Infusion from 300 g potatoes To make one liter

Dextrose 20.0

Agar 15.0

Appendix-III

Interview Schedule

Name:	
Address and Ph. No:	
Age	
Education	a) Literate b) Primary c) Middle d) Intermediate e) Graduation f) Post Graduation g) Technical degree
Occupation	a) Service (Private) b) Service (Govt) b) Business
Type of work:	a) Sedentary b) Moderate c) Hard
Type of Family	a) Joint b) Nuclear
No of family members	
Food habits:	a) Vegetarian b) Non –Vegetarian c) Ovo-Vegetarian
Physical Examination:	Weight _____ kg Height _____ cms. BMI.....
Biochemical Examination: Blood sugar level	Pre Feeding Day 1 _____ Day 2 _____ Day 3 Post Feeding Day 1 _____ Day 2 _____ Day 3
How long have you been in Diabetes mellitus	a) < 1 year b) 1-5 years c) >5 years
Family history	a) Positive b) Negative
Clinical Manifestation	a) Polyuria b) Polyphagia c) Polydypsia e) Delayed wound healing f) Weakness of eyes g) General weakness h) Weight loss i) Tiredness j) Pain in legs k) Any other
Treatment taken for the disease	a) Dietary modification only b) Oral hypoglyceamic drugs with dietary modification c) Insulin
Medication	a) Alopathic b) Ayurvedic c) Homeopathic
Sleep Time	
Physical exercise	Yes..... No..... if Yes Duration.....

DIETARY PATTERN BY USING THREE DAY RECALL METHOD:

Day 1			
Meal	Menu	Household amt.	Ing. Amt.
<u>Breakfast</u>			
<u>Mid Meal</u>			
<u>Lunch</u>			
<u>Evening Tea</u>			
<u>Dinner</u>			

<u>Day 2</u>			
<u>Meal</u>	<u>Menu</u>	<u>Household amt.</u>	<u>Ing. Amt.</u>
<u>Breakfast</u>			
<u>Mid Meal</u>			
<u>Lunch</u>			
<u>Evening Tea</u>			
<u>Dinner</u>			

<u>Day 3</u>			
<u>Meal</u>	<u>Menu</u>	<u>Household amt.</u>	<u>Ing. Amt.</u>
<u>Breakfast</u>			
<u>Mid Meal</u>			
<u>Lunch</u>			
<u>Evening Tea</u>			
<u>Dinner</u>			

Appendix-IV

INSTITUTIONAL ETHICAL COMMITTEE FOR HUMAN RESEARCH
Maharana Pratap University of Agriculture and Technology
Udaipur, Rajasthan

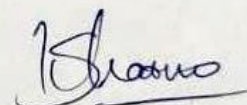
Ref. No.

MPUAT/DR/2017/428
Date: 24/2/20

ETHICAL COMMITTEE CERTIFICATE

TO WHOM IT MAY CONCERN

This is to certify that a clinical trial entitled "EFFECT OF GOAT MILK BASED PROBIOTIC PRODUCTS ON DIABETIC PATIENTS" is going to be started in this institution by Miss Jaspreet Kaur, Ph.D. Research Scholar, Department of Food Science and Nutrition, College of Community and Applied Sciences. This project was presented before the Institutional Ethical Committee for Human Research and there is no objection to conduct this work. The project is approved by the committee.



Dr. S. K. Sharma

Chairman

MPUAT, Udaipur, Rajasthan

Appendix-V



DEPARTMENT FOOD SCIENCE & NUTRITION
COLLEGE OF COMMUNITY AND APPLIED SCIENCES, UDAIPUR
Maharana Pratap University Of Agriculture And Technology, Udaipur

Dr. Sarla Lakhawat
Assoc.Prof.& Head

No.CCAS/FSN /2020/ 907

Date: 22-02-2020

To

The Dr. *Suresh Missaria*
Missaria Multi Specialist Hospital
Hanumangarh, Rajasthan

Through: Proper Channel

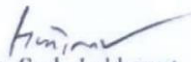
Subject: To seek cooperation and supervision for intervention work

Respected Ma'am / Sir

Greetings with respect, My student Jaspreet Kaur, Ph.D. (Research Scholar) working on research project entitled "Effect of goat milk based probiotic products on diabetes mellitus". In this intervention patients will be fed the *Lassi* which will be made from goat milk with *S. thermophilus* and *L. helveticus* and Stevia Extract. It is ensure that patient will not suffer from any adverse effect on health from intake of this *Lassi* which approved by university ethical committee. So to conduct the research she needs your cooperation and supervision. This is humble request to you to provide her the same.

Thanking you

Your's sincerely


Dr. Sarla Lakhawat
Associate Professor and Head,
Department of Food Science and Nutrition,
College of Community and Applied Sciences
Maharana Pratap University of Agriculture and Technology,
Udaipur, Rajasthan

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HEAD
DEPARTMENT OF FOOD SCIENCE & NUTRITION
COLLEGE OF COMMUNITY & APPLIED SCIENCES
MPUAT, UDAIPUR

Appendix-VI

जसप्रीत कौर
पीएच.डी. छात्रा, खाद्य व पोषण विभाग
सामुदायिक एवं व्यावहारिक विज्ञान महाविद्यालय, उदयपुर

सादर नमस्कार

मैं सामुदायिक एवं व्यावहारिक विज्ञान महाविद्यालय के खाद्य व पोषण विभाग की विद्यावाचस्पति की छात्रा हूं। इस शोध कार्य की सफलता आपके सहयोग पर निर्भर करती है।

प्रोबायोटिक्स व बकरी के दूध का मधुमेह नियन्त्रण में लाभकारी प्रभाव देखा गया है। इस अध्ययन के अन्तर्गत मैं यह ज्ञात करना चाहती हूं कि मधुमेह की स्थिति में बकरी के दूध व प्रोबायोटिक्स से बनी लस्सी का रक्त शर्करा पर क्या प्रभाव पड़ता है।

मेरा आपसे यह निवेदन है कि अध्ययन के दौरान मुझे पूर्ण सहयोग करें एवं इस अध्ययन में भाग लेने के लिए अपनी सहमति निम्नलिखित पत्र में अंकित करें।

धन्यवाद!

जसप्रीत कौर

प्रिय जसप्रीत कौर

मुझे आपके द्वारा किये जा रहे अनुसंधान के बारे में आवश्यक जानकारी प्रदान की गई है। मैं इस शोध कार्य में भाग लेने के लिए सहमत हूं। मेरा पता निम्न है :-

नाम:-.....

पता:-.....

दूरभाष:-.....

मधुमेह की विस्तृत जानकारी



जसप्रीत कौर एवं डॉ. सरला लखावत



सामुदायिक एवं व्यवहारिक विज्ञान महाविद्यालय

महाराणा प्रताप कृषि एवं प्रौद्योगिकी विश्वविद्यालय, उदयपुर (राज.)

लेखक के बारे में

डॉ. सरला लखावत वर्तमान में सामुदायिक एवं व्यवहारिक विज्ञान महाविद्यालय, महाराणा प्रताप कृषि एवं प्रौद्योगिकी विश्वविद्यालय में आहार विज्ञान एवं पोषण में प्रमुख व सहआचार्य पद पर कार्यरत हैं। आपको 10 साल के शोध व शिक्षण अनुभव हैं। आपके द्वारा शोध पत्रों को विभिन्न राष्ट्रीय व अन्तर्राष्ट्रीय शोध पत्रिकाओं में प्रकाशित किया गया है। आपके द्वारा स्वामी सहजानन्द सरस्वती पुरस्कार (ICAR) राष्ट्रीय स्तर पर पुरस्कार प्राप्त किया गया एवं शिक्षण के साथ ही आप क्षेत्रिय कार्यों में पारंगत हैं।

जसप्रीत कौर वर्तमान में सामुदायिक एवं व्यवहारिक विज्ञान में रिसर्च स्कॉलर हैं। आपके बीएस.सी. (B.Sc.) स्वामी केशवानन्द राजस्थान कृषि विश्वविद्यालय बीकानेर एवं एमएस.सी. (M.Sc.) सरदार कृषि नगर दातीवाड़ा कृषि विश्वविद्यालय बनासकांठा गुजरात से की गईं। आपने नेस्ले (Nestle) में मातृ पोषण विशेषज्ञ पद पर लगभग 2 वर्ष तक कार्य किया गया।

अनुक्रमणिका

क्र.स.	विषय
1.	प्रस्तावना
2.	मधुमेह के प्रकार
3.	मधुमेह के कारण
4.	मधुमेह के लक्षण
5.	जांच करने के तरीके
6.	डायबिटीज का प्रभाव
7.	ग्लाइसेमिक इन्डेक्स ;ळपद्ध
8.	आर्टीफिशियल एवं नेचुरल स्वीटर व मधुमेह
9.	प्राकृतिक मिठास वाले खाद्य पदार्थ
10.	प्रबंधन
11.	आहार पर ध्यान दें
12.	पोषण गणना
13.	आहार तालिका

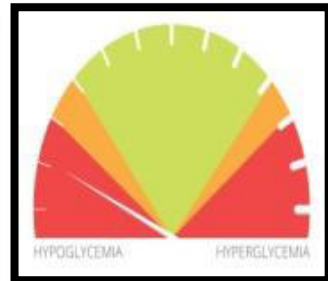
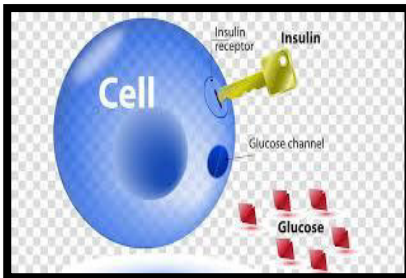
प्रस्तावना

भारत में विश्व दूसरी सबसे बड़ी आबादी वाला देश है। धरती पर रहने वाला हर 6 में से 1 मनुष्य भारत में रहता है। विश्व के कुल मधुमेह रोगियों में से हर 6 में से एक रोगी भारत से है जो कि चीन के बाद दूसरे नम्बर पर है। इन आंकड़ों से समस्या की गंभीरता को समझना आसान है। शहरी क्षेत्रों में इसकी व्यापकता ग्रामीण क्षेत्रों की तुलना से अधिक है। मधुमेह का इतना प्रसार होने से इसकी जागरूकता बहुत कम है।



मधुमेह एक दीर्घकालिक चयापचयी विकार (Metabolic Syndrome) है जिसमें शरीर की शर्करा को उपयोग की क्षमता आंशिक या पूर्ण रूप से विकृत हो जाती है। मधुमेह में रक्त की शर्करा का स्तर बढ़ जाता है एवं कार्बोहाइड्रेट के चयापचय विकृत होने से वसा व प्रोटीन का चयापचय भी प्रभावित होता है।

*Protect You and
your Family from
Diabetes*

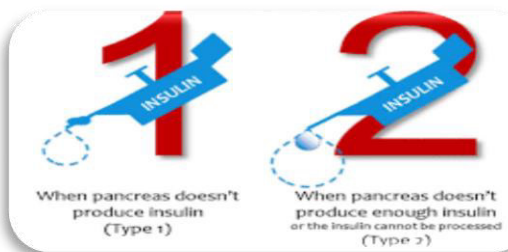


मधुमेह के प्रकार

प्रकार I: इस किशोरावस्था या बाल्यावस्था का मधुमेह भी कहा जाता है। जिसमें प्रभावित व्यक्ति इन्सुलिन पर निर्भर होता है क्योंकि अग्नाशय पर्याप्त मात्रा में इन्सुलिन का उत्पादन नहीं कर पाता। अग्नाशय का किसी कारण से क्षतिग्रस्त होना जैसे कोई दुर्घटना या वायरस इन्फेक्शन इत्यादि कारणों से हो सकता है। इससे प्रभावित रोगियों में कम वजन की समस्या व एसिडोसिस सामान्य है।

प्रकार II: इस प्रकार का मधुमेह धीरे-धीरे विकसित होता है। इसमें इन्सुलिन तो स्त्रावित होता है परन्तु इसका पर्याप्त उपयोग नहीं हो पाता। यह मुख्यतः व्यस्क लोगों में होता है इससे एसिडोसिस सामान्यतः नहीं होता है।

Over 50% of type 2 diabetes is preventable)



कुपोषण सम्बन्धी मधुमेह : कुपोषण के कारण अग्नाशय पर्याप्त मात्रा में इन्सुलिन स्त्रावित नहीं कर पाता जिसके कारण रक्त में शर्करा का स्तर बढ़ जाता है।

गर्भावस्था सम्बन्धी मधुमेह : यह मुख्यतः गर्भावस्था के 24वें से 28वें सप्ताह के मध्य विकसित होता है। इसमें अग्नाशय आवश्यकतानुसार इन्सुलिन का उत्पादन नहीं कर पाता जिससे रक्त में शर्करा का स्तर बढ़ने लगता है।

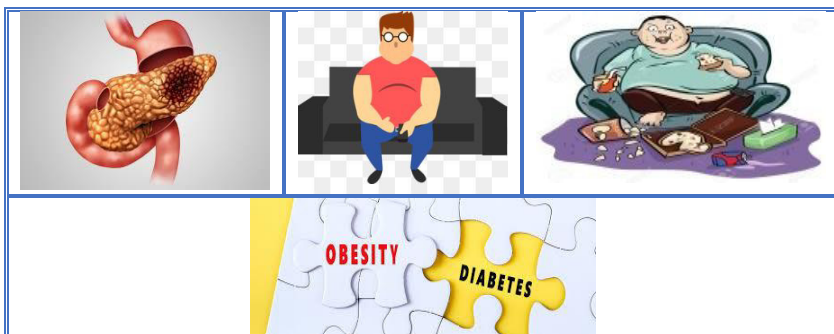


मधुमेह के कारण

आनुवांशिकी : माता-पिता या परिवार में रक्त सम्बन्धी में मधुमेह होने से इसकी संभावना बढ़ जाती है।



वातावरणीय कारण (इन्फेक्शन) : कुछ वायरस इन्फेक्शन के कारण कम उम्र में अग्नाशय का क्षतिग्रस्त हो जाने से इन्सुलिन का स्रावण नहीं हो पाता है। रोग प्रतिरोधक कारक रोग प्रतिरोधक क्षमता में कमी कारण इन्सुलिन ठीक प्रकार से कार्य नहीं कर पाता अतः रक्त में शर्करा का स्तर बढ़ जाता है।



मोटापा : फैट सेल्स हार्मोन के साथ कुछ जलन पैदा करने वाले रसायनों का उत्पादन करते हैं एवं नान एस्टेरीफाइड फैटी एसिड जारी करते हैं जो कि चपापचय को प्रभावित करते हैं।

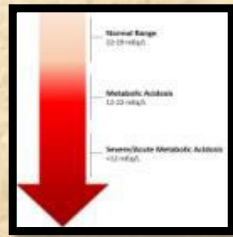
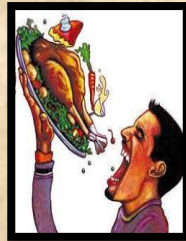
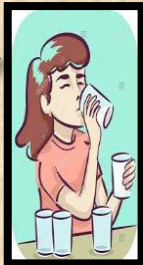
BMI बॉडी मास इन्डेक्स

वर्गीकरण	बीएमआई (किलोग्राम/मी ²)
कम वजन	< 18.5
सामान्य वजन	18.5 - 22.9
ज्यादा वजन	23-24.9
मोटापा 1	25-29.9
मोटापा 2	>30

रहन-सहन : आरामदायक जीवन यापन व व्यायाम न करना, समय पर न खाना, जंक फूट खाना इत्यादि रक्त की शर्करा के स्तर को व इन्सुलिन की कार्यक्षमता को प्रभावित करते हैं।

मधुमेह के लक्षण

- ✚ रक्त में शर्करा का स्तर अधिक होना, पेशाब में शर्करा का उपस्थित होना, बहुत ज्यादा प्यास लगाना, भूख लगना, थकान, वजन कम होना, बार-बार पेशाब आना,
- ✚ एसिडोसिस
- ✚ आँखों की रोशनी कम होना,
- ✚ घाव भरने में समय लगाना।



जांच करने के तरीके

फास्टिंग प्लाज्मा ग्लूकोज टेस्ट : इस जांच में रक्त में शर्करा का स्तर भूखे पेट किया जाता है जिसमें व्यक्ति ने कम से कम 8 घण्टे तक कुछ न खाया हो। इस जांच में किस अंगुली के सिरे से खून निकाला जाता है उसे ग्लूकोमीटर में लगे स्ट्रिप पर डाला जाता है। निम्न आंकड़ों से पता चलता है कि शर्करा स्तर कितना है –



स्थिति	शुगर की मात्रा (डी.एल. में)
सामान्य	100
प्री-डायबीटीज	100-125
डायबीटीज	126 से ज्यादा

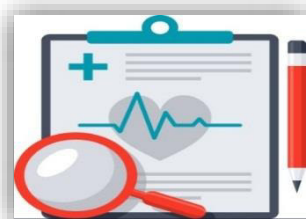
खाने के बाद रक्त में शर्करा का स्तर : इसमें रक्त शर्करा का स्तर नाश्ता करने या कुछ खाने के 2 घंटे के बाद किया जाता है। इसमें किसी अंगुली के सिरे से खून निकाल कर ग्लूकोमीटर में लगी स्ट्रिप पर डाला जाता है।

स्थिति	शुगर की मात्रा (डी.एल. में)
सामान्य	140 से कम
प्री-डायबीटीज	141 से 199
डायबीटीज	200 से ज्यादा

ओरल ग्लूकोज टोलरेंस टेस्ट : इस टेस्ट में किसी व्यक्ति को फास्टिंग के बाद ग्लूकोज दिया जाये तो उसका शरीर किस तरह की प्रतिक्रिया देता है। इसे भी ग्लूकोमीटर की मदद से ही जांच जाता है तथा 2 घंटे के पश्चात्

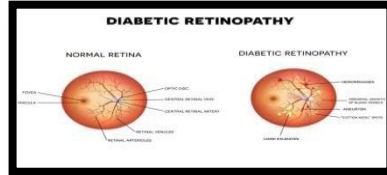
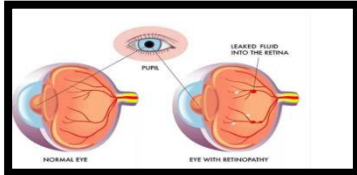
स्थिति	शुगर की मात्रा (डी.एल. में)
प्री-डायबीटीज	141 से 199
डायबीटीज	200 से ज्यादा

रेण्डम प्लाज्मा ग्लूकोज टेस्ट : इसमें खाने पीने के वक्त की कोई पाबन्दी नहीं होती है। इसकी जांच भी ग्लूकोमीटर की सहायता से की जाती है। अगर 200 से स्तर होता है तो डायबीटीज की जांच को सकारात्मक दिखाता है।



डायबिटीज का प्रभाव

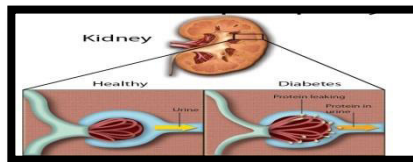
आंख : रक्त में शर्करा के स्तर को नियमित बढ़े रहने से नजर धुंधली होने लगती है। समय पर ध्यान न देने पर मोतियाबिंद होने के आसार बढ़ जाते हैं। आंख को प्रभावित करने वाली इस गंभीर समस्या को डायबिटिक रेटिनोपैथी कहते हैं।



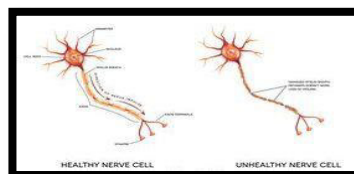
त्वचा : डायबिटीज को रोगियों में त्वचा पर संक्रमण होना बहुत आम है। जिसमें त्वचा शुष्क हो जाती व इस पर चकते पड़ जाते हैं जिसे सेबोरिक केरेटोसिस के नाम से जाना जाता है।



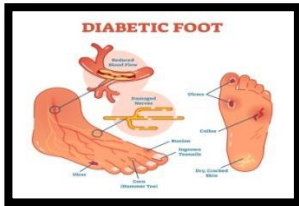
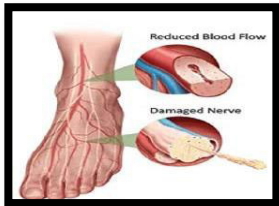
किडनी: शर्करा के बढ़े स्तर से किडनी पर अनावश्यक भार जिससे किडनी खराब होने अथवा फेल होने का खतरा बढ़ जाता है।



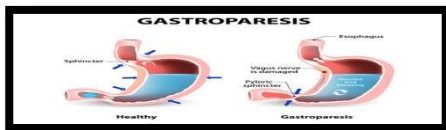
तंत्रिका क्षति : रक्त में शर्करा का स्तर बढ़ने से तंत्रिका क्षति होने लगती है। जिसे न्यूरोपैथी कहते हैं।



पैरो पर : मधुमेह में छोटे व बड़ी दोनों प्रकार की रक्त वाहिनियों में रक्त की आपूर्ति कम हो जाती है जिससे पैर की संरचना विकृत होने लगती है।



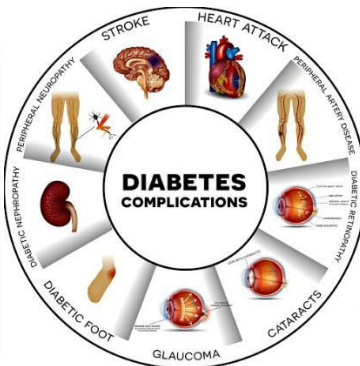
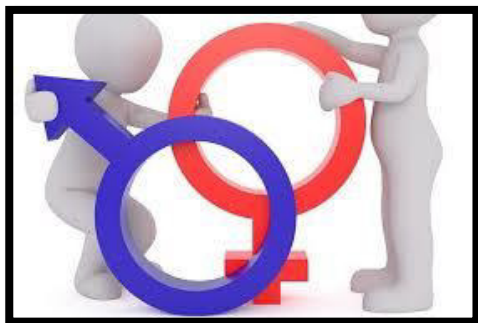
पाचन तंत्र : तंत्रिका तंत्र में क्षति होने से गेस्ट्रोपेरेसिस की स्थिति उत्पन्न होती है। गेस्ट्रोपेरेसिस तब होता है जब पेट से भोजन को छोटी आंत में स्थानान्तरित करने में तंत्रिका तंत्र पाचन तंत्र की क्षमता में हस्तक्षेप करता है।



रक्त प्रवाह तंत्र : मधुमेह उच्च रक्तचाप के जोखिम को बढ़ाता है। रक्त में अधिक शर्करा से हृदय पर अधिक दबाव बढ़ता है जिससे रक्त वाहिनियों की दिवारों पर वसा के प्रभाव का जोखिम बढ़ जाता है एवं हृदयरोग होने का खतरा बढ़ जाता है।



प्रजनन प्रणाली : मधुमेह का महिला व पुरुष दोनों की प्रजनन प्रणाली पर नकारात्मक प्रभाव पड़ता है।



ग्लाइसेमिक इन्डेक्स(GI)

यह रक्त शर्करा के स्तर पर कार्बोहाइड्रेट के प्रभाव का माप है। जिसे रक्त शर्करा की मात्रा बढ़ाने के अनुसार विभिन्न अंकों द्वारा निरूपित किया जाता है।

कैसे समझे ग्लाइसेमिक इन्डेक्स : ग्लाइसेमिक इन्डेक्स एक प्रकार का अंक है जिसकी गणना आसानी से की जा सकती है। सफेद ब्रेड या ग्लूकोज को रेफरेंस फूड के रूप में उपयोग किया जा सकता है।

उदाहरण : किसी व्यक्ति को 50 ग्राम ग्लूकोज दिया जाये एवं निश्चित समयानुसार (2 घंटे पश्चात) रक्त में उपस्थित ग्लूकोज शर्करा को ग्लूकोमीटर की सहायता से जांच कर लें। इसका उपयोग दूसरे खाने की मात्रा के रक्त शर्करा के स्तर से भाग देकर 100 से गुणा करने पर उस खाने का ग्लाइसेमिक इन्डेक्स ज्ञात किया जा सकता है।

वर्गीकरण

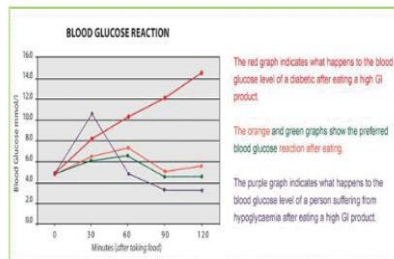
वर्गीकरण	जीआई विस्तार	उदाहरण
निम्न जीआई	55 या उससे कम	ज्यादातर फल सब्जियां, मोटा अनाज, फलियां, दाले दूध दही, बादाम इत्यादि
मध्यम जीआई	55-69,	चावल, कंद
उच्च जीआई	70 व इससे अधिक	पके आलू, तरबूजा, फ्रेंच रोल, सफेद, ब्रेड, ग्लूकोज इत्यादि

लाभ : ग्लाइसेमिक इन्डेक्स ज्ञात होने पर कम ग्लाइसेमिक इन्डेक्स वाले भोज्य पदार्थों के सेवन से रक्त शर्करा का स्तर नियमित किया जा सकता है।

Glycemic Index

Low GI (<55), Medium GI (56-69) and High GI (70+)

Grains / Starches	Vegetables	Fruits	Dairy	Proteins
Rice Bran 27	Asparagus 15	Grapefruit 25	Low-Fat Yogurt 14	Peanuts 21
Rice-Cereal 42	Broccoli 15	Apple 38	Plain Yogurt 14	Beans, Dried 40
Spaghetti 42	Celery 15	Peach 42	Whole Milk 27	Lentils 41
Wild Rice 54	Cucumber 15	Orange 44	Soy Milk 30	Licury Beans 41
Sweet Potatoes 57	Lettuce 15	Grape 46	Fat Free Milk 32	Split Peas 45
White Rice 61	Peppers 15	Banana 54	Skim Milk 32	Lima Beans 46
Whole Wheat Bread 64	Spinach 15	Mango 56	Chocolate Milk 35	Chickpeas 47
Corn Cobs 65	Tomatoes 15	Pineapple 66	Fruit Yogurt 36	Pinto Beans 55
Whole Wheat Bread 71	Chickpeas 33	Watermelon 72	Ice Cream 61	Black-Bean 59
Muesli 80	Cooked Carrots 39			
Baked Potatoes 85				
Oatmeal 87				
Taco Shells 97				
White Bread 100				
Bagel, White 103				



Source: GI Foundation SA endorsement

आर्टिफिशियल एवं नेचुरल स्वीटर व मधुमेह




चीनी का विकल्प आर्टिफिशियल स्वीटर व प्राकृतिक मीठी चीजें दोनों हो सकती हैं। आजकल मधुमेह रोगियों में आर्टिफिशियल स्वीटर का प्रचलन काफी बढ़ गया है। जो कि सामान्य उपयोग होने वाली चीनी से कई सौ गुणा ज्यादा मीठे होते हैं। एफडीए (FDA) ने कुछ आर्टिफिशियल स्वीटनटर को इंसानों के लिए सुरक्षित बताया है। परन्तु इन्हें सीमित मात्रा में उपयोग करने की सलाह दी गई है।

मुख्य आर्टिफिशियल स्वीटर

एस्पार्टेम		यह सामान्य शुगर से 200 गुणा ज्यादा मीठा होता है। इसके एमिनो एसिड, एसपार्टिक एसिड, एथानाल जैसे घटक सूक्ष्म मात्रा में मेटाबोलाइज हो जाते हैं। प्रतिदिन मात्रा : 50 mg/kg/day उपलब्ध ब्रांड : शुगर फ्री गोल्ड
सेकरीन :		यह सामान्य चीनी के मुकाबले से 300 गुणा ज्यादा मीठा होता है। प्रतिदिन मात्रा : 5उहधहधकंल उपलब्ध ब्रांड : स्वीट एड लो, शुगर फ्री।
सुक्रालोज :		यह चीनी की तुलना से 600 गुणा ज्यादा मीठी होती है। इसका उपयोग आईसक्रीम, डेयरी व ठण्डे पेय पदार्थ बनाने में उपयोग किया जाता है। प्रतिदिन : 5mg/kg/day उपलब्धब्रांड : स्पलेण्डा।
ऐसेसल्फेम :		इसकी घुलनशीलता अच्छी होती है। यह चीनी की तुलना में 200 गुणा ज्यादा होती है। उपलब्ध ब्रांड : स्वीट वन प्रतिदिन मात्रा : 15 mg/kg/day
नियोटोम :		नियोटोम चीनी की तुलना में 8000 गुणा ज्यादा मीठा होता है। प्रतिदिन मात्रा : 18mg/kg/day





लगातार सिंथेटिक स्वीटर के बढ़ते उपयोग में इनकी निर्धारित मात्रा की ओर विशेष ध्यान देना चाहिए। अन्यथा इनके दुष्परिणाम भी हो सकते हैं।

प्राकृतिक मिठास वाले खाद्य पदार्थ

गुड :		कम मात्रा में गुड का उपयोग चीनी के स्थान पर किया जा सकता है। इसमें आयस व मैगनीज जैसे जरूरी तत्व भी मौजूद होते हैं।
शहद :		प्राकृतिक स्रोतों में शहद चीनी का अच्छा विकल्प है। इसका ग्लाइसेमिक इन्डेक्स 55 है जो कि चीनी से कम होता है। स्टीविया यह मीठे के प्राकृतिक विकल्प के लिए जाना जाता है।
स्टिविया :		स्टिविया जिसे मुधरगुणा, स्वीटलीफ, शुगर लीफ व मीठी तुलसी नाम से जाना जाता है। यह एक शून्य कैलोरी हर्ब (Herb) है जो कि खाने में बिना कैलोरी प्रदान किये चीनी का स्वाद दे सकता है। यह साधारण चीनी से 200 गुणा अधिक मीठा होता है। 2015 में FSSAI ने स्टीविया को मीठे के तौर पर दूध व दूध से बनी मिठाईयां, दही, फ्लेवर्ड ड्रिंक इत्यादि में उपयोग को मंजूरी दी है। हाल ही हुए शोधों से पता चलता है कि टाइप 2 डायबिटीज से ग्रसित लोगों को स्टीविया का सेवन कराने से शर्करा के स्तर में कमी पाई गई है।

हाइपोग्लाइसेमिया : जब रक्त में शर्करा की मात्रा 70 मिग्रा/डेसी या 3.9 मिग्रा/ली से नीचे हो जाती है। इस अवस्था को निम्न रक्त शर्करा (हाइपोग्लाइसेमिया) कहते हैं।



हाइपोग्लाइसेमिया के कारण

बहुत अधिक इन्सुलिन एवं मधुमेह की दवाइयाँ लेना	→	
खाना न खाना	→	
सामान्य से अधिक व्यायाम करना	→	
शराब पीना	→	

हाइपोग्लाइसेमिका के शुरूआती लक्षण

अस्थिरता	चक्कर आना	पसीना आना	सरदर्द
			
ज्यादा भूख लगना	चिड़चिड़ापन	घबराहट	
			

गंभीर लक्षण

मांसपेशियों में कमजोरी	बोलने में कठिनाई	धुंधली या दोहरी दृष्टि	बेहोशी तथा मौत
			

निवारण :

- ✚ भोजन या नाश्ता न छोड़े,
- ✚ नाश्ता देर से न करे,
- ✚ इन्सुलिन व दवाईयां जरूरत से ज्यादा न ले।
- ✚ रक्त शर्करा को लगातार जांच करावें,
- ✚ दर्वाइया या इन्सुलिन लेने के पश्चात् हल्का नाश्ता ले,
- ✚ शराब ना लें, अगर शराब ले तो उसके साथ कुछ खाये,
- ✚ कम रक्त शर्करा प्रतिक्रियाओं को रिकार्ड करें।



क्या करें :

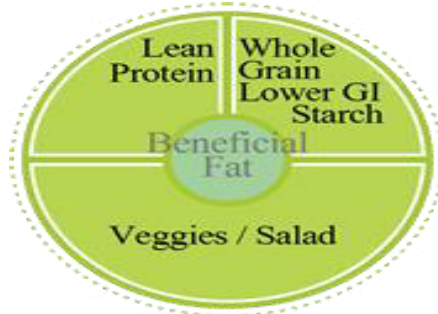
- ✚ मधुमेह की मूल जानकारी रखे,
- ✚ मधुमेह के कारण व प्रबंधन के बारे में जाने
- ✚ निश्चित समय पर चिकित्सक से मिले,
- ✚ चिकित्सक द्वारा बताई दर्वायों व इन्सुलिन लें,
- ✚ थोड़ा खायें व थोड़ी देर में खायें।
- ✚ वजन ज्यादा हो तो कम करें,

प्रबंधन

मधुमेह प्रबन्ध एक नजर में

योगा एवं व्यायाम	दवाईयाँ	कार्बोहाइड्रेट गणना	संतुलित आहार
			
रक्त ग्लूकोज मॉनिटरिंग	मद्यपान व धूमपान न करें।	खाना न छोड़े व खाने में बहुत देर न करें।	
			

आहार में परिवर्तन करके : आहार में परिवर्तन करके मधुमेह को नियंत्रित किया जा सकता है।



कैलोरी : मधुमेह से प्रभावित व्यक्ति को लम्बाई, आयु, शारीरिक वजन के अनुसार प्रस्तावित ऊर्जा जरूरत 5 प्रतिशत कम दिया जाना चाहिए। कैलोरी को जटिल कार्बोहाइड्रेटस स्रोतों से पूरा किया जाना चाहिए।

प्रोटीन : मधुमेह की स्थिति में नकारात्मक नाइट्रोजन संतुलन हो जाता है। अतः अधिक प्रोटीन की आवश्यकता होती है। इसलिए वजन के अनुसार प्रोटीन की आवश्यकता से लगभग दुगुनी मात्रा में प्रोटीन देना चाहिए।

विटामिन एवं खनिज: मधुमेह की स्थिति में खनिज लवण व विटामिन की आवश्यकता बढ़ जाती है। अतः अधिक फल सब्जियों का सेवन करना चाहिए व आवश्यकता लगने पर मल्टी विटामिन की गोली दी जानी चाहिए।

आहार पर ध्यान दें

✚ क्या लें

कार्बोहाइड्रेट

गेहूँ	दलिया	रागी	बाजरा
			
बाजरा	क्विनोआ	ज्वार	
			

वसा

सरसों का तेल	तिल का तेल	सोयाबीन तेल	मूंगफली का तेल
			
जैतन का तेल			मछले का तेल
			

सब्जियाँ

पालक	मेथी	टमाटर	गाजर
			

मूली	धनिया	बैंगन	लौकी
			
कद्दू	गोभी	करेला परवल	फलियाँ
			
ग्वारफली	मटर	सेमफली	भिण्डी
			
खीरा	ककड़ी	चुकन्दर	
			

दालें

मूंग	मसूर	चना	मौठ
			
उड़द	राजमा	तुअरदाल	चवलां
			

दूध

कम वसा का दही	दूध	पनीर	लस्सी
			

क्या न लें

कार्बोहाइड्रेट

मैदा	सफेद ब्रैंड	चावल	तला हुआ खाना
			
समोसा	कचौड़ी	लड्डू	प्रोसेस्ड सीरियलस
			

फल

केला	चीकू	सीताफल	आम
			

न लें

जेम जैली	चीनी मिले हुए ज्यूस इत्यादि
	

सब्जियाँ

आलू	शक्कर कंद	तली हुई सब्जियां	साबूदाना
			

ज्यादा वसा वाले दूध व दूध से बने पदार्थ

मलाई	ज्यादा वसा का दूध	दही	पनीर
			

वसा

वनस्पति घी	पाम तेल	लार्ड	मार्जरीन
			

अन्य

सोडा	शराब	कैफीन	टेबल सॉल्ट	शुगर
				

पोषण गणना

- वजन के अनुसार

BMI बॉडी मास इन्डेक्स :

वर्गीकरण	बीएमआई (किलोग्राम/मी ²)
कम वजन	< 18.5
सामान्य वजन	18.5 - 22.9
ज्यादा वजन	23-24.9
मोटापा 1	25-29.9
मोटापा 2	>30

- कार्य के अनुसार

आहार तालिका

कार्य सेडेंटरी कैलोरी 1800		
समय	आहार	मात्रा
उठने के बाद	गर्म पानी मैथी का पानी नींबू शहद पानी	1 गिलास 1 गिलास 1 गिलास
नाश्ता	चाय या दूध या दही या छाछ + दलिया या उबला अण्डा या पराठा (मैथी, पालक, बथुआ, गोभी, इत्यादि) कम तेल का आमलेट	1 कप 1 गिलास 1 कटोरी 1 गिलास 1 कटोरी 1 अण्डा 1 पराठा
मध्य भोजन	छाछ या नींबू पानी या मौसमी फल या अंकुरित चाट या	1 गिलास 1 गिलास 1 फल 1 प्लेट
दोहपर का भोजन	सब्जी (मौसमी) दाल रोटी दही या रायता (मिश्रित अनाज की) / चटनी / चावल / सलाद	1 कटोरी 1 कटोरी 2 रोटी 1 कटोरी
शाम की चाय	मिश्रित अनाज के बिस्किट / भुने-चनेभुनी हुई मुंगफली चाय / कॉफी / दूध	50 से 80 ग्राम 1 कप
रात का खाना	रोटी सब्जी दाल +सलाद	2 रोटी 1 कटोरी 1 कटोरी
सोते समय	दूध + इसबगोल का छिलका	1 गिलास 1 चम्मच