DEVELOPMENT OF LOW FAT FIBER ENRICHED MASCARPONE CHEESE

THESIS SUBMITTED TO THE

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FOR THE DEGREE OF

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IN

DAIRY TECHNOLOGY

By

ZADE SANDIP BAPU

B. Tech. (Dairy Technology)

DAIRY TECHNOLOGY SECTION

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This is to certify that the thesis entitled, "**DEVELOPMENT OF LOW FAT FIBER ENRICHED MASCARPONE CHEESE**" submitted by **ZADE SANDIP BAPU** towards the partial fulfillment for the award of the degree of **M. Tech.** in **DAIRY TECHNOLOGY** of the **ICAR-NATIONAL DAIRY RESEARCH INSTITUTE (Deemed University)**, Karnal (Haryana), India, is a bonafide research work carried out by him under my guidance and no part of the thesis has been submitted for any other degree or diploma.

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Dedicated to My Loving Family

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ABSTRACT

Mascarpone is a soft and unripened variety of Italian cheese newly introduced in the Indian cheese market. It contains high fat (~40%) with high spreadable property and can be used as substitute of butter. Consumption of high-fat dairy products is linked with cardio vascular disease. Cheese does not contain any fiber but role of ditery fiber in health is well known. Considering the ill effects of high fat consumption and beneficial effects of fiber, an attempt was made to standardize the manufacturing process of fibre enriched, low fat Mascarpone cheese with suitable fat replacer.

Among three stages of fat addition, 50% fat addition in milk and 50% fat in coagulum resulted in higher yield with better spreadability than its addition in milk before coagulation or after coagulation. Four per cent lactic coagulation showed 2.0 -2.5% more yield and lowest fat loss in whey (about 0.85%) and highest mean spreadability value (3.35 N. sec^{-1}) as measured by Texture Analyzer among the three acids namely acetic acid, citric acid and lactic acid. Increase in fibre (Inulin) incorporation exhibited an increasing spreadability and decreasing moisture content in cheese up to 8%. Mascarpone cheese containing 16, 20 and 24% fat (corresponding to 60, 50 & 40% fat reduction) was prepared and compared with control cheese with 40% fat. The influence of fat replacers in combination of Inulin, WPC and N-creamer was studied by adding fat replacer to the reduced fat cheese on weight basis in 3 combinations: 10% (6% inulin, 2% WPC & 2% N-creamer), 15% (6% inulin, 4.5% WPC & 4.5%N-creamer), 20% (6% inulin, 7% WPC & 7% N-creamer). The results indicated that in general low-fat cheese samples had higher moisture contents. However, as the level of incorporation of combined fat replacer increased, the moisture content in the cheese decreased. The pH of Mascarpone cheese showed a direct relation to the quantity of fat replacer used in the cheese; as the quantity increased pH value of the cheese also increased. The mean spreadability of 1.83 (N.sec^{-1}) in Mascarpone cheese containing 16% fat (60% fat reduction) added with 10% fat replacers was more than that of cheese made with 24 and 20% fat (40% and 50% fat reduction) and it was very similar to that of control cheese. Shelf life studies showed that fat replaced sample had 2 days more than that of control sample when packed in polyethylene pouch and stored at 5-8^{0}C.

सारांश

Mascarpone नव भारतीय पनीर बाजार में पेश इतालवी पनीर (Cheese) का एक नरम और Unripened किस्म है। यह उच्च Spreadable संपत्ति के साथ उच्च वसा (~ 40%) शामिल है और मक्खन के विकल्प के रूप में इस्तेमाल किया जा सकता है। उच्च वसा वाले डेयरी उत्पादों की खपत कार्डियो वैस्कुलर रोग के साथ जुड़ा हुआ है। पनीर (Cheese) किसी भी फाइबर शामिल नहीं करता है, लेकिन स्वास्थ्य में आहार फाइबर की भूमिका अच्छी तरह से जाना जाता है। उच्च वसा की खपत और फाइबर के लाभदायक प्रभाव के दुष्परिणामों को देखते हुए, एक प्रयास समृद्ध फाइबर के निर्माण की प्रक्रिया का मानकीकरण करने के लिए बनाया गया था, Inulin तरह उपयुक्त वसा Mimeticके साथ कम वसा Mascarpone पनीर (Cheese), एन क्रीमर और मट्ठा प्रोटीन ध्यान (डब्ल्यूपीसी)।

वसा (Fat) अलावा के तीन चरणों के अलावा, रक्तकण में दूध और 50% वसा में 50% वसा अलावा जमावट से पहले या जमावट के बाद दूध में अपनी अलावा तुलना में बेहतर Spreadabilityसाथ उच्च उपज में हुई। प्रतिशत लैक्टिक जमावट प्रति चार एसिटिक एसिड के तीन एसिड के बीच बनावट विश्लेषक द्वारा मापा के रूप में 2.0 -2.5% अधिक उपज और मट्ठा (लगभग 0.85%) और उच्चतम मतलब Spreadability मूल्य (3.35 एन धारा -1) में सबसे कम वसा हानि से पता चला है, साइट्रिक एसिड और लैक्टिक एसिड। फाइबर (Inulin) समावेश में वृद्धि 8% करने के लिए पनीर में एक Spreadabilityबढ़ रही है और घटते नमी सामग्री का प्रदर्शन किया। (60, 50 और 40% वसा में कमी करने के लिए इसी) 16, 20 और 24% वसा युक्त Mascarpone पनीर तैयार है और 40% वसा के साथ नियंत्रण पनीर के साथ तुलना में किया गया था। 10% (6% Inulin, 2% डब्ल्यूपीसी और 2% एन क्रीमर: Inulin, डब्ल्यूपीसी और एन क्रीमर के संयोजन में वसा replacers के प्रभाव 3 संयोजन में वजन के आधार पर कम वसा पनीर के लिए वसा Replacer जोड़कर अध्ययन किया गया था), 15% (6% Inulin, 4.5% डब्ल्यूपीसी और 4.5% एन क्रीमर), 20% (6% Inulin, 7% डब्ल्यूपीसी और 7% एन क्रीमर)। परिणामों सामान्य कम वसा वाले पनीर के नमूने में उच्च नमी सामग्री को संकेत दिया कि। संयुक्त वसा Replacer के समावेश के स्तर में वृद्धि हुई है के रूप में हालांकि, पनीर में नमी की मात्रा कम हो। Mascarpone पनीर का पीएच पनीर में इस्तेमाल वसा Replacer की मात्रा के लिए एक सीधा संबंध दिखाया; मात्रा पनीर का पीएच मूल्य में वृद्धि के रूप में भी वृद्धि हुई है। 1.83 एन एस -1 Mascarpone पनीर (Cheese) में 16% वसा (60% वसा की कमी) युक्त का मतलब Spreadability 24 और 20% वसा (40% और 50% वसा की कमी) के साथ बना पनीर की तुलना में अधिक था 10% वसा Replacers के साथ जोड़ा जाता है और यह नियंत्रण पनीर की है कि बहुत समान था। प्रयोगात्मक नमूने की शेल्फ जीवन पॉलीथीन थैली में पैक किया और 5-8^{0}C पर संग्रहीत जब नियंत्रण नमूना की तुलना में 2 दिनों में अधिक से पता चला है।

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LIST OF ABBREVIATIONS AND SYMBOLS

ACF	:	Aberrant crypt foci
ACP	:	Aberrant crypts
ANOVA	:	Analysis of Variance
AR	:	Analytical Grade
CC	:	Control in Cup
CP	:	Control in Pouch
CVD	:	Cardio Vascular Diseases
Df	:	Degrees of Freedom
DM	:	Dry Matter
FFEC	:	Full Fat Edam Cheese
FR	:	Fat Replacer
GFT	:	General Food Texturiometer
GRAS	:	General Recognized as Safe
LA	:	Lactic acid
LDL	:	Low Density Lipoprotein
LFEC	:	Low Fat Edam Cheese
ML	:	Milligram
MPa	:	Mega Pascal
N	:	Newton
PDA	:	Potato Dextrose Agar
Ppm	:	Parts per Million
RBGL	:	Red, blue, green, Luminosity
RT	:	Room Temperature
SC	:	Sample in Cup
SNF	:	Solids Not Fat

SP	:	Sample in Pouch
SPC	:	Standard plate Count
TA	:	Titrable Acidity
TBA	:	Thiobarbutric Acid
TPA	:	Texture Profile Analyzer
UF	:	Ultra filtration
UTM	:	Universal Testing Machines
w/w	:	Weight by weight
WPC	:	Whey Protein Concentrate

Chapter- 1

Introduction

1.0 INTRODUCTION

The immense popularity of cheeses items from its nutritional package, versatility in tastes and many varieties. Cheese is the generic name for a group of fermented dairy products, produced throughout the world in a great diversity of flavours, textures and forms. All cheeses are made from milk or its by products but different manufacturing and aging processes are used to produce the array of cheeses. They are categorised in several ways; natural versus process cheeses, unripened versus ripened cheeses and soft versus hard cheeses. Cheese is a concentrated source of milk fat, protein and many of milk nutrients. There are thousands of different cheeses all over the world, each is unique in their own way. Each has a history of origin; making process, specific characteristics and uses. Some cheeses are wellknown and commonly recognized like Cheddar, Gouda, Mozzarella and Swiss, while others are less familiar to the common consumers like Quark and Mascarpone. New varieties of cheeses are constantly being made and developed by artisan cheese makers, as they focus on creating new and unique product that fit into and create niche markets.

Among the various unripened, soft varieties of cheeses, Mascarpone, an Italian soft cheese is gaining wide popularity because of its taste and spreadable nature which can be used in place of butter. It contains about 40-45% fat. Its mild flavour and soft texture allow it to be used in savoury and sweet dishes making the possibilities in nearly endless items in the kitchen. Mascarpone cheese is made from cream by heat and acid coagulation with diluted lactic, acetic or citric acid, to pH 5.0. The curd is drained in cloth and packaged. Recently, higher fat creams have been used in conjunction with UF in order to speed up the long curd drainage time required for the traditional process. This high fat content in cheese prevents health conscious consumer to include in diet, as high fat consumption is linked with cardio vascular diseases.

Consumers are increasingly demanding foods with dietetic and functional properties such as those with low calories, low or reduced fat and health benefits. Low fat foods have been recommended for weight loss and maintenance. In this regard, various cheese based functional foods are already launched in the markets. Functional foods are claimed to have health promoting or disease preventing property beyond the basic function of supplying nutrients. The general category of functional foods includes processed foods or foods fortified with health promoting additives like probiotics, peptides, fibre and fat replacers etc. Fibre provides health benefits liked, improved bowel functions, reduced blood glucose level, attenuates blood cholesterol and increased faecal bulkiness. Therefore, fibre enriched functional cheeses are gaining popularity among the health conscious consumers.

Mascarpone cheese is the new arrival in Indian cheese market in recent times. It is being used with bread, *roti*, c*hapattis* and other Indian snack foods in place of butter because of its rich flavour and unique spreadable properties. Many brands of Mascarpone cheeses are getting imported to Indian market. A few private entrepreneurs have shown their interest to commercialize this variety in India. Information indicates that till now no systematic approach have been made to enrich with fibre or to reduce fat content in Mascarpone cheese to make it more functional for the health conscious consumers in India. Therefore, considering the benefits of fibre and ill effects of high fat dairy products, an attempt was made to standardize its manufacturing process with different organic acids and to incorporate fibre in reduced fat cheese with suitable fat replacer for similar taste and improved health benefits. Reduction of fat level in the Mascarpone cheese by using suitable fat replacer may result in reduced risk of atherosclerosis and CVD (Cardio vascular diseases) of the consumers.

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Chapter- 2

Review of Literature

2.0 REVIEW OF LITERATURE

2.1 INTRODUCTION

Cheese is the generic name for a group of fermented dairy products, produced throughout the world. Every cheese has a story. There are thousands of different cheeses all over the world and each is unique in their own way. Each has a history of origin; making process, specific characteristics and uses. Some cheeses are well-known and commonly recognized like Cheddar, Mozzarella, Gouda and Swiss etc, while others are less familiar to the common consumer like Quark and Mascarpone. New varieties of cheeses are constantly being made and developed by artisan cheese makers, as they focus on creating new and unique product that fit into and create niche markets. The beauty of making cheese is being able to take milk, a simple raw material, and create a value added product with special, one-of-a-kind qualities that please hundreds, maybe even thousands of consumers. Supply of cheese to the world market mainly comes from the European Union followed by New Zealand and Australia. Cheese are well persevered milk foods contributing protein, fat, calories and vitamins to consumer while also providing a diversity of flavor, aromas and textures to the diet (Steinkraus, 1994). With gradual acquisition of knowledge on the chemistry and microbiology of milk and cheese, it becomes possible to direct the changes involved in cheese making in a more controlled fashion (Fox and McSweeney, 2004). Cheese is nutrient-dense food. Cheese provides calories; high quality protein; vitamins such as vitamin A, riboflavin (B2) and B12 and mineral such as calcium, phosphorus and zinc. Consuming cheese immediately after meals or as a between meal and snack helps to reduce the risk of tooth decay (Ahola, *et al.* 2002). Certain cheese such as aged Cheddar, Gouda, Swiss, Blue, Cream, Monterey jack and Brie cheese have been shown to prevent tooth decay Kashket *et al.(*2002). Calcium, phosphorus and other components in cheese may contribute to these beneficial effects. The primary objectives of cheese manufacturing were to extent the shelf life of milk and conserve its nutritive value. This can be achieved by either acid production and/or dehydration.

Cheese means the ripened or unripened, soft or semi hard, hard and extra hard products, which may be coated with food grade waxes or poly film. Cheese is obtained by coagulating wholly or partly milk and /or products obtained from milk through the action of non-animal rennet or other suitable coagulating agents and by partially draining the whey resulting from coagulation of milk and / or products obtained from milk, which gives a final product with similar physical, chemical and organoleptic characteristics. The product may contain starter cultures of harmless lactic acid and /or flavor producing bacteria and cultures of harmless microorganism, safe and suitable enzymes and sodium chloride. It may be in the form of blocks, slices, cut, shredded or grated cheese (Food Safety and Standards Act, 2006). Davis (1965) defines cheese as the product formed by coagulation of the milk of certain mammals by rennet or similar enzymes in the presence of the lactic acid produced by added or adventitious microorganism, from which part of the moisture has been removed by cutting, warming, and/or pressing, which has been shaped in mould and then by holding for some time at suitable temperature and humidity.

2.2 NUTRITIONAL ASPECTS OF CHEESE

Apart from water, man requires four major groups of nutrient in food, which in the past were often quoted as fat, protein, carbohydrate and salt. Modern nutritional standards are more demanding in respect of many minor components such as vitamins and minerals etc.

2.2.1 Milk Fat

Consumers generally prefer high fat cheese because a high fat content contributes significantly to flavor. Cheese from whole milk is known to contribute fatty acid but there has been an effort to introduce more unsaturated fatty acids (linolenic) into the diet of the animal in order to augment those in normal milk (Scott, 1986). The typically aroma some types of cheese, for instance Cheddar, develops only when the fat-in dry matter content is at least 40-50% because the aroma is due to mainly to the breakdown products of fat formed during cheese ripening. A high fat product causes cardio vascular diseases and other health problems and hence, there is a very high demand for low fat and functionally enriched food products. The production of low fat cheese offers great opportunities to the market, new produced which are perceived as ‘healthy’ due to the their low fat content (Scott, 1986). It is however essential that low fat cheeses are organoleptically acceptable. Scott (1986) reported the cholesterol content of cheese is rather low (0-100 mg/100 g, depending upon fat content), therefore cheese contribute only 3-4 % of the total cholesterol. Furthermore cholesterol in the diet has only a limited effect on the level of blood cholesterol.

2.2.2 Protein

In cheese manufacturing, casein of milk is contributed into the cheese while most of the biological valuable whey proteins pass into the whey. Thus, 75-80% of the total protein and about 95% of the casein are transferred from the milk to the cheese. Therefore, in cheese produced from pasteurized milk, the content of whey protein in total cheese proteins is low of 4-6 %. Since the whey proteins are nutritionally superior to casein which is somewhat deficient in sulphur amino acids. The biological value of proteins in cheese is somewhat lower than that of the total milk protein but is still higher than that of casein alone. The daily requirement per day of adult is approximately 1g/kg of the body fat weight. Cheese is the suitable source of protein since it normally contents all the essential amino acid (Scott, 1986).

2.2.3 Lactose and lactic acid

There is no or very less amount of lactose in many cheeses (1-3 g/100 g) because most of the lactose of the milk passes into the whey and that entrained in the cheese curd is partly or fully converted in the lactic acid during cheese ripening therefore cheese is suitable for the diet of persons suffering from lactose malabsorption and of diabetics (Blanc, 1982). The average composition of the lactose is as follows, Parmesan- 0.7%, Cheddar cheese- 1.3%, Tilsiter- 1.0%, Quarg - 0.7%, Blue cheese - 0.6%, Emmentaler- 0.4%, cottage cheese 0.3% and Camembert - 0.2%. Those people allergic to lactose can normally eat cheese, except for those very soft fresh unripened cheese which may still contain appreciable amounts of unfermented lactose.

2.2.4 Minerals and vitamins

Cheese contains appreciable levels of minerals, of which calcium (for bone and tooth formation), iron (for red blood) and phosphorus (for bone and teeth) are the most important.

The levels of fat soluble vitamins in a cheese depends upon the its fat content. In a full fat cheese approximately 80% of the vitamin A in milk passes into the product. The concentration of B vitamins is also useful, but the precise amount will depend on number of factors i.e. type of starter culture and length of maturation period. Thus, a number of starter culture bacteria have the ability to synthesize B vitamins (Scott, 1986). Various trace elements such as zinc, iron, iodine, selenium and copper are all found in milk to some extent. Although the total dietary intake from cheese may not be high, the regular consumption of cheese is beneficial with respect to these minerals.

2.3 CLASSIFICATION OF CHEESE

The great range of cheese varieties, excluding minor local variant, make classification of cheese extremely complicated. It is obvious that a well-known variety has certain distinctive characteristics, such as size, shape, weight, color, external appearances and analytical data for fat-in-dry-matter, salt content or water-in-fat-free substance, but it is more difficult to determine flavor and aroma, especially if milk from cow, sheep, goat or buffalo has been used singly or mixed for manufacturing of the cheese.

Personal preferences determine which variety or brand is to be purchased. In the major dairy countries like France or Italy in Europe, the range of cheese is market extensive, the variations within each variety and classification become difficult. Table 2.1 shows some popular variety of Italian cheeses.

Table 2.1 Classification of Italian cheeses

Type	Examples
Extra Hard/ Hard	Asiago, Parmesan, Grana, Reggiano, Romano, Sardo, Fiori Sardo
Pasta filata(mature)	Caciocavallo, Provolone, Incanestrato, Cartonese, Foggiano
Pasta filata(fresh)	Mozzarella, Provatura, Scamorze, Trecce
Hard with ‘eyes’	Asin, Fontina, Montasio
Internal mould	Castelmagno, Gorgonzloa, Moncenisio, Stracchino di Gorgonzola
Smear coated	Bel Paese, Cacitta, Crescenza, Raviggiolo, Taleggio
Soft (fresh)	Formaggini, Fresa, **Mascarpone**, Nostrale, Ricotta

(Fox and Guinee, 2000)

2.4 SOFT CHEESES

Fresh acid-curd cheeses are defined as those varieties produced by the coagulation of milk, cream or whey via acidification or a combination of acid and heat, which are ready for consumption immediately after manufacture (Guinee *et al.,* 1993). Volikakis *et al.* (2004) observed that addition of ß-gulcan concentrate in a low fat white brined cheese, affects adversely on cheese appearance and flavor in comparison with the control sample and the product fortified with ß-gulcan concentrate exhibited higher yield & significantly lower pH than control sample. Suarez-solis *et al.* (2002) prepared a fresh cheese of very good sensory quality with a shelf life of 15 days during which cell viability was 10^{-7} using probiotic lactic acid bacteria. Nayra *et al.* (2002) used different types of probiotics culture in the manufacturing of soft cheeses. Optega *et al.* (2003) prepared a substitute of cream cheese with low fat (15%) cheese and soya isolate, cheese whey & vegetable fat. The sensory quality of imitated cheese was excellent.

2.5 CHEESE TEXTURE

Texture of foods is a highly subjective human experience with foods during their consumption. The International Organization for Standardization (ISO) defines texture of food products as, all the rheological and structure (Geometric and surface) attributes of the products perceptible by means of mechanical, tactile and wherever appropriate. The textural attributes of foods play a major role in consumer appeal, buying decision and even for consumption. For some foods, texture is more important to consumer than flavor and color (Szczesniak *et al.,* 1963). Food texture is the single most dominant attributes for consumer preference of food as reported by Rohm (1990) based on several studies. Texture means different things to different people and the texture attributes expected from different foods vary wieldy. Chewing force and chewing movements are strongly influenced by food texture (Ahkgren, 1966). The texture profile analysis was originally developed at the General Foods Corporation Technical Center in the early 1960 (Friedman *et al.,* 1963). Kucukoner *et al.* (2003) studied the low fat (17%) and full fat (27%) fresh Edam cheese and aged up to 6 months at 5°C, and observed that low fat edam cheese (LFE) had significantly higher stress value than full fat Edam cheese (FFE). Koca *et al.* (2004)

observed that when prepared low fat cheese fresh Kasher cheeses by using fat replacer. It decreased the hardness, springiness gumminess & chewiness and increased the cohesiveness of the product. Adhikari *et al,* (2003) studied to find textural difference in cheeses by sensory and instrumental methods and attempted to correlate the sensory and instrumental data. Rahimi *et al.* (2007) concluded that use of gum tragacanth as fat replacer in Iranian white cheese, showed an adverse effect on cheese yield, sensory and texture & it increases the instrumental hardness & also increased the whiteness of cheese. Sahan *et al.* (2008) reported that texture attributes and meltability significantly increased with the addition of fat replacer in low fat Kashar cheese. Zisu *et al,* (2006) observed that control Mozzarella cheese exhibited poor meltability and stretchability and greater hardness, springiness, chewiness as compared to the capsular & ropy microbial expolysacchari‌ed low fat Mozzarella. Pottorak *et al*. (2015) observed the values of meltability for cheese with reduced fat content were lower than those of full-fat cheese.

2.5.1 TPA testing for cheese

Cream cheese and Process cheese have been used as anchors for hardness and cohesiveness scales on the original TPA test as described by Szzezesniak *et al.* (1963a). Brennam *et al.* (1970) performed TPA on cheddar cheese using the GFT (General food texturiometer) and obtained good correlation with sensory evaluation and Hardness. Brennam *et al.* (1975) compared the measurements made with the GFT and UTM (universal testing machine) and reported that the UTM measurement was better. The TPA remains to be among the most widely used instrument for measurement of cheese texture evaluation (Tunick, 1991). Imoto *et al.* (1979) investigated the effect of compression ratio (20 to 80%) on the firmness of different cheeses. They found little effect on the extent of compression on correlation between firmness and sensory evaluation.

2.6 MASCARPONE CHEESE

Mascarpone is high fat, soft variety of Italian cheese. It is milky-white in color and easily spreadable. Mascarpone is used in various dishes of the Lombardy region of Italy, where it is a specialty. It is a main ingredient of modern Tiramisu (Italian dessert). It is sometimes used instead of butter or Parmesan cheese to thicken and enrich Risotto (Italian rice dish). Its mild flavor and soft texture allow it to be used in savory and sweet dishes, making the possibilities nearly endless items in the kitchen. It is added to enhance flavor without overwhelming the original taste. The cheese tastes best with anchovies, mustard and spices or mixed with cocoa or coffee. Composition of Mascarpone cheese varies; Moisture – 44 to50%, Fat- 45 to 55%, Protein- 7 to 8% and Ash - 0.5 to 0.7% (Maggi, 1987).

2.6.1 History and origin

Mascarpone originated in the area between Lodi and Abbiategrasso, southwest of Milan in Italy, probably in the late 16th or early 17th century. Italy is a very popular country for fresh cheese where more than 40 % fresh cheese is sold (Potenza, 2002). The name is said to come from Mascarpa, a milk product made from whey or from Mascarpia, the word in the local dialect for ricotta. The correct name of the cheese should be Mascarpone, originally stemming from Cascina Mascherpa, a farmhouse that once was located halfway between Milan and Pavia, belonging to the Mascherpa family where it was reportedly first made.

2.6.2 Methods of Manufacture

Mascarpone is a traditional soft Italian cheese, made from hot (85-90°C) cream or high fat milk which is acidified with acid (edible) solution to pH about 5.0; the curd is drained in cloth bags and packed after complete draining of whey. Traditionally, Mascarpone cheese is manufactured by collecting cream and heated to 85-90°C in a double jacketed steam kettle to prevent scorching the cream solids. Then edible acid solution is added to the hot cream. This mixture coagulates shortly and is then transferred to a draining cloth to allow the whey to drain away. This process is similar to Paneer making in India but with very high fat milk.

Alternative process for traditional Mascarpone cheese is to use of naturally ripened cream with a bacteria culture. Cream is normally heated to 29-30°C, the culture is added and allowed to ripen for 10-12 hours to form a thick but soft curd. Although ripening sometimes takes more time but results in much smoother and creamier consistency than the traditional process. This process of Mascarpone cheese manufacture takes long time but an alternative to use higher fat cream (45-55%) which is mixed with 4:1 to 2.5:1 with milk retentate (whole milk concentrated by 4-5 time by ultra filtration). This method is quicker, economical in terms of labor with improved shelf life (Resmini *et al.*, 1984). Pagani *et al.* (1995) prepared a Mascarpone cheese by using a milk ultra filtration technology. Continuous production of Mascarpone cheese with using ultra-filtration technology decreases time of preparation and heating by plate exchangers rather than by steam injection, improves product quality (Sordi, 1984).

2.6.3. Types of acid used

Mascarpone cheese is heat-acid coagulated cheese where organic acid solution is commonly used to coagulate the high fat milk. Its taste and yield depend on the acid used for coagulation. It mainly is prepared by using citric, acetic or lactic acids. Among three acids, lactic acid is the costliest. Therefore, use of this acid for commercial manufacture of Mascarpone cheese is not economical. Vinegar (4% acetic acid solution), which is easily accessible and commonly available, can be used for cheese manufacture. Citric acid is a weak organic acid which is present in a variety of fruits and vegetables, most notably citrus fruits, with lemons and lime having the highest concentration. Citric acid is commonly used as a natural food preservative. It is added to obtain acidic or sour taste to foods and soft drinks. Most commercially used citric acid is obtained from a bacteria and sugar process and is sold as a white crystalline powder.

Buratto (2010) observed that use of vinegar showed about 1% more yield and higher fat recovery than that of cheese made with citric acid. However, the use of citric acid was recommended due to better textural characteristics to manufacture Mascarpone cheese. He has also observed that Mascarpone cheese made by citric acid showed white in color than other acid cheese and citric acid Mascarpone cheese prepared by citric acid easily spreadable. It gives clean taste whereas other acid gives cooked creamy and tangy flavor with less spreadable property.

2.6.4. Rheological properties

Mascarpone is soft, fresh and high fat cheese. Rheological properties of the product mostly depend on its composition, temperature and manufacturing conditions. Due to this high fat and high moisture content, Mascarpone cheese has spreadable body and texture. It is also used as a bread spread where it requires higher spreadability and smoothness. Negative correlation was found between shear stress values and panel response, while slope values showed to be directly correlated with sensory score. On the basis of this observation, it was possible to describe spreading properties of Mascarpone cheese (Cattaneo *et al.*, 2005).

Cheese curd heated with constant stirring and dynamic cooling followed by storage at 5°C for 7-9 days. Result of curd showed the time dependent hysteresis of the flow properties and dynamic visco-elastic behavior. Curd mixing at 500 rpm and 70°C and dynamic cooling at 20°C or 13°C of double cream cheese resulted in decrease of firmness with increase of viscosity but this result became totally opposite when the curd was homogenized at 20 MPa +5 MPa at 85°C followed by cooling at 13°C. The product became more firm (Sanchez *et al.*, 1994). Double cream cheese homogenized at (0, 5,10,20,30 and 50 MPa) showed the extrusion shear force increasing due to increasing the homogenization pressure till 20 MPa thereafter double cream cheese could not be extruded because it became brittle. Further, if the homogenization pressure increases, simultaneously storage modulus increases and loss modulus decreases. The combine result of both modules in double cream cheese became more solid in state. Therefore, it was set to keep maximum homogenization up to the 20 MPa for good result (Sanchez *et al.*, 1995). In double cream cheese incensing the curd cooling rate produces soft body with weaker structural organization which means decreased in loss and storage modulus (Sanchez *et al.*, 1994).

2.6.5 Microbial, Chemical and Physical Quality

Mascarpone cheese has high moisture and fat. High moisture products have low shelf life. It has 3-4 day shelf life at room temperature. Microbial spoilage is less as compare to other cheese because Mascarpone cheese is heat-acid coagulated cheese. Acid acts as a preservative but it is highly susceptible to the yeast and moulds. Lactic acid bacteria in cheese are indicators of high/good quality of cheese. Lodi *et al.* (1994) have suggested that the presence of high content of lactic acid bacteria in cheese was indicative of good quality of cheese or called as fresh cheese and they also observed Pasta Filata cheeses with high content of LAB but in Mascarpone and Ricotta cheeses, absence of lactic acid bacteria was reported. Carminati *et al.* (2001) investigated the ability of a biological control system to inhibit the growth of *Clostridium sporogenes* spores during storage of Mascarpone cheese under temperature abuse condition.

Color of Mascarpone cheese depends upon its raw material used. Cow milk cream produces Mascarpone cheese with slightly milky-yellow color whereas buffalo milk cream Mascarpone cheese gives white color. Content of β carotene and fatty acid composition of the cream has maximum influence on physical and sensory properties of Mascarpone cheese. Yellowness index of cream is less than 20, RFM 2.9 to 3.1 with DM greater than 58% and RPL 30 to 35 for Mascarpone cheese manufacturing and the desired characteristics of the cheese can be obtained by blending different types of cream (Battelli *et al.,* 1995). Cream color is the most important factor for the final Mascarpone cheese color. Giangiacomo *et al.* (1991) have done work on the instrumentally measuring the cream color index and classified the cream on basis of the color for good Mascarpone preparation. During storage of Mascarpone cheese, discoloration happens from creamy yellow to orange brown color due to *Pseudomonas aeruginosa, P. fluorescens*, *P. aureofaciens, Shewanella putrefaceiens, Serratia marcescens, Enterobacter agglomerans* and *Micrococcus luteus* (Cantoni *et al.,* 1997).

Presence of D-Amino acid in cultured food and cheese function as ripening markers. D-Amino acid may also be present in Non- cultured food due to the microbial contamination of raw material. Mascarpone cheese also contents D-Amino acid (Chiavaro *et al.,* 1998). Presence of *Clostridium botulinum* type A spore in dessert *Tiramisu* was reported through its key ingredients of Mascarpone cheese (Aureli *et al.,* 2000). A fatal case of *C. botulism* in Italy due to the consumption of Mascarpone cheese from an dairy was reported and possible sources of contamination were discussed by Spolaor (1996). Presence of toxin in Mascarpone cheese produced by *C. botulinum* due to the different temperature conditions was reported (Franciosa *et al.,* 1999). Joseph *et al.,* (1998) have discussed the detection of botulinal toxin producing organisms from Mascarpone cheese using an Amplified Elisa System.

2.6.6. Reduction of fat content in cheese

Mascarpone cheese contains high fat. The consumption of fat is linked to increase low density lipoprotein (LDL) cholesterol levels, which in turn is linked to increase risk of atherosclerosis and cardio vascular diseases (CVD). Therefore, reductions of fat level in Mascarpone cheese by using suitable fat replacer may result in reduce the risk of atherosclerosis and CVD of the consumers. Quiblier *et al.* (1990) have reported that the use of tapioca as a partial fat substitute in fresh cheese. Corradini (1998) reviewed the use of high protein as fat replacer in the production of high fat Mayonnaise and Mascarpone cheese by incorporation of pasteurized skim milk whey. Similarly, Sensidoni (1994) investigated the possibility to mix the traditional Mascarpone cheese with pasteurized whey to achieve low fat content.

2.6.7 Use of Mascarpone cheese

Mascarpone cheese is used to prepare Italian desserts like Tiramisu. It is frequently used to thicken and enrich the Italian rice dish *Risotto* instead of butter or Parmesan cheese. Its mild flavor and soft texture allow it to be used in savory and sweet dishes, making the possibilities in nearly endless items in the kitchen. Mascarpone cheese can be used for direct consumption, as a topping for desserts, with *roti, chapatti* and bread as spread. It can also be added to pasta for creamy finish, dishes to increase the richness and in fresh cut fruits.

Recipes for frozen desserts such as mousses, ice creams and cocktail sauces were made with fresh cheese such as Quarg and Mascarpone (Mehrens, 1990). Use of fresh Mascarpone cheese (85 % fat in DM) in frozen dessert as a base formulation has also been reported by Mehrens (1991).

2.7 INULIN AS A FUNCTIONAL INGREDIENT

2.7.1 Preamble

Inulin appears as an important food ingredient that could be widely explored in research and by the food industry for the production of functional foods. It has many interesting functional attributes that are useful in formulating variety of cheese. The consumer of today is health conscious and demands foods with low fat and health benefits. The influence of inulin as a prebiotic on the activity of starter probiotic cultures and the pH value of cheese depend on the type of bacteria used in the products, as well as the type of inulin. Considering the diversity of cheese manufacturing procedure, most important factors that affect the final quality of different types of cheeses containing inulin depend on chain length of inulin, inulin concentration, preparation temperatures and amount of shear. Generally, application of inulin in cheese is of great interest in the development of prebiotic cheese that maximizes nutritional benefits and sensory characteristics to meet consumer demands. The inclusion of inulin in cheese as a fat replacer/texturizer/texture modifier has different influences on the rheological and textural properties depending on the structure and composition of each type of cheese matrix. Sensory properties can vary among the different inulin contents and/or blends. For modification and optimization of different applications of inulin in cheese, an adequate understanding of the physiochemical characteristics of inulin polymers is needed. More information on versatile aspects regarding the effects of inulin in different types of cheese is needed before using it on a large industrial scale. Solowjiej *et al* (2015) suggested that milk fat in processed cheese analogues can be partially replaced with inulin to improve the functional properties of the final product.

Inulin is a soluble dietary fibre forming a subset of nutraceutical ingredients that are increasily used in food products (Fagan, 2006). Basically inulin is a carbohydrate of fructan family. Fructan, in general is a term used for any carbohydrate in which fructosyl-fructose links constitute the majority of the glycosidic bond fraction which are linear or branched fructose polymer that are joined together by either ß (2-1) or ß (2-6) bond.

2.7.2 Health beneficiary effect of inulin

2.7.2.1. Inulin as a dietary fibre

While universally accepted definition for dietary fibre doesn’t exist, it is generally agreed that this term include saccharides (lignin) that are not hydrolyzed or absorbed in the upper part of the gasto intestinal tract (Flamm *et al.,* 2001). Dietary fibre reach the colons intact, where they may be totally fermented, partially fermented or remain unfermented. Fibre also contributes to fecal bulkiness in a number of ways. They can contribute directly via their own mass and/or the mass of the water they attract. In addition, they can influence fecal bulking in a direct manner, by being fermented colonic microflora thus stimulating their growth and resulting in a microbial biomass when fermented they are metabolized to hydrogen, methane, CO_2 and short chain fatty acids. These short chain fatty acids are absorbed and further metabolized in a colonocytes, the liver cells or the peripheral tissues (Flamm *et al.,* 2001). Indeed, the five basic attributes of dietary fibre are:

- Component of edible plant cell
- Carbohydrate (both oligosaccharide and polysaccharides)
- Resistance to hydrolysis by human alimentary enzymes
- Resistance to absorb in small intestine
- Hydrolysis and fermentation by bacteria in the large intestine

Inulin is known as soluble dietary fibre because of the B (2-1) configuration of fructosyl-fructose linkage. Inulin type fructoses are plant carbohydrates that resist digestion in upper gastro intestinal tract but are fermented in the colon by increasing fecal biomass and water content of stools. They also improve bowel habits. Inulin-type fructanse are:

- Part of edible plants
- Carbohydrate that are composed of a mixture of either oligosaccharide or oligosaccharides and polysaccharide
- Resist hydrolysis by human digestive enzymes
- Not appear to be significantly absorbed in the small intestine except possibly for the very short oligosaccharide (Menzies, 1974)

- Hydrolyzed and completely fermented by the colonic micro flora and are oxidized to produce gasses and short chain fatty acids

For all these reasons they are undoubtedly part of the dietary fibre complex (Roberforiod, 2005).

2.7.2.2 Inulin and gastrointestinal mineral absorption

Besides the amount of mineral (especially Ca and Mg) in the diet, the absorption of dietary minerals in foods is also a critical factor in determining their bioavabilitiy. Thus, there is a need to identify food components and/or functional food ingredients that may enhance mineral absorption in order to optimize their bioavability from foods (Weaver *et al.,* 2002). A remarkable increase in the absorption of calcium, a nutrient that helps to build and maintain structure of teeth and bones has been shown by ingredients from chicory roots. Several investigations have demonstrated that rats fed with oligofructose and/or inulin absorbed more calcium and magnesium than control rats despite an increase in total fecal mass (Ohta *et al.,* 1994). Inulin which acts as a soluble fiber increased the intake of calcium in the bone tissue that resulted in improving bone mineral density thus showing the potential to prevent or postpone osteoporosis (Kaur, 2002). Increased calcium absorption could be due to its increased availability by transfer of calcium from the small intestine into the large bowel. The osmotic effect of inulin and oligofructose transfer water into the large intestine, thus allowing it to become more soluble (Carbin, 1999). Improved calcium availability in the colon could also be as a result of hydrolysis of calcium phytate complex by bacterial phytase releasing calcium (Lopez *et al.,* 2000). Coudhray *et al.* (2003) noted that inulin improved the absorption of calcium but not of magnesium, iron and zinc in humans. Mechanism by which ingestion of non-digestible carbohydrates improves mineral absorption is not very clear.

2.7.2.3 Inulin and lipid metabolism

Modulation of either the digestion/absorption or the metabolism of lipid is another physiological effect of inulin-type frutans that affect trigyceridaemia and cholesterolaemia as well as the distribution of lipids between the different lipoproteins in favor of a more beneficial health pattern (Delzenne *et al.,* 2002). Jackson *et al.* (1999) reported that daily addition of 10 g inulin in the diet lowered the plasma triacylglycerol levels in 54 healthy middle aged men and women during the 8 weeks test period. The effect on cholesterolaemia is less constant. Kaur *et al.* (1998) reported the cholesterol lowering effect of inulin rich diet in caffeine- fed rates. These decreases are likely to be due to a reduction in the number of VLDL partials with the same composition in the lipids and the same size (Delzenne and Kok, 1998).

2.7.2.4. Inulin and anticancer activity

Cancer of the colon is one of the leading causes of cancer morbidity and mortality among men and women. Aberrant crypts (ACP) are putative precursor lesion from which adenomas and carcinomas may developed in the colon. Administration of inulin in the diet significantly suppressed the total number of aberrant crypt foci (ACF) compared with control diet. The role played by inulin and oligofructose in reducing ACF formation, an early pre-neoplastic marker of malignant potential in the process of colon carcinogenesis, suggests that they may suppress colon tumourigenesis (Wargovich *et al.,* 1996). Taper and Roberfroid (1999) studied the influence of inulin and oligofructose on breast cancer and tumour growth. In a preliminary study on methyl nitoso-urea induced mammary carcinogenesis in Sprague-Dawley female rats, 15% oligofructose added to the basal diet lowered the number of tumor bearing rats and also decreased the total number of mammary tumors. The anticarcinogenic and antitumor effect of inulin is in conjunction with the prebiotics. It appears that inulin inhibits preneoplastic lesion, probably by changing the composition of micro flora (Wang *and Gibson* 1993).

2.7.3 Effects of process and process conditions on inulin

There are various methods for the incorporation of inulin into cheese. In a study of fresh cream cheese, inulin was incorporated into the cheese after whey draining through homogenization resulting to form a smooth and homogeneous cream cheese (Buriti *et al.,* 2007). In another study of cream cheese, the curd mass was mixed with remaining ingredients such as salt after completing whey removal, and inulin was then added in the desired amounts followed by mixing (Alves *et al.,* 2013). Inulin should be added in cheese production before inoculation of milk protein concentrate (Miocinovic *et al.,* 2011). In

Karish cheese, skim milk was heated at 74°C for 15s and inulin was added to warm milk (Alnemr *et al.,* 2013). It has been suggested for low fat fresh cheese spreads that inulin should be added to the cream, heated to approximately 80°C and mixed several minutes to hydrate. This resultant is then mixed to the fresh cheese followed by pasteurization at 80°C and homogenization at 200 bars. The pH of the most cheeses in which inulin is used as a fat replacer, are never lower than 4 (Blecker *et al.,* 2001). In different studies, the formulations containing the highest inulin content have been shown to have the lowest moisture contents, as the prebiotic contributed to the total solid content (Akalin *et al.,* 2007).

2.7.4 Application of inulin as a fat replacer

The presence of fat in dairy products plays an important role for their physical, rheological, and textural properties (Barclay *et al.,* 2010 and Dave, 2012). Fat, apart from its nutritional significance in cheese, contributes to the sensory and functional properties of dairy products (Miocinovic *et al.,* 2011). Consumers are increasingly demanding foods with dietetic and functional properties, such as those with low calories, low or reduced fat and health benefits. Low-fat food plans have been recommended for weight loss and maintenance (Carmichael *et al.,* 1998 and Peterson *et al.,* 1999). Low-fat or reduced fat foods are less desirable because they have poor organoleptic qualities (Hamilton *et al.,* 2000). There are some reviews about specific applications and potential effects of fat replacers (Ognean *et al.,* 2006). The challenge of using fat replacers in cheese while keeping the same functional and organoleptic properties as full fat cheeses has attracted great attention (Kebary, 2002). Inulin is widely used as texturizing agents in low-fat foods, particularly in the European Union and increasingly in the U.S.A. and Australia (Devereux *et al.,* 2003). Inulin seems particularly suitable for fat replacement in low-fat cheeses, as it may contribute to an improved mouth feel (Meyer *et al.,* 2011). A creamy mouthfeel is achieved when inulin is used as a fat replacer in dairy products due to its interactions with whey protein and caseinate (Karaca *et al.,* 2009). High performance (HP) inulin with long chain and high molecular weight is most desirable as a fat replacer. Longer chain lengths reduce the solubility of inulin type fructans and result in the formation of inulin micro crystals when mixed with water or milk. These microcrystals are not discretely perceptible and have a smooth, creamy mouth feel. The fat mimetic property of HP inulin is double than standard inulin, while it has no sweetness (Niness, 1999). The different functional attributes of inulin and oligofructose are due to the difference in their chain lengths. As noted above, due to its longer chain length, inulin is less soluble than oligofructose, and has the ability to form inulin microcrystal when sheared in water or milk. Inulin has therefore been used successfully to replace fat in dairy products (Kaur & Gupta, 2002). The ability of inulin as fat replacer is not only related to the modification of rheological behavior or the thickness or hardness of the product, but also to changes in other mouth feel attributes, such as creaminess or smoothness (Meyer *et al.*, 2011). To obtain low-fat products with rheology and thickness closer to those of full fat products, higher concentrations of inulin are needed than is necessary to merely mimic their creaminess or smoothness (Meyer *et al.*, 2011). Fadaei *et al.* (2012) studied the chemical characteristics of low-fat whey less cream cheese containing inulin as a fat replacer. No significant difference was found in the pH and salt values of cream cheeses. They indicated that an inulin proportion of 10% was enough to obtain a low-fat cream cheese with chemical attributes near to those of high fat cream cheese that does not contain inulin. They also reported that inulin has an excellent water binding capacity which inhibits syneresis in spreads and fresh cheeses (Fadaei *et al.*, 2012). It is expected that long chain inulin versus short chain has considerable water binding/retention capacity and capability to prevent syneresis. Wadhwani *et al.*, (2011) conducted several preliminary studies to select the most efficient fiber type from four inulin fibers; low methoxy pectin, polydextrose and resistant starch–to improve the quality of low-fat Mozzarella and Cheddar cheeses (Wadhwani, 2011). Results from their preliminary studies indicated that inulin had better efficacy in cheese systems than the other three fibers. They also found that incorporating inulin led to improved texture in low-fat cheese by decreasing hardness and gumminess while maintaining cohesiveness, adhesiveness and springiness (Wadhwani, 2011). Overall, studies have shown that the effect of fat replacement on cheese texture depends on the nature of the fat being replaced (Lobatocalleros, 1998).

2.7.5 Inulin application in dairy products

Incorporation of inulin in a variety of foods, especially dairy products is mostly due to two reasons. One reason can be attributed towards the various physiological functions which confer to the consumer (i.e. dietary fibre, prebiotic etc). Other reason is the different technological properties of inulin and its functionality in the food matrix (i.e. mimic texture modifier etc). For example, when inulin replaced corn syrups in reduced fat in ice cream formulation, a chewier texture was created. However, ice crystal formation was reduced when 50% of the corn syrup was replaced with inulin during thermally abusive storage condition (Schaller-Povolny, 1999). Akin *et al.* (2007) showed that addition of inulin (1 and 2%) in ice cream stimulated the growth of *L. acidophilus* and *B. lactic* and improved viability of this organism. Elewa *et al.* (2009) also observed the prebiotic effect of inulin (2%) in UF-probiotic white soft cheese. Yoghurt with inulin was identified as being creamier in appearance, having a less chalky and creamier texture, and was sweeter with a less sour/fermented taste and aftertaste (Spiegel *et al.,* 1994). Due to the unique functional properties, inulin has to manage water effectively, affect rheology and improves texture in foods and acts synergistically with high water binding hydrocolloids, which has allowed inulin to be used across all food product application areas, particularly in low and no fat and low and no sugar system as fat-replacers. Because of various functional characteristics, it is possible to use of inulin in water based products, such as dairy products and table spread, salad dressing, baked products, frozen desserts and drink etc. The most important physiological role, besides the soluble dietary fibre, played by inulin is its prebiotic ability. Several studies show that inulin addition can improve the viability of the probiotic in various milk products.

The technological reasons for adding inulin to foods relate to its capacity to act as fat and sugar replacer as well as emulsifier, thickener and stabilizer. The functions vary with the nature of the inulin (e.g. chain length), its concentration in a food and the food itself. The technological reasons relate to the dispersing properties of inulin in particular its ability to mimic fat droplets dispersed in water. This dispersion can then be used in food to replacer fat or to impart texture qualities in foods. The amount of inulin derived substance used for these purposes will vary depending on the technological purpose to be fulfilled.

In fermented milk beverage the effect of inulin addition seems to follow similar pattern. Inulin tends to increase firmness and viscosity of the low fat fermented milks if added in the range of 2 to 6 % (Tratnik *et al.,* 2006 and Pinheiro *et al.,* 2009). Another study by Villegas *et al.* (2007) showed that the skimmed milk vanilla beverage can mimic the whole milk vanilla beverage with inulin addition from 4-10%. For semi-solid products like yoghurt (low fat or fat free), addition of inulin resulted in increased creaminess and mouth feel (Guven *et al.,* 2005; Kip *et al.,* 2006 and Guggisberg *et al.,* 2009). Brennan *et al.* (2008) observed that above 2% of inulin was needed to exert significant improvements in apparent viscosity and storage modulus values and to obtain values closer to those of full-fat yoghurt. Narender (2009) during incorporation of different levels (1, 2, 3 and 4.5%) of inulin in Misti dahi found that inulin at higher levels of incorporation (4.5%) significantly reduced the body and texture score than control. Up to 4.5% addition of inulin, although, decreased the overall acceptability score, but the product was still acceptable. Paseephol *et al.* (2008) showed that low fat yoghurt with 4% inulin gives comparable properties to full fat yoghurt. The taste and texture of symbiotic Cottage cheese remained unchanged up to 8% inulin addition (Araujo *et al.,* 2010). Gahane (2008) reported that more than 10% inulin addition in Quarg cheese significantly increased the firmness and decreased the work of shear and stickiness. During storage (21 days) body and texture score decreased on the 7th day of storage. This was mainly attributed to hard and firm body. Inulin being soluble makes the product comparatively hard during storage. But, after reaching the limit, it remains constant. The Quarg cheese was spreadable when tested at 15-20°C. Koca *et al.* (2004) observed the fat mimetic effect of inulin and found that addition of 5 % inulin to the low fat Kasher cheese improve slightly higher than that of full fat control cheese. However, the sensory scores decreased over 60th and 90th days of storage. Pagliarni *et al.* (1994) prepared low fat Mozzarella cheese by adding inulin and reported that inulin addition significantly improved the sensory quality particularly increasing softness and wateriness which were perceived as positive sensory attributes by consumers. El-Nagar *et al.* (2004) manufactured Karish cheese from reconstituted skim milk (11%

SNF) by using fat replacer inulin at the rate of 0.5, 1.0 and 1.5 %. Due to inulin addition hardness and elasticity decreased but adhesiveness, cohesiveness, gumminess and chewiness increased. Inulin was incorporated as a fat mimetic into natural cheese at levels up to 10g/100g. It has been observed that it had no affect on cheese aroma (Gijs *et al.*, 2000).

2.7.6 Application of inulin as a texturizer

Highly soluble fibers are highly branched and those that are relatively short chain polymers, such as inulin, have low viscosities. They are generally used to modify texture or rheology, manage water migration, influence the colligative properties of the food system and enhance the food product's taste, mouth feel and shelf life without significantly altering its specific application characteristics and improve its marketability as a health promoting or functional food product. As inulin content increases, its effect on the product's structure and texture becomes important, because at higher levels of inulin, the physic-chemical properties can modify the texture of dairy products and may significantly influence their sensory quality. It has been observed that the viscosity of the products increases with increasing levels of inulin (Akin *et al.*, 2007), Hennelly *et al.* (2006) compared the use of shear induced inulin gels and heated inulin solutions to replace 63% of the fat in imitation cheese. They also found that at equivalent moisture levels, the inulin cheeses had significantly higher hardness values than the control sample with fat. However, there was no difference in hardness among the cheeses containing different levels of inulin (5% or 13.75%). Cheeses manufactured with 3% of inulin were characterized by a more compact structure, denser protein matrix and more uniform disposition of protein chains and the pores between them compared to other cheeses (Miocinovic *et al.*, 2011). It has been speculated that inulin may become part of the protein structural network by complexion with protein aggregates if inulin is present during fermentation and coagulation (Kip *et al.*, 2006) or if water phase insoluble submicron crystalline inulin particles form a particle gel network (Franck, 2002). According to the study of Juan *et al.* (2013) on the sensorial properties of reduced fat fresh cheese, the addition of 5% inulin in milk resulted in a retention of 3% of inulin in the resulting cheeses. They found that the pH and microbiological quality of cheeses were not affected by the presence of inulin. In their study, cheeses produced with inulin were less hard, springy, cohesive and chewy than reduced fat cheeses. Inulin's water retention capacity could increase the water available for salvation of the protein chains, resulting in a softer, more easily deformed cheese (Creamer, 1982). Alnemr *et al.* (2013) investigated the effect of adding texturizing inulin at levels 2 and 4% on physicochemical properties of Karish cheese. The use of inulin significantly enhanced the yield of cheese and moisture content compared to control. They related the increase in yield to the form of a gel network and increase of the ability of water holding in cheese containing inulin. It was found that the pH values of Karish cheese with inulin were higher than that of the control cheese during the storage. Higher addition of inulin led to decrease in hardness of Karish cheese.

2.8 LOW FAT CHEESE

The term low fat cheese generally refers to cheeses whose fat content is lower than its corresponding full fat variety. Specifically, as the fat content of Cheese is lowered, moisture content increases and protein plays a greater role in texture development. The moisture in nonfat substance of cheese is generally equal to that in full fat cheese (Mistry *et al.*, 1993). Cheeses with reduced fat levels are usually also excessively dry and possibly grainy, due to the greater structural matrix per unit cross-sectional area (Emmons *et al.*, 1980). In the past years, the commercialization of low fat cheese production around the world has significantly accelerated. Even though the concept of low fat cheese manufacture is not a new idea. past 20 years has largely been responsible for the growth in low fat cheese markets. Reduced fat and low fat cheeses accounted for approximately 20% of supermarket sales of cheese in United States in 1998 and in Sweden low fat cheese consumption doubled over a three-year period (De, 1991). Fat is not only of nutritional significance in cheese, but also contributes to sensory and functional properties. Low fat cheeses, especially hard varieties, are usually characterized as having rubbery body and flavor notes that are a typical of corresponding full fat varieties. Functional properties of low fat cheeses also are not adequate. Soft, unripened varieties have had more commercial success. Reduction of the fat content in cheese also alters the main parameters defining texture and rheology of cheese. In low-fat Cheddar and Mozzarella cheeses, for instance, values of springiness and hardness are higher and cohesiveness lower than in full-fat cheese, because of the changes that take place in the microstructure (Tunick *et al.,* 1993). An increase in viscoelasticity was obtained in reduced-fat Cheddar and Mozzarella cheeses (Ustunol *et al.,* 1995). Low fat unripened varieties such as Cream, Cottage, Mozzarella and others also possess certain unique characteristics that are not desirable to the consumer. These qualities are related to compositional differences between full and low fat cheeses.

The difference in the rate of release of flavor compounds from cheese during chewing is also a factor in flavor perception (Delahunty *et al.,* 1996). Low fat ripened cheeses generally have flavor that is a typical for the variety. In low fat Cheddar cheese the lack of and an imbalance of flavor has been associated with lowered levels of fatty acids such as butanoic and hexanoic acids and methyl ketones (Banks *et al.,* 1989). Bitterness in low fat Cheddar cheese may be lowered by increasing the salt in moisture phase of cheese to >4.5% to control microbial activity, but this also makes the cheese harder (Mistry *et al.,* 1998). Lee *et al.* (1996) concluded that when a bitter-tasting fraction was incorporated into reduced and full fat Cheddar cheese, the intensity of bitterness was more significant in the Cheeses with reduced-fat content. The origin of flvour defects that normally appear in low fat cheeses seems to be related to the reduced fat content (Lee *et al.,*1996), lower rate of lipolysis, modification of the protein matrix and the rate of proteolysis in these cheeses (Ardoe, 1993). The higher moisture content of low-fat cheeses modified the activity of the starter culture due to the effects of higher moisture in non-fat solids (MNFS) and lower salt-in moisture contents (S/M), both of which have critical effects on the ripening process (Leliavre *et al.,* 1982). Texture development in cheese occurs due to the breakdown of αs1-casein during ripening (Lawrence *et al.,* 1987). Milk fat normally provides a typical smoothness to a full fat cheese by being evenly distributed within the casein matrix of cheese. The extent of hydrolysis depends on the moisture and salt content of cheese (Mistry *et al.,* 1998). A cream dressing is then added to adjust the fat content. The curd absorbs a small amount of dressing, hence low fat Cottage cheese can be rubbery in the absence of adequate quantities of cream dressing. Though Mozzarella cheese is not considered a ripened cheese, a small degree of casein breakdown is required for body and texture development and functionality. For example, when fat content was reduced to below 15%, proteolysis during storage decreased and the hardness of cheese increased, which in turn also adversely affected its functionality (Rudan *et al.,* 1999). Bitterness develops early in the aging process and is a common defect in aged low fat cheeses, partly because of low salt and high moisture content. These compositional factors along with manufacturing procedures that are typically used for low fat cheese making induce excessive growth of starter organisms (Ardo, 1993) and proteolysis (Mistry *et al.*, 1998).

Cheese making procedures, i.e., use of low fat milk, specialized cultures, etc., directly influence its quality characteristics. A common immediate consequence is a significant change in appearance. The hard varieties and others such as Mozzarella, lowering fat content imparts a dull translucency to the surface, especially along the edges, of the cheese due to fat related changes in light scattering (Kosikowski *et al.,* 1997). Upon heating, low fat Mozzarella cheese undergoes a large increase in whiteness which decreases upon cooling (Metzger *et al.,* 2000). These changes apparently are caused by gels that are formed by casein and casein proteolysis products during heating (Metzger, *et al.,* 2000). Color change may not be as apparent in higher fat, softer unripened cheeses. Low fat Mozzarella cheeses typically have poor meltability (Fife *et al.*, 1996 and Tunick *et al.*, 1993). Low fat and fat free Cream cheese have been developed (Meilinger *et al.,* 1995). These products have smoothness that is similar to conventional Cream cheese but are not suitable for baking because of the high moisture content and burning which may occur during baking. Fat and nitrogen recoveries in cheese are also important in cheese yield. The percentages of expected recoveries depend on variety and are affected by a number of cheese making factors. For Mozzarella fat recovery is 85% (Rudan *et al.*, 1999) and Cheddar it is 93% (Kosikowski *et al.,* 1997). Homogenization of the milk or cream prior to processing helps to reduce fat loss in the whey and increases cheese yield (Metzger *and Mistry*. 1994). Similar results can be obtained by the use of ultra-filtered (UF) sweet buttermilk (Mistry *et al.,* 1996) or UF milk (Rodroaguez *et al.*, 1996) in the manufacture of cheeses with reduced-fat content. Different alternatives for increasing the moisture content of low-fat cheese by modifying the conventional cheese making technologies have been proposed, such as higher milk pasteurization temperature, preacidification of milk, briefer curd handling time, reducing the scald temperature, lower temperature during drainage of whey, milder physical treatment of the curd and shorter pressing time (Katsiari *et al.,* 1994: Banks *et al.,* 1994 and Rosenberg *et al.,* 1995). However, such modifications to the conventional cheese making process have to be very precisely regulated to avoid problems like milk clotting, loss of cheese yield and defective sensory characteristics (Olson *et al.,* 1990.)

In this connection, it has been proposed to add non- fat dry milk, condensed skim milk or skim milk to whole milk, although the amounts to be added must be regulated to prevent problems like milk clotting, curd handling and excessive lactose levels in cheese. Further- more, addition of condensed skim milk with an excessive total solids (TS) content results in reduced-fat Cheddar cheese with an extremely firm and crumbly texture (Anderson *et al.,* 1993). The use of UF milk has been reported as showing promise as a means of improving low and reduced fat cheeses (McGregor & White 1990), UF cheeses ripen more slowly than conventionally-made cheeses due to the presence of rennet and plasmin inhibitors in the whey protein fraction (De *et al.,* 1980). Because of the high water holding capacity of whey proteins, UF cheeses are normally softer, and this could offset the hardness that is commonly found in low-fat cheeses (McGregor & White 1990 and De *et al.,* 1980). In low-fat cheese, the predominant feature of the structure is a higher proportion of protein, which causes texture defects (Mistry *et al.,* 1993). Homogenization of milk increases the surface area of fat globules, which may improve the body and texture of reduced fat cheese (Mistry *et al.,* 1993 and Metzger *et al.,* 1994), although it can also produce adverse effects on the protein structure and cause casein membrane interactions in the fat globules which adversely affect the characteristics of the curd formation (Tunick *et al.,* 1995). Homogenization of milk can also produce alterations in the characteristic color of cheese (Drake *et al.,* 1995). It is possible to improve the texture of reduced-fat Cheddar cheese by homogenizing only the milk fat without producing any negative effect on the other parameters for defining cheese quality (Metzger *et al.,* 1994 and Metzger *and Mistry* 1995). Nowadays, different fat mimetics are used in the industrial production of low-fat cheeses. McMahon *et al,* (1996) used two protein-based fat mimetics (Simplesse1 and Dairy- Lo1) and two carbohydrates based fat replacers (Stel-lar2 and Novagel2) for the production of low-fat Mozzarella cheese. The moisture content of the low-fat cheeses with added fat replacers was higher than that of the control cheese containing no fat replacer. The combined use of carrageenan and microcrystalline cellulose to make reduced-fat Cheddar also improves texture due to the changes that occur in the microstructure, which has been found to resemble that of a full-fat cheese (Bullens *et al.,* 1994). Also, good results were obtained in the utilization of fat mimetic for the production of reduced-fat Cheddar cheese, although some fat mimetic may have a negative effect on flavor (Drake *et al.*, 1996).

Technology of manufacturing procedures developed for manufacturing low fat cheeses involve three broad approaches namely (a) processing techniques, (b) starter culture selection, and (c) use of additives such as stabilizers and fat replacers. Combinations of these procedures are also used.

2.8.1 Processing Techniques

In low fat Cheddar cheese making, the cooking temperature varies between 30–51°C, depending on the moisture content desired (Banks *et al.*, 1989). For low fat Cheddar, pH may range between 5.6 and 5.8. Washing curd with cold water (21°C) also helps to retain moisture, remove excess lactose, and solubilize calcium, which helps soften cheese texture. Excessive whey protein addition is likely to interfere with rennet curd formation and ultimately adversely affect cheese quality (Guinee *et al.,* 1998). Tunick *et al.* (1993) reported on the use of milk homogenized at 10, 300 and 17,200 kPa for manufacturing low fat Mozzarella cheese. Hargrove *et al.* (1964) received patent for procedures for the manufacture of low fat cheese. These early studies highlighted the important processing parameters for manufacturing low fat heeses. Milk may be fortified with nonfat dry milk or may be condensed up to 1.8 (Anderson *et al.,* 1993) fortified with dried ultra-filtered or micro filtered retentate (St-Gelais *et al.,* 1998). Rodriguez *et al.* (1999) concluded that semi hard low fat cheese made with milk concentrated by microfiltration had sensory qualities similar to full fat counterparts because of the retention of less (35%) whey proteins. The ratio of casein to fat in milk is also important. For manufacturing a 33% fat reduced Cheddar cheese, a Casein to fat ratio of 1.58 is desirable (Kosikowski *et al.,* 1997) whereas for Mozzarella cheese with 50% fat reduction, a ratio of 2.4 was suggested (Merrill *et al.*, 1994). Other primary cheese making parameters that may be manipulated include temperature of cooking, time of holding during cooking, pH at milling, and rate of salting (Johnson *et al.*,1995a).

2.8.2 Starter, adjunct cultures and enzymes

Starter organisms play an important role in cheese manufacture because of their contribution to proteolysis, texture and flavor development. Because of the altered microenvironment in low fat cheese, various alterations in microbial metabolism will occur which will influence cheese characteristics. Regular starters may be used for cheese manufacture e.g. mesophilic lactococci and thermophilic starters for low fat variants of Cheddar and Mozzarella, respectively, Starters selected for low fat cheese making should be able to undergo autolysis at the low cook temperatures and high cheese moisture content (Ardo, 1997). Adjunct cultures not only have a role in flavor development in ripened cheeses but may also be used to enhance functionality of low fat heese. For example, the photolytic activity of *Lb. casei* subsp. *casei* is also useful in development of functional properties of low fat Mozzarella cheese (Merrill *et al.*, 1996).

2.8.3 Fat replacers and other additives

Various additives are applied in low fat cheese making with a view to actually replacing the void left by fat in terms of its sensory and functional characteristics. These additives include various commercially available fat replacers as well as blends of stabilizers that help in moisture retention. Applications include a wide range of cheeses including soft cheeses, hard and semi-hard ripened cheeses, and functional cheeses such as Mozzarella and Process cheeses. Bullens *et al.*, (1994) reported on the blending of microcrystalline cellulose, carrageenan, and nonfat dry milk in cheese milk for Cheddar cheese of 11% fat. Cheese structure is softened by the interference of the casein–casein interaction by carregeenan and microcrystalline cellulose particles that function similarly to fat globules by imbedding within the curd matrix. Viscosity is attained by use of additives such as starch, hydrocolloids, carrageenan, and locust bean gum in the cream dressing (Kosikowski *et al.*, 1997).

2.9. FAT REPLACER

The prevalence of obesity and overweight in most Western countries has increased in recent decades. High dietary fat intake has been implicated in the aetiology of obesity and certain cancers. Obesity, in turn, is a risk factor for type 2 diabetes and hypertension. Furthermore, a high intake of saturated fats has been shown to elevate total blood cholesterol level, in particular, LDL-cholesterol level. High total and LDL cholesterol levels are key risk factors for coronary heart disease and ischemic stroke. Many expert groups worldwide have recommended a reduced intake of dietary fat, in particular, saturated fats. For example, in 1995, United States Dietary Guidelines recommend reducing total fat intake to not more than 30% of total daily caloric intake, with saturated fats reduced to not more than 10% of daily calorfic intake.

Health conscious consumers continue to look for ways to improve nutritional habits without sacrificing psychological satisfaction (Kostias, 1997). Consequently, health conscious individuals are modifying their dietary habits and eating less fat (Miller, 1996 and Cengiz, 2005). Consumer acceptance of any food product depends upon taste and consumers want foods with minimal calories (Caceres, 2004). Saturated fat intake is associated with high blood cholesterol and coronary heart disease. High fat intake is associated with increased risk for some types of cancer (Krauss, 2001 and Poppitt, 1995). Consumption of a diet rich in fat has been identified as a risk factor for excess energy intake, positive energy balance and the development of obesity (Thomas, 1992 and Siggaard, 1996). The development of low-fat foods with the same desirable attributes as the corresponding full-fat foods has created a distinct challenge to food industry (Zoulias, 2002 and Zalazar, 2002). Fat has functional properties that influence processing and the eating qualities of a food item. These functions must be accounted for when lowering the fat in a product (Kavas, 2004 and Koca, 2004). As a food component, fat contributes sensory and physiological benefits. Fat contributes to flavor, taste and aroma (Lucca & Tepper, 1994: Mistry, 2001 and Sampaio, 2004). Fat also contributes to creaminess, appearance, palatability and texture (Romanchik-Cerpovicz, 2002). Fat can also carry lipophilic flavor compounds which act as a precursor for flavor development and stabilize the flavor

(Romeih, 2002). Fat is the most concentrated source of energy in the diet, providing 9 kcal/g compared to 4 kcal/g for proteins and carbohydrates. Fat may be replaced in food products by traditional techniques such as substituting water (Chronakis, 1997). Air for fat, using lean meats in frozen entrées (Hsu, 2005), skim milk instead of whole milk (Zalazar, 2002) in frozen desserts (Specter, 1994), and baking instead of frying (Haumann, 1986) for manufacturing or preparing snack foods. Some lipids may be replaced in foods by reformulating with selected ingredients that provide some fat like attributes (Sipahioglu, 1999). Fats and oils impart many functional, nutritional and sensory properties in food products.

Fat interacts with other ingredients to develop texture, mouthfeel, structure and lubricity in foods (Giese, 1996). Fat also acts as a flavor carrier, enhancing the perception of taste. Nutritionally, fat aids in the absorption of fat-soluble vitamins: A, D, E, and K (Giese, 1996). Fat provides numerous qualities in foods. Fat replacement becomes a difficult (Jones, 1996). Fat replacers play a role in substituting some of fat's functions. Unfortunately, most fat replacers cannot recreate the functional and sensory properties of fat. Several carbohydrate, protein, or lipid-based ingredients individually or in combination is often used (Akoh, 1998). Fat replacers are generally categorized into two groups: fat substitutes and fat mimetics.

2.9.2 Fat mimetics

Fat mimetics are ingredients that have distinctly different chemical structures from fat. They are usually carbohydrate and/or protein-based. Fat substitutes are macromolecules that physically and chemically resemble triglycerides (conventional fats and oils) and which can theoretically replace the fat in foods on a one-to-one, gram-for-gram basis. Often referred to as lipid or fat-based fat replacers. Fat substitutes are either chemically synthesized or derived from conventional fats and oils by enzymatic modification. Many fat substitutes are stable at cooking and frying temperatures (Duflot, 1996 and Harrigan & Breene, 1989). They have diversed functional properties that mimic some of the characteristic physico chemical attributes and desirable eating qualities of fat: viscosity, mouthfeel and appearance (Johnson, 2001 and Harrigan, 1989).

2.9.2.1 Protein-Based Fat mimetics

Protein-based fat substitutes are derived from protein sources such as milk, egg, whey, or vegetable proteins (Giese, 1996). Some protein-based fat mimetics undergo microparticulation (sheared under heat), which produces microscopic round particles similar to fat particles that mimic the mouthfeel and texture of fat (Akoh, 1998). Simpless is a microparticulated protein-based fat mimetic that was given GRAS status in 1990 for use in frozen desserts and in 1994 for use in yogurt, cheese spreads, frozen desserts, cream cheese, and sour cream (Akoh, 1998). Simpless cannot be used in high-temperature food applications, which could easily denature the proteins. On a dry basis, Simpless provides 4 kcal/g, whereas a hydrated gel provides 1 kcal/g.

2.9.2.1.1 WPC

WPC besides being a nutritional ingredient in various food products can serve as functional components for binding, texturization, color development and whipping properties in a wide variety of food formulations. WPCs by virtue of their high protein content and functionally are widely used to replace traditional additives in various dairy products. It can replace milk powder in infant formulas, dietetic foods, ice cream, frozen beverages and desserts, meat products, pasta and Indian traditional milk products like khoa etc. (Patel and chakraborty, 1985). Olson & Johnson (1990) observed incorporation of undenatured whey proteins into low fat Gouda UF cheese which reduced its undesirable toughness.

2.9.2.2 Carbohydrate Based Fat Mimetics

For several years, carbohydrate based fat substitutes have been used to partially or fully replace fat. These mimetics are derived from cereals, grains and plants that include digestible and nondigestible carbohydrates (Giese, 1996). Digestible carbohydrates provide 4 kcal/g while non digestible carbohydrates provide negligible calories. Carbohydrate based fat substitutes include polydextrose, pectin, cellulose, gums, and starch derivatives (modified starches, dextrins). Carbohydrate based fat substitutes provide some of the functions of fat by binding water, and providing texture, mouthfeel, and opacity (Giese, 1996). Polydextrose is a bulking agent made by the random polymerization of glucose, sorbitol and citric acid. Polydextrose is an approved food additive often added to nonnutritive sweeteners to maintain texture, body and mouthfeel in low-calorie products (Glueck, 1994). It is also used as humectant to replace fat or sugar in baked goods, chewing gum, salad dressings etc. (Glueck, 1994). Cellulose based fat replacers are used to stabilize foams and emulsions, modify texture, increase viscosity and add dietary fiber (Giese, 1996). Gums or hydrocolloids are long chain polymers of monosaccharides that easily dissolve in water and produce thickening or viscosity increasing effects. Gums can be used with other gums or fat replacers to mimic the texture of fat. The most commonly used gums are guar, xanthan, locust bean gum, carrageenan, and gum arabic. Their thickening properties are used in salad dressings, soups and sauces, desserts and ice cream, dairy products, baked goods etc. Starches and starch derivatives provide many functions in fat replacements systems. When moist heat is applied to starch, the granules gelatinize, forming a mixture of thick, soft and creamy consistency. Starches are commonly derived from corn, potato, rice, wheat and tapioca. However, most starches are further modified when used in fat replacement systems. Starches are modified by acid or enzymatic hydrolysis to produce smaller polymers or by cross-linking or substitution. Starches are effective in high moisture systems such as salad dressings, low-fat spreads, meat emulsions, icings and fillings and baked goods.

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Chapter- 3

Scope and Plan of Work

3.0 SCOPE AND PLAN OF WORK

3.1 SCOPE

With modernization, there is a change in lifestyle as well as dietary habits. This resulted in the development of various healthy foods for the benefit of consumers. Cheese and cheese based products are one of them, and are gaining importance in the Indian food market. The food industry is also responding to the expectation of the consumers by delivering newer products with functionality, health benefits and additional nutrients. Likewise, Mascarpone cheese has arrived in the Indian market. The demand of soft fresh cheese (Mascarpone cheese) in India is increasing especially due to its taste and flavour as well as its use as butter substitute. Its mild flavour and soft texture allow it to be used in savoury and sweet dishes, making the possibilities in nearly endless items in the kitchen. Mascarpone cheese can be used for direct consumption, as a topping for desserts, with *roti*, *chapatti*, bread as spread. It can also be added to pasta for creamy finish, dishes to increase the richness and fresh cut fruits. Recipes for frozen desserts such as ice creams and cocktail sauces can be made with fresh cheese such as Quarg and Mascarpone.

High fat products, seems to cause cardio vascular diseases and other health problems and hence, there is higher demand of low fat and functional foods. The fat content of Mascarpone Cheese is high. High fat content in food is also linked with increase low density lipoprotein (LDL) cholesterol levels which in turn are linked to increase the risk of atherosclerosis and cardio vascular diseases (CVD). A reduction of fat level in the Mascarpone cheese by using suitable fat replacer may reduce the risk of atherosclerosis and CVD of the consumer.

Again, intake of dietary fibre & low fat products have reported to reduce the risk for developing coronary heart disease, stroke, hypertension, diabetes, obesity and certain gastrointestinal diseases. Therefore, the addition of dietary fibre and or suitable fat replacers to Mascarpone cheese may help to overcome these types of health disorders.

Mascarpone cheese contains high moisture and has low shelf life. This can be improved with ideal packaging and technological intervention.

Considering all these factors as mentioned above, the present study entitled "**DEVELOPMENT OF LOW FAT, FIBRE ENRICHED MASCARPONE CHEESE**" was planned to undertake with following objectives:

- **To replace the fat with suitable fat replacer and/or fibre**
- **To evaluate the quality characteristics of the developed Mascarpone Cheese**
- **To enhance the shelf life of the product**

3.2 PLAN OF WORK:

3.2.1 Preparation of Mascarpone Cheese

The following parameters were studied to prepare low fat Mascarpone Cheese

a) Types coagulating or acidifying agent

-Citric acid,

- Lactic acid,

-Acetic acid

b) Fat replacement

Levels: 40, 50 & 60 %

c) Replacers

Fat mimetic: WPC (protein base) and N-creamer

Fibre: Inulin

3.2.2 Evaluation of quality characteristics of low fat Mascarpone Cheese

a) Textural characteristics –Firmness, spreadability

b) **Sensory evaluation –** 9 point hedonic scale

c) **Analysis –** Fat, protein, carbohydrates, moisture, ash, acidity, pH and water activity

3.2.3Shelf life enhancement study of low fat Mascarpone Cheese

a) **Packaging:** Polyethylene pouch and polystyrene cup (100 gm)

b) **Temperature:** Refrigerated (7-8^{0}C)

3.2.4 Physico-chemical and microbiological analyses of the developed Cheese during storage

a) **Sensory evaluation –** 9 point hedonic scale

b) **Chemical analysis –** TBA, acidity, pH and changes in colour

c) **Microbial analysis-** Coliform count, yeast & mould counts.

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Chapter- 4

Materials and Methods

4.0 MATERIALS AND METHOD

This chapter deals with the various materials including ingredients and instruments used, methods employed for the preparation of sample and their analysis.

4.1 Materials

4.1.1 Milk

Milk was obtained from the Experimental Dairy of NDRI, Bangalore.

4.1.2 Cream

Cream was obtained from the Experimental Dairy of NDRI, Bangalore.

4.1.3 Fat replacer

Fat mimetic like WPC (Protein base) and N-Creamer (Carbohydrates base) were procured from local market.

4.1.4 Fibre

Fibruline Instant (Inulin) in white powder form was procured from M/s. Vilco Ingredients PVT. LTD, Mumbai and was used as fibre to the Mascarpone Cheese.

4.1.5 Chemical

Analytical grade chemical were procured from reputed firms for chemical analysis.

4.1.6 Packaging Material

Polyethylene pouches (65 u) & polystyrene cup (100 gm capacity) were procured from local market for the packaging of the Mascarpone Cheese.

4.1.7 Microbiological Media

Potato dextrose agar for yeast & mould counts and Mcconkey agar for coliform count were obtained from Hi-media, Bangalore and were used for microbiological analysis.

4.2 Equipments

4.2.1 pH Meter

Digital pH meter-Digsiun Electronics, Hyderabad, Model: DI707 was used to measure pH during the investigation.

4.2.2 Micro Kjeldhal Unit

Kjeldhal digestion unit and kjel plus distillation unit of Gerhardt Instruments were used to estimate nitrogen content.

4.2.3 Water Activity Meter

Water activity meter (Rotroni-Hygroskop, Germany) was used for the measurement of water activity. The equipment was stabilized before measurement of water activity.

4.2.4 Adobe Photoshop

Adobe Photoshop (RBGL scale 0-255) was used for the colour measurement of Mascarpone cheese.

4.2.5 Gerber centrifuge

Gerber centrifuge (Sanraj industries, New Delhi) was used to determine fat in milk and cream.

4.2.6 Spectrophotometer – Anthelie 2 Light, Secomam, France was used to measure TBA (Absorbance) during the investigation.

4.2.7 Texture Analysis

TA-XT plus, Stable Micro System, England was used for measuring the firmness and spreadability.

4.2.8 Glassware

Glassware of Borosil and Schott Duran brands were used for the chemical analysis of Mascarpone Cheese. They were thoroughly cleaned by detergent solution, rinsed under running water and dried in hot air oven before use.

4.3 Chemicals

Analytical Grade (AR) of chemicals from reputed standard companies were procured from the local market and was used by adopting standard procedures.

4.4 Other equipments

Vortex shaker, Electronic balance, Water bath, Incubator with Thermostatic control, Muffle furnace, Hot air oven, Air flow chamber, Muslin cloth, Refrigeration etc. of standard companies were used for the study.

4.5 Methods

Mascarpone Cheese was manufactured from fresh cow milk. The milk was standardized to fat content of about 11-12% for high fat (control) & 3-5% for low fat redundant. Cream of 50% fat was used for the standardization of fat content in cheese. Out of calculated amount total fat, 50% fat was added in milk before coagulation and remaining 50% fat in the form of pasteurized cream was added in coagulum. Milk was heated at 85-90 °C/10 min and was coagulated at 80°C by using diluted organic acid (2 or 4%). This coagulated mass cooled at room temperature by natural cooling (30 min). Whey was drained at from the coagulated mass using Muslin cloth. After removing maximum amount of whey, remaining 50% fat in the from of pasteurized cream was added in the coagulated mass and mixed well. It was kept in cold condition for overnight for further removal whey. Coagulated mass was taken out from the cloth, weighed and fat replacer in different proportion (4 -10% Inulin, 2- 8% WPC & 2-8% N-creamer) was added on weight basis to the cheese & mixed it well for smooth and homogenous body and texture. The cheese was packed either in polystyrene cup or polyethylene pouches and stored at 5-8^0 C.

4.6 Physico-chemical analysis

4.6.1 Fat in milk and cream

Fat in milk and cream were determined using Gerber method (IS: 18 Part XI, 1981).

4.6.2 pH

The pH of cheese was measured as described by Awad *et al*. (2005). The combined glass electrode was calibrated against standard buffer of pH 7.0 and 4.0. Approximately 20 g Cheese was mixed with 20 ml distilled warm (35-40°C) water & slurry was prepared. pH of cheese was measured directly by inserting the electrode into the slurry

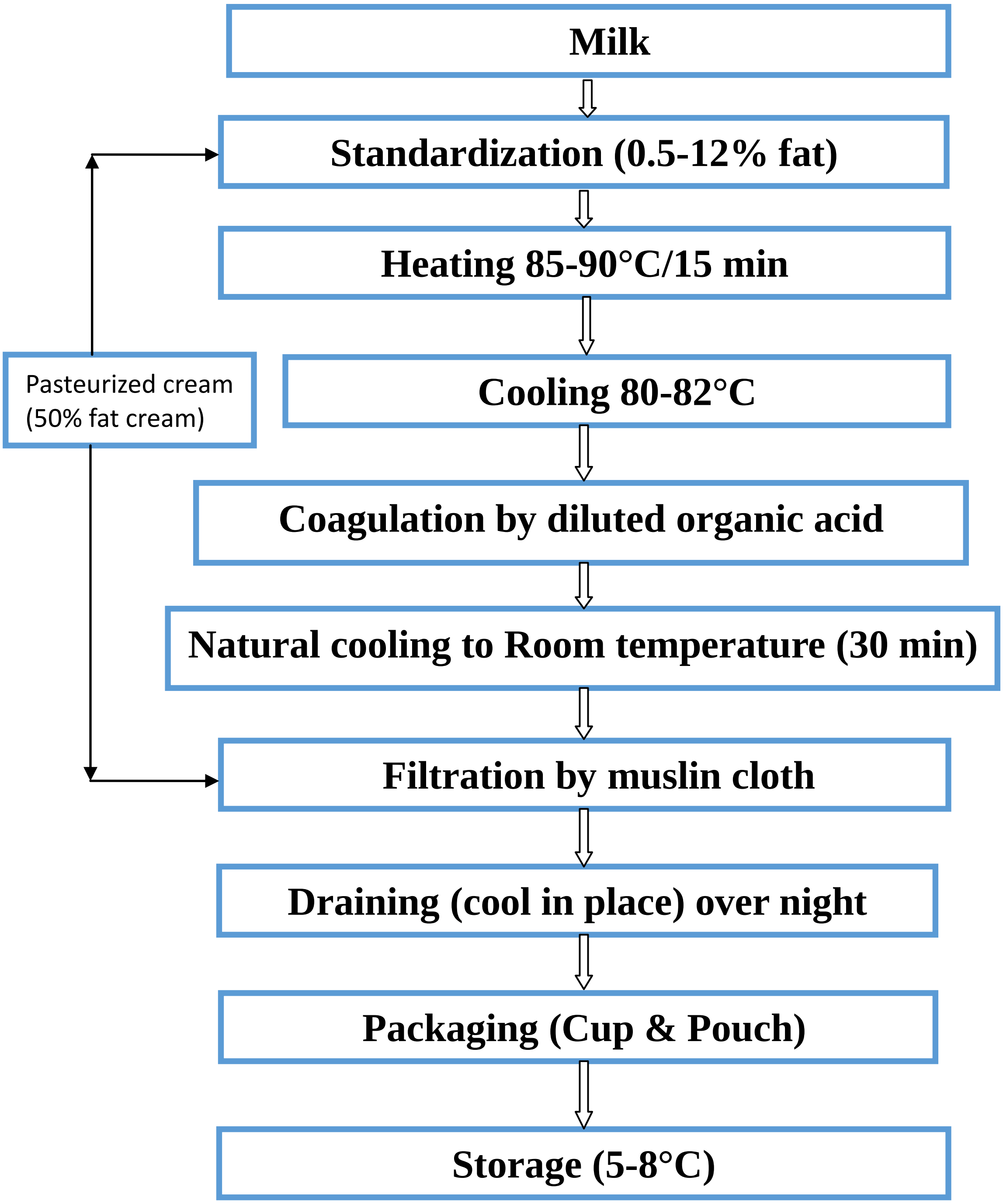

Figure 4.1: Flow diagram for the manufacture of Mascarpone cheese

4.6.3 Acidity

Acidity in Mascarpone cheese was determined by titration method No. 920.124 of AOAC (1990). One gram of cheese sample was mixed with 10 ml warm water (40°C) in 100 ml conical flask, was shaken vigorously and filtered. The filtrate was titrated with 0.1 N NaOH using phenolphthalein as indicator. Acidity was calculated by using the following expression:

Per cent (TA) = 0.0090 × volume of NaOH used x 100/ weight of the sample in g.

4.6.4 Water Activity

The water activity meter was allowed to stabilize for 10 min. Cheese sample 15-20 g sample was taken in a dish and fan of the meter was switched on. Water activity was measured directly from the instrument.

4.6.5 Thiobarbituric acid (TBA) value

The extent of oxidation of fat in product was measured in terms of TBA value as outlined by Sidwell *et al*. (1955).

Mascarpone cheese of 10 gm was transferred into Kjeldahl flask. Sixty ml of water was added and the sample was properly dispersed. Thereafter, 7 ml of 3 N HCl was added and mixed thoroughly (the ratio of sample to water was so adjusted as to get the pH of the mixture between 1.5 – 2.0 after the addition of 3 N HCl). The contents of the flask were then distilled and 50 ml distillate was collected. Distillate of 20 ml was pipette out into a 60 ml glass stoppered tubes and 2 ml of TBA reagent (0.67 g in 100 ml glacial acetic acid) was added in it. The tubes were stoppered, shaken and placed in boiling water bath for 35 min. The contents were cooled to room temperature and absorbance was measured at 530 nm. Blank determinations were made using distilled water in place of sample. TBA absorbance reading was noted down directly from the instruments.

TBA value was expressed as absorbance at 530 nm.

4.6.6 Reflectance

Colour of the sample was measured in terms of reflectance/lightness by using Adobe photoshop The sample was taken in a clean small petri-plate and then scanned by a scanner. The scanned image was taken in adobe photoshop where whiteness was measured the in term of lightness. The same petri-plate was used throughout the study. The results are expreseed in luminosity value of 0-255 RGB scale.

4.7 Microbial analysis

The enumeration of coliform count and yeast & mould counts were carried out as per procedure given in IS: (SP: 18, Part IX, 1981)

4.7.1 Coliform count

Mascarpone cheese sample of 11 g was transferred aseptically in to 99 ml sterile saline. Thus a 1:10 dilution was obtained. Further dilution was carried out serially by transferring 1 ml of diluted sample to test tube having 9 ml sterile saline. The dilutions were prepared so as to give 30 to 300 colonies per plate from one ml of the dilution which was poured into sterile petri plate. Duplicates were prepared.

The sterilized agar medium (mcConkey agar) was melted and kept at 48-50°C. The medium was introduced aseptically at 42-44°C into petri plate and spreaded by rotating the plates. Care was taken to prevent spreading over the edges. The mixture was evenly distributed over the bottom of the plate. It was then allowed to solidify. The plates were inverted and incubated at 37°C for 48 hours. Thereafter count was taken and expressed as colony forming units per gm.

4.7.2 Yeast and mould counts

Mascarpone Cheese sample of 11g was transferred into 99 ml of sterile saline aseptically. Thus, a 1:10 dilution was obtained. Further dilution to desired level was carried out serially by transferring 1ml of diluted sample to test tube having 9 ml sterile saline. The dilutions were prepared so as to select the desired dilution give between 30 and 300 colonies per plate after incubation. One ml of the dilution was poured into a sterile petri plate. Duplicates were prepared.

The sterilized agar medium (Potato Dextrose Agar) was melted and was kept at 48-50^{0}C. The medium was introduced aseptically at 42 to 44^{0}C into petri dishes and mixed by rotating the dishes. Care was taken to prevent spreading over the edges. The mixture was spread evenly over the bottom of the plate. It was then allowed to solidify. Yeast and mould counts were determined by plating 1:10, and 1:100 dilutions using potato dextrose agar, the pH of which was adjusted to 3.5 by adding 1-2 drops of sterile tartaric acid solution (10%) to each plate before pouring the medium. The count was taken after 2-3 days of incubation at 25-30^{0}C and results were expressed as colony forming units per gm.

4.8 Compositional analysis

4.8.1 Moisture

Moisture content of cheese was determined by gravimetric method (IS, SP: 18 Part XI, 1981). About 3 g of grated cheese was weighed accurately and transferred to a previously dried, tarred aluminium dish containing dried sea sand (10-15 g) and a glass rod. The cheese was uniformly spreaded using the glass rod so that the sample was covered with sea sand and weighed accurately. The sample was dried at 102 ± °C for 4 hours. It was cooled and weighed. The process was repeated by re-drying for 1 hour until change in weight between two successive drying was less than 0.5 mg. The moisture was calculated as below:

$$\text{Moisture \%} = \frac{W_1 - W_2}{W_1 - W} \times 100$$

Where,

W= weight in g of the dish+ sand+ glass rod.

W_1= weight in g of the dish+ sand+ glass rod and cheese sample.

W_2= weight in g of the dish+ sand+ glass rod+ cheese after drying.

4.8.2 Fat Mojonnier-Gravimetric Method

Fat in Cheese was determined as per the method AOAC 933.05, 2005.

First extraction: Weigh accurately about 1 g of grated cheese sample into dried beaker to which 9 ml of water was added followed by 10 ml of concentrated HCL and then it was placed in boiling water for 20 minutes. The contents of the beaker was transferred to Mojonnier extraction tube and allowed to cool. 10 ml ethanol was added into the extraction tube followed by addition of 25 ml diethyl ether and 25 ml petroleum ether (40-60°C). The mixture was mixed thoroughly after each addition of the solvents for 90 seconds. This was allowed to stand for 30 minutes undisturbed and thereafter the clear supernatants were decanted into pre-dried and weighed beaker. Addition of ethanol prevented gelation of sample matrix with ether and aids in ether-water phase separation. Ethyl ether and petroleum ether serve as lipid solvents. Petroleum ether decreases the solubility of water in the ether phase and is selective for hydrophobic lipids. After clear separation of solvents, the supernatant was transferred to a previously dried and weighed fat dish.

Second extraction: Five ml ethanol, 15 ml diethyl ether and 15 ml petroleum ether were added in order, after each addition of solvent the tube contends was mixed by shaking for 30 seconds. The solvent and water phase were allowed to separate and supernatant was carefully transferred to fat dish.

Third extraction: Avoiding ethanol, 5 ml of each diethyl and petroleum ether were added in same manner to fat dish. The solvent mixture was evaporated over boiling water and the final trace were removed using hot plate and was dried for 1 hour in hot air oven at 100°C to remove traces of water. Cooled in desiccators and weighed accurately.

$$\% \text{ Fat} = \frac{\text{Weight of fat residue}}{\text{Sample weight}} \times 100$$

4.8.3 Ash

Ash content in cheese was determined as per the method described in AOAC 935.42, (2005). Cheese sample of 3-5 g of was weighed into dried crucible and ignited to ≤550°C in a Muffle furnace, cooled and weighed. Samples were dried for 1 hr. prior to ashing over water bath)

$$\% \text{ Ash} = \frac{\text{Weight after ashing} - \text{Weight of crucible}}{\text{Sample weight}} \times 100$$

4.8.4 Total Protein

Total nitrogen or total protein in cheese was determined as per the method described in AOAC 2001.14, (2005).

Approximately 0.2-0.3 g of cheese sample was weighed accurately on a butter paper and transferred carefully along with the butter paper to 300 ml Kjeldahl flask. Two gram of digestion mixture (K_2SO_4 and $CuSO_4$), followed by 15-20 ml distilled water and 12.5 ml of concentrated sulphuric acid (AR) were added to the flask. The contents were digested over block digester until clear and colourless solution was obtained. After cooling, the Kjeldhal flask was washed with 10-15 ml of distilled water and was transferred to the Kjeldhal distillation unit. Twenty ml of 40% NaOH was added to make the solution alkaline. The contents were steam distilled and the liberated ammonia was collected in 25 ml of 2% boric acid solution containing 2-3 drops of the mixed indicator (methyl red and methylene blue). The distillation was continued until about 100-120 ml of distillate was collected. The distillate was titrated against N/10 H_2SO_4 to light pink colour. A blank test was carried out simultaneously using all the reagents except the test material. Per cent protein was calculated as follows:

$$\textbf{\% Nitrogen} = \frac{1.4007 \times (A-B) \times N}{\text{Sample weight in g}}$$

Whereas-

A= Sample titrate value, B= Blank titrate value N= Normality of H_2SO_4 used.

%Total Protein = % Nitrogen x 6.38

6.38 is the conversion factor of milk and milk products

4.8.5 Total carbohydrate

The total carbohydrate content of the sample was obtained by difference i.e. Total carbohydrate (%) = 100 – (% Moisture + % Fat + % Total protein + % Ash).

4.9 Rheological characteristics

4.9.1 Texture Profile Analyser

The textural analysis of Mascarpone Cheese was done by using Texture Profile Analyser. as shown in Figure 4.2. The following TPA step was followed.

1) Texture exponent programme was opened and following project setting was loaded as mentioned in Table 4.1.

2) Heavy duty platform was placed on the base of the machine.

3) P/60 Probe selection and calibration of the probe height was done. A typical texture profile analyzer curve is shown in Figure 4.3.

4) Cheese was place to the empty cone holder and slightly pressed it down to eliminate air pockets. Scrape off any excess sample with knife/ spatula to leave a flat test area. Cone tip was brought down to the sample surface. Then click “TA” and “run a test” option was started with filling details such as name, batch number, saving option etc. A force Vs time plot was displayed on the monitor corresponding to the cone probe movement into the sample and withdrawal thereafter.

5) Maximum positive force on positive peak was determining a firmness of sample and spreadability was inversely proportional to the area of positive peak.
6) Table 4.1: Setting specifications of Texture Profile Analyser

Test mode	:	Compression
Pre-test speed	:	1.0 mm/sec
Test speed	:	3 mm/sec
Post-test speed	:	10mm/sec
Target mode	:	Distance
Distance	:	18 mm
Trigger type	;	Button
Stop plot at	:	Start position
Tare mode	:	Auto
Advance options	:	on
Control oven	:	Disable

4.10 Statistical analysis

Data obtained from various analyses of cheeses were statistically analysed by using SPSS software.

4.11 Sensory evaluation

The organoleptic qualities of low fat, fibre enriched Mascarpone Cheese was evaluated by an expert panel. The evaluation was carried out room temperature under proper lighting. The judged parameters were: flavour, body & texture, colour and appearance and overall acceptability.

Fig 4.2: Texture Analyzer instrument

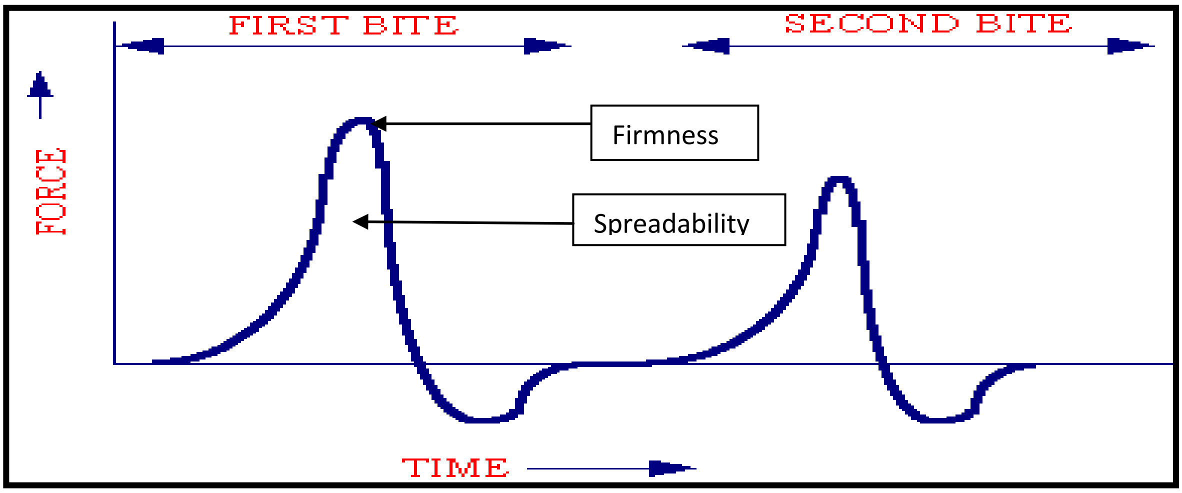

Fig 4.2: Texture profile analysis curve

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Chapter- 5

Results and Discussion

5.0 RESULTS AND DISCUSSION

5.1 PROLOGUE

Investigations were carried out to develop low fat fibre enriched Mascarpone cheese and also to analyse the physic-chemical, sensory and microbial properties of the Mascarpone cheese. Attempts were also made to assess the shelf life of developed low fat Mascarpone cheese at refrigeration temperature.

5.2 OPTIMIZATION OF PROCESSING PARAMETERS

The following parameters were optimized for the development of low fat fibre enriched Mascarpone cheese

5.2.1 Process optimization of cream addition at different stages

5.2.2 Selection of acid for coagulation

5.2.3 Effect of inulin (fibre) incorporation on Mascarpone cheese

5.3 FAT REDUCTION

5.3.1 Optimization of fat reduction level and fat replacer level in Mascarpone cheese

5.4 PREPARATION OF STANDARDIZED LOW FAT FIBRE ENRICHED MASCARPONE CHEESE

5.5 EVALUATION OF QUALITY CHARACTERISTICS OF THE DEVELOPED LOW FAT FIBRE ENRICHED MASCARPONE CHEESE

5.6 SHELF LIFE STUDIES OF DEVELOPED LOW FAT, FIBRE ENRICHED MASCARPONE CHEESE

5.2 OPTIMIZATION OF PROCESSING PARAMETERS

5.2.1 Process optimization of cream addition at different stages

In order to optimize the process, 50% fat cream was added either in milk or in coagulum or in combination to obtain 40% fat in final Mascarpone cheese (Table 5.1). Cream was added in the product at three different stages to obtain 40 % fat in the final product. First, cream was added in milk to standardize 11-12% fat before coagulation. Second, 50% fat was added in milk to standardize 5-6% fat before coagulation and balance 50 % fat was added in coagulum after coagulation. Third, full amount of fat was added in coagulum after coagulation. In all the three occasions, milk was coagulated with 4% lactic acid solution at 80-82^{0}C. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the form of pasteurized cream was added in the coagulated mass and mixed well. It was kept in cold condition for overnight for further removal whey. Coagulated mass was taken out from the cloth, weighed and analyzed. Three different stages of cream addition were compared for yield, moisture retention, fat loss in whey and sensory characteristics of products.

<u>5.2.1.1 Influnces on physico-chemical parameters</u>

It was observed that addition of 50% fat in milk and 50% fat in coagulum resulted in highest yield and lowest fat loss in whey than those of two other stages of fat addition (Table 5.1). Higher yield was significantly ($p<0.05$) more in 50-50% than the entire fat addition in milk. The lowest moisture content was observed in the cheese when entire amount of cream was added in milk before coagulation. Moisture content of three different methods of cream addition has no statistically significant different ($p<0.05$) as shown in Table 5.1. Since the moisture content was lowest, the yield was lowest and the product became granular. When the entire amount of cream was added in coagulum (third process), it was difficult to mix the 100% cream in the coagulum uniformly and the product was rubbery. Mascarpone cheese obtained from the 50 % fat addition in milk and 50% fat in coagulum showed good appearance and body & texture because of 50% fat addition in

Table 5.1 Effect of cream addition at different stages of Mascarpone cheese Preparation on yield, moisture, fat loss and sensory parameters

PARAMETER	Before coagulation	50-50%	After coagulation
Yield (%)	25.06±2.20[a]	28.38±0.79[b]	27.0±1.60[ab]
Moisture (%)	51.02±3.49[a]	54.75±2.95[a]	53.12±1.58[a]
Fat loss in whey (%)	1.21±0.36[a]	1.56±0.28[a]	1.79±0.11[b]
SENSORY PARAMETER			
Color& appearances	7.26±0.55[ab]	7.42±0.51[b]	7.11±0.44[a]
Body& Texture	7.29±0.50[a]	7.35±0.66[a]	7.22±0.43[a]
Flavor	7.13±0.61[a]	7.42±0.56[b]	7.01±0.51[a]
Overall acceptance	7.25±0.53[ab]	7.44±0.59[b]	7.13±0.41[a]

Means with different superscript in a row differ significantly (p<0.05)

Table 5.1a Anova for physico-chemical parameters

		Yield		Moisture		Fat loss	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Between the Methods	2	13.99	5.204*	17.470	2.230	0.444	6.669*
Error	12	2.689		7.802		0.067	
Total	14						

**Significantly differ at levels of 5% (p<0.05)*

Table 5.1b Anova for sensory parameter

		C&A		B & T		Flavor		OA	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Between the Methods	2	1.18	4.57	0.20	0.70	2.19	6.80*	1.21	4.47
Error	147	0.25		0.29		0.32		0.27	
Total	149								

**Significantly differ at levels of 5% (p<0.05)*

Milk inhibited rubbery body & texture formation and 50% fat addition in coagulum produced smoothness in the product.

5.2.1.2 Influence of addition of fat at different stages on sensory characteristics

Table 5.1 reveals that cheese made by addition of 50 % fat in milk and 50% fat in coagulum using pasteurized cream of about 50% resulted in a best product in terms of good appearance, homogeneous body & texture due to uniform mixing of fat among three stages of fat addition during processing. This may be due to the addition of 50% fat in the from of coagulated mass where a small amount fat got adsorbed on the surface of the curd particle and facilitated in smoothening of the final product. Good quality of Mascarpone cheese using ultra-filtration technology decreased time of preparation and heating by plate exchangers rather than by steam injection, improved product quality where cream dressing was added in cheese for fat adjustment (Pagani *et al.,* 1995). Flavor quality of the cheese made with 50% fat in milk and 50% fat in coagulum showed significantly ($p<0.05$) more than the other two stages of fat addition. Overall acceptance score was significantly ($p<0.05$) more than the other two stages of fat addition i.e. all fat in milk and all fat in coagulum.

Hence, based on physical and sensory characteristics evaluations, the Mascarpone cheese made by addition of 50% fat in milk and 50% fat in coagulum was adjudged the best among three stages of fat addition for Mascarpone cheese preparation. Therefore, this method of manufacturing process was subsequently used for Mascarpone cheese.

5.2.2 Effect of various acids on physic-chemical & textural parameters of Mascarpone cheese

To identify the ideal acid to be used to manufacture good quality of Mascarpone cheese, three different acids of acetic, citric and lactic acid were used to coagulate milk. Milk was standardized to 5-6% fat with 50% fat cream. Acid solutions of 4% were used to coagulate milk at 80-82^{0}C. The quantity of acid used to complete the coagulation process for the manufacture of Mascarpone cheese was different. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out

Table 5.2: Influence of different acids used on Mascarpone cheese preparation on physical and rheological properties of cheese

PHYSICAL PARAMETER	Acetic Acid	Lactic Acid	Citric Acid
Yield (%)	27.12± 2.09^{a}	29.22 ±3.09^{a}	27.92 ±3.74^{a}
Moisture (%)	48.70 ±3.94^{a}	55.77 ±2.75^{b}	55.24 ±3.63^{b}
Fat loss in whey (%)	1.11± 0.13^{b}	0.85± 0.028^{a}	1.28±0.25^{b}
Speradability (N.s^{-1})	1.90 ±0.16^{a}	2.19 ±0.075^{b}	1.75 ±0.13^{a}
Firmness (N)	57.58± 1.66^{b}	45.08 ±2.03^{a}	51.22± 2.70^{b}
SENSORY PARAMETERS			
Color & Appearance	7.19±0.60^{a}	7.54±0.48^{b}	7.39±0.54^{ab}
Body & Texture	6.73±0.86^{a}	7.52±0.67^{b}	7.36±0.60^{b}
Flavor	7.11±0.62^{a}	7.42±0.65^{ab}	7.49±0.54^{b}
Overall acceptability	6.93±0.73^{a}	7.50±0.58^{ab}	7.25±1.11^{b}

Means with different superscript in a row differ significantly (p<0.05)

Table 5.2a Anova for physico-chemical and textural parameters

		Yield		Moisture		Fat loss		Spreadability		Firmness	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Between the Acid	2	5.618	0.601	109.70	9.050*	0.233	8.081*	0.246	14.509*	195.33	41.162*
Error	12	9.343		12.122		0.029		0.017		4.745	
Total	14										

**Significantly differ at levels of 5% (p<0.05)*

Table 5.2b Anova for sensory parameters

		C & A		B & T		Flavor		OV	
Source of variation	**Df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Types of Acid	2	1.557	5.20*	8.722	16.55*	2.083	5.60*	4.095	5.77*
Error	147	0.299		0.527		0.372		0.709	
Total	149								

**Significantly differ at levels of 5 % (p<0.05)* from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the from of pasteurized cream was added in the coagulated mass and mixed well to obtain 40% fat in the final product. It was kept in cold condition for overnight for further removal of whey. Coagulated mass was taken out from the cloth, weighed and analyzed. Addition of three different acids of acetic, citric and lactic acid (coagulants) were compared for physic-chemical and sensory parameters of products obtained from different coagulations.

5.2.2.1 Influence of various acids on physico-chemical and textural parameters of mascarpone cheese

There was definite differences between each batches of Mascarpone cheese due to the different acids used for coagulation. The quantity of 4% acid solutions used to coagulate about one liter of milk were 25-28 ml, 32-35 ml and 35-40ml. Highest quantity of lactic was utilized to complete the coagulation because it is the weakest acid among acetic, citric and lactic acids.

Mascarpone cheese manufactured using lactic acid showed maximum yield, moisture retention and minimum fat loss in whey followed by citric acid coagulation and thereafter acetic acid coagulation (Table 5.2). Although the yield of Mascarpone made by using lactic was about 1.5% higher, but the higher yield did not differ significantly ($p>0.05$) from the cheese made by using either acetic or citric acids. Among the three acids used for coagulation, significantly higher moisture content and significantly lower fat loss in whey observed when lactic acid was used as coagulant. Speradability of cheese made by using lactic acid cheese was significantly ($p<0.05$) more than the cheese made by using citric and acetic acids. Significantly ($p<0.05$) lowest firmness was observed in the cheese made with lactic acid followed by coagulation with citric and acetic acid. These observations are in contradict with observations made by Buratto (2010) where use of vinegar showed 1% more yield and higher fat recovery than that of cheese made with citric acid. Use of citric acid was recommended by Buratto (2010) due to better textural characteristics to manufacture Mascarpone cheese.

5.2.2.2 Influences on sensory characteristics

Sensory scores showed that there was no significant ($p>0.05$) difference in all the parameters between the cheese made by lactic acid and citric acid whereas these parameters were significantly different between the cheese made with lactic acid and acetic acid. Mascarpone cheese made with lactic acid resulted in softest body, easily spreadable and clean taste whereas cheeses with citric acid and acetic acid showed tangy taste and higher firmness. Cheese made with lactic acid obtained maximum sensory scores in all sensory parameters except flavor.

Based on the results obtained from physical and sensory parameter of the cheeses made with acetic, citric and lactic acids, cheese made with lactic acid was the best . Therefore, lactic acid was selected for ideal coagulant to manufacture further studied on Mascarpone cheese.

5.2.3 Effect of inulin (fibre) incorporation in Mascarpone cheese

Fibre has great role in gastrointestinal tract. It helps to reduce amount of cholesterol and triglycerides deposition, reduces the constipation and favours the microbiota present in intestinal tract and inhibits growth or activities of certain pathogenic bacteria, thereby promoting colonic health. Inulin is considered as ideal soluble fibre with beneficial attributes in human physiology. Today, inulin is used as an active ingredient for functional foods. Solowjiej *et al* (2015) suggested that milk fat in processed cheese analogues can be partially replaced with inulin to improve the functional properties of the final product. Inulin is not digested by enzymes in the human alimentary system, contributing to its functional properties: reduced calorie value, dietary fibre and prebiotic effects. Without color and odor, Inulin has little impact on sensory characteristics of food products. Inulin is increasingly used in processed foods because it has unusually adapTable characteristics. Its flavor ranges from bland to subtly sweet (approx. 10% sweetness of sugar/sucrose). It can be used to replace sugar, fat and flour. This is advantageous because inulin contains 25-35% of the food energy of carbohydrates (starch, sugar). In addition to being a versatile ingredient, inulin has many health benefits.

Considering above benefits of inulin, an attempt was made to incorporate inulin in Mascarpone cheese for better health benefit. Therefore, Mascarpone cheeses were made with the incorporation 4, 6 and 8% of inulin on weight basis of the cheese. Milk was standardized to 5-6% fat with 50% fat cream and was coagulated with 4% lactic acid solution at 80-82^{0}C. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the form of pasteurized cream was added in the coagulated mass and mixed well to obtain 40% fat in the final product. It was kept overnight in cold condition for further removal of whey. Coagulated mass was taken out from the cloth and weighed. Heat processed and cooled inulin was incorporated at 4, 6 and 8% of the coagulated and mixed them homogeneously. Moisture content, spreadability, firmness and sensory characteristics of products obtained were compared. A control Mascarpone cheese was manufactured to obtain 40% fat in cheese in the same way without addition of inulin.

Table 5.3 Effect of different levels of inulin addition of Mascarpone cheese preparation

PHYSICAL PARAMETER	Control	4%	6%	8%
Moisture (%)	57.65±3.22[b]	55.44±2.41[b]	54.69±2.77[ab]	51.31±3.69[a]
Spreadability ($N.s^{-1}$)	1.99±0.12[a]	2.81±0.13[b]	3.35±0.09[c]	3.33± 0.14[c]
Firmness (N)	77.20±4.48[c]	55.96±3.90[b]	47.35±3.06[a]	48.66±3.49[a]
SENSORY PARAMETER				
Color & Appearance	7.96 ±0.33[b]	7.63± 0.51[a]	7.90± 0.46[ab]	7.65± 0.53[a]
Body & Texture	7.86 ±0.40[b]	7.49 ±0.48[a]	7.99± 0.44[b]	7.59 ±0.47[a]
Flavor	7.78± 0.37[a]	7.58 ±0.57[a]	7.69± 0.74[a]	7.48 ±0.53[a]
Overall acceptability	7.79 ±0.42[ab]	7.51± 0.52[a]	7.89± 0.61[b]	7.56 ±0.51[a]

Means with different superscript in a row differ significantly (p<0.05))

Table 5.3a Anova for physic0-chemical and textural parameter

		Moisture		Spreadability		Firmness	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Between the Inulin levels	3	34.537	3.671	2.020	128.999*	952.279	66.767*
Error	16	9.407		0.016		14.263	
Total	19						

**Significantly differ at levels of 5%*

Table 5.3b Anova for sensory parameter

		C & A		B & T		Flavor		OA	
Source of variation	df	MS	F	MS	F	MS	F	MS	F
Between the Inulin levels	3	0.702	3.162	1.323	6.453*	0.436	1.348	0.815	3.024
Error	96	0.222		0.205		0.323		0.270	
Total	199								

**Significantly differ at levels of 5%*

5.2.3.1 Effect of inulin incorporation on physico-chemical and textural parameters

The effect of different levels of inulin incorporation on physical parameters of Mascarpone cheese is presented in Table 5.3. Moisture content of cheese decreased significantly with increases of inulin levels. However, this decrease was non significant ($p>0.05$) among the control, 4 and 6% incorporation of inulin. This decrease was due to the dry blending of inulin cheese. Water retention capacity of inulin could increase the water available for salvation of the protein chains, resulting in a softer, more easily deformed cheese (Creamer & Olson 1982). The spreadability was found to increase with the increase in inulin incorporation. Spreadability increased significantly more with the increase in inulin incorporation except for 8% level. Highest spreadability was observed in the cheese with 6% inulin and it was lowest in control samples. It was observed that the spreadability of cheese increased with increasing the inulin content up to 6% but thereafter the spreadability started decreasing as it was observed in case of 8% inulin incorporation. Firmness was observed to decrease with the increase in inulin content of the cheese. This decrease in firmness differed significantly among the control and three levels inulin incorporation except between 6 and 8% levels of inulin incorporation.

Moisture content of cheese was affected with the level of inulin incorporation. Our observation is well matched with the observation of Dave (2012), found higher spreadability with the addition of 6-7% inulin in low fat processed cheese spread. Incorporation of inulin led to improvement in the texture in low-fat cheese by decreasing hardness and gumminess while maintaining cohesiveness, adhesiveness and springiness (Wadhwani, 2011).

5.2.3.2 Effect of inulin incorporation on sensory parameters

The effect of inulin levels on color and appearances score of cheese is shown in Table 5.3. The average color & appearances scores of control, 4% inulin, 6% inulin and 8% inulin were 7.96, 7.63, 7.90 and 7.65 respectively. The control sample had a better score compared to others because of non blending of inulin. Control sample scored maximum score of 7.96 followed by 7.90 for 6% inulin incorporated cheese and 7.65 for 8% and 7.63 for 4% inulin added cheese. Addition of inulin in cheese improved the spreadability and smoothness as it is evident in Table 5.3. Highest spreadability was observed in the cheese with 6% inulin, scored maximum score of 7.99. Body and texture did not differ significantly ($p>0.05$) between control sample and 6 % inulin added sample whereas the difference significant ($p<0.05$) between 4 and 8% inulin added cheese. Cheeses manufactured with 3% of inulin were characterized by a more compact structure, denser protein matrix and more uniform disposition of protein chains and the pores between them compared to other cheeses (Miocinovic *et al.*, 2011). The average flavor score of all samples is near about same as shown in Table 5.3. Addition or incorporation of inulin in cheeses has no effect on the flavor of sample but slightly sweetest taste arose at time of tasting. No statistically significant different ($p>0.05$) observed in all samples. The average over all acceptability score of cheese shown in the Table 5.3. Acceptability of cheese is the summation of all color & appearances, body & texture and Flavor. Since 6% inulin sample had better color & appearance and body & texture it which resulted in higher score than other samples. Higher sensory score in the cheese was due to the good spreadable and lees firm because of inulin addition. Whereas, Raju (2009) during incorporation of different levels (1, 2, 3 and 4.5%) of inulin in misti dahi found that inulin at higher levels of incorporation (4.5%) significantly reduced the body and texture score than control. Up to 4.5% addition of inulin although decreased the overall acceptability score, but the product was still accepTable.

Based on physico chemical and sensory parameter analysis, 6% incorporation of in inulin cheese was adjudged the best among three levels of incorporation.

5.3 FAT REDUCTION

A low-fat diet is one that restricts fat and often saturated fat and cholesterol as well. Low-fat diets are intended to reduce diseases such as heart_disease and obesity. Reducing fat in the diet can make it easier to cut calories. Fat provides nine calories per gram while carbohydrates and protein each provide four calories per gram, so choosing low-fat foods makes it possible to eat a larger volume of food for the same number of calories. The Institute of Medicine recommends limiting fat intake to 35% of total calories to help for preventing obesity and to help control saturated fat intake.

Mascarpone cheese contains high fat. The consumption of fat is linked to increase low density lipoprotein (LDL) cholesterol levels, which in turn is linked to increase risk of atherosclerosis and cardio vascular diseases (CVD). Therefore, reduction of fat level in Mascarpone cheese by using suiTable fat replacer reduces the risk of atherosclerosis and CVD of the consumers. Therefore, attempts were made to reduce fat levels of 40, 50 and 60% with the addition fat replacers of 10, 15 and 20% in the final Mascarpone cheese. To achieve this study, cheese were made as per the followings:

5.3.1 Optimization of fat reduction levels (40, 50 and 60% fat reduction) in Mascarpone cheese with addition of fat replacer (10, 15 and 20%)

Fat reduction in Mascarpone cheese is an important step as reduction of fat has influences on flavor and moisture content in the product. Therefore, to determine the optimum level of fat reduction in cheese with addition of 10, 15 and 20% fat replacer, effects of each level of fat reduction was studied keeping constant addition of three levels fat replacer.

5.3.1.1 Effect of 40% fat reduction with addition of 10, 15 and 20% fat replacers (w/w) on the physico-chemical, textural & sensory characteristics of Mascarpone cheese

Milk was standardized to 4-5% fat with 50% fat cream and was coagulated with 4% lactic acid solution at 80-82^{0}C. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the form of pasteurized cream was added in the coagulated mass and mixed well to obtain 24% fat in the final product. It was kept in cold condition for overnight for further removal of whey. Coagulated mass was taken out from the cloth and weighed. Three mixtures of fat replacers were heat processed and cooled. Fat replacers (FR) of 10% (6% inulin, 2% WPC & 2% N-creamer), 15% (6% inulin, 4.5% WPC & 4.5%N-creamer) and 20% (6% inulin, 7% WPC & 7% N-creamer) were added in Mascarpone cheese of 24% fat and mixed them homogeneously. Moisture content, pH, color, spreadability, firmness and sensory characteristics of products obtained were compared. A control Mascarpone cheese was manufactured using 50% fat in milk and 50% fat incorporation in coagulum to obtain 40% fat in final product without addition of fat replacers.

5.3.1.1.1 Effect on physico-chemical and textural parameters

The effects of 40% fat reduction with addition of different levels of FR on the physico-chemical and textural parameters of Mascarpone cheese is presented in Table 5.4. Moisture content, pH, color, spreadability and firmness were determined. It was observed that with the addition of FR, the moisture content in cheese decreased. Higher as the level of incorporation of combined fat replacer increased as it is evidence moisture content in the cheese decreased but higher than control due to the dry blending of the fat replacer was the addition, lower was the moisture in cheese. The cheese with 10% FR showed significantly higher moisture than control and other two levels of FR. In general low fat cheese samples had higher moisture content as it is shown in Table 5.4. However, as a result shown Table 10% fat replacer addition showed significantly difference from other sample including control also. The pH of Mascarpone cheese showed a direct relation to the quantity of fat replacer used in the cheese. The control sample showed a pH 5.23±0.015 where as pH of the cheeses made by addition of 10, 15 and 20% FR were 5.30±0.036, 5.33±0.12 and 5.43±0.124 respectively indicating addition of FR has directly relation with pH value. Higher was the quantity of FR, higher was the pH value of the cheese. The whiteness of cheese was significantly ($p<0.05$) increasing with the levels of FR addition and control. Whiteness of cheese mainly depends upon type milk and cream or per cent fat as β-carotene lowers the whiteness in cheese and this β -carotene is fat soluble compound. The spreadability value of control and 10, 15 & 20% FR added cheeses were 2.03, 1.64, 1.38 and 1.17 ($N.S^{-1}$) respectively. It has been observed spreadability decreases with the increase in FR level. That fat content of cheese is decreasing the spreadability of cheese also decreasing. Spreadability of cheese differed significantly ($p<0.05$) among the three levels of FR and control. Whereas firmness increased with increased levels of FR addition in cheese. 10% fat replacer sample showed significantly different ($p<0.05$) in spreadability and firmness.

5.3.1.1.2 Changes in sensory attributes

The average color & appearances scores of control, 10,15 & 20% FR added cheeses were 7.82,7.85,7.31and 7.08 respectively (Table 5.4). Cheese with 10% FR scored very similar to that of control cheese and better than 15 and 20% FR added cheese. Similar was the case for body & texture scores. The average B &T scores of 10% added cheese significantly ($p<0.05$) higher than that of 15 and 20% FR added cheese but the difference was not significant between control and 10% FR added cheese. This higher score in 10% FR added cheese may be due to the higher spreadability and lower the firmness of sample. The average flavor score of samples revealed that highest flavor score was in control sample followed by 10, 15 & 20% FR added cheese. This may be due to reduction in fat content of the cheese as fat contributes flavor profusely in the cheese. Acceptability of cheese is the summation of color & appearances, body & texture and Flavor scores. Since 10% FR added cheese sample had better color & appearance and body & texture scores it obtained in maximum score than other samples. Therefore, 10% FR addition in the cheese was selected among three levels of far replacement.

Table 5.4. Effect of 40% fat reduction with addition of 10, 15 and 20% fat replacers (w/w) on the physico-chemical, textural & sensory characteristics of Mascarpone cheese

PARAMETER	Control	10 %	15%	20%
Moisture (%)	54.02±1.16[a]	58.13±1.66[b]	54.66±1.27[a]	53.21±1.24[a]
pH	5.23±0.01[a]	5.30±0.03[ab]	5.33±0.06[ab]	5.43±0.12[b]
Color (0-255)	195.5±1.10[a]	208.6±1.41[d]	203.03±1.44[c]	199.3±0.70[b]
Spreadability($N.S^{-1}$)	2.03±0.15[d]	1.64±0.036[c]	1.38±0.05[b]	1.17±0.05[a]
Firmness(N)	78.08±2.35[a]	107.11±1.86[b]	121.70±3.39[c]	174.68±4.18[d]
SENSORY PARAMETER				
C& A	7.82±0.40[b]	7.85±0.29[b]	7.31±0.63[a]	7.08±0.63[a]
Body & Texture	7.68±0.30[b]	7.81±0.34[b]	7.29±0.5[7a]	7.02±0.55[a]
Flavor	7.74 ±0.46[b]	7.24 ±1.97[ab]	7.12 ±0.75[ab]	6.90 ±0.65[a]
Overall Actability	7.78 ±0.51[b]	7.86 ±0.27[b]	7.32± 0.61[a]	7.07±0.56[a]

Means with Different superscript in a row differ significantly (p<0.05)

Table 5.4a Anova for physico-chemical and textural parameter

		Moisture		**pH**		**Spreadability**		**Firmness**		**Color**	
Source of variation	df	MS	F	MS	F	MS	F	MS	F	MS	F
Between the FR levels	3	14.09	7.69*	0.020	3.870	0.417	51.522*	4915.0	515.7*	92.58	64.07*
Error	8	1.832		0.005		0.008		9.530		1.447	
Total	11										

**Significantly differ at levels of 5%*

Table 5.4b Anova for sensory parameter

		C & A		**B & T**		**Flavor**		**OA**	
Source of variation	df	MS	F	MS	F	MS	F	MS	F
Between the FR levels	3	2.178	8.197*	1.958	9.175*	1.924	1.506	2.111	8.122*
Error	56	0.266		0.213		1.274		0.260	
Total	59								

**Significantly differ at levels of 5%*

Based on physical and sensory characteristics, 10% FR addition in 40% fat reduced Mascarpone cheese was adjudged best among the three level of FR addition.

5.3.1.2 Effect of 50% fat reduction with addition of various levels of fat replacer (w/w) on the physico-chemical, textural & sensory characteristics of Mascarpone cheese

The second fat reduction of 50% fat in the cheese was studied. To study 50% fat reduction in the cheese, milk was standardized to 3.5- 4.5% fat with 50% fat cream and was coagulated with 4% lactic acid solution at 80-82^{0}C. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey drained out, remaining 50% fat in the from of pasteurized cream was added in the coagulated mass and mixed well to obtain 20% fat in the final product. It was kept overnight in cold condition for further removal of whey. Coagulated mass was taken out from the cloth and weighed. Three mixtures of fat replacers were heat processed and cooled. Fat replacers of 10% (6% inulin, 2% WPC & 2% N-creamer), 15% (6% inulin, 4.5% WPC & 4.5% N-creamer) and 20% (6% inulin, 7% WPC & 7% N-creamer) were added in Mascarpone cheese of 20% fat and mixed homogeneously. Moisture content, pH, color, spreadability, firmness and sensory characteristics of products obtained were compared. A control Mascarpone cheese was manufactured using 50% fat in milk and 50% fat incorporation in coagulum to obtain 40% fat in final Mascarpone cheese without addition of fat replacers.

<u>5.3.1.2.1 Effect on physico-chemical and textural parameters</u>

Mascarpone cheese with 50% fat reduction was manufactured and 10, 15 and 20% FR was added on weight basis. It was evident from the Table 5.5 that with the increase of fat reduction level, moisture content in cheese increased. Again, with increase in FR levels, the moisture content in cheese decreased. It was observed that the moisture content in cheese with 10% FR was significantly ($p<0.05$) maximum (59.07%) of amongst other levels except 15%. The pH was found to increase with the increase of FR levels in cheese. The control sample had pH of 5.19 whereas samples with 10, 15 and 20% FR showed pH

Table 5.5. Effect of 50% fat reduction with addition fat replacer (10, 15 and 20 FR on weight basis) on the physico-chemical, textural & sensory characteristics of Mascarpone cheese

PARAMETER	Control	10%	15%	20%
Moisture (%)	53.90±0.95[a]	59.07±1.66[b]	56.30±3.01[ab]	54.36±1.26[a]
pH	5.19±0.00[a]	5.32±0.03[b]	5.34±0.07[b]	5.45±0.06[c]
Color (0-255)	198.7±1.19[a]	209.7±0.36[d]	205.8±0.35[c]	202.3±1.24[b]
Spreadability($N.S^{-1}$)	2.10±0.05[d]	1.74±0.04[c]	1.48±0.03[b]	1.29±0.02[a]
Firmness(N)	77.82±2.66[a]	98.19±2.26[b]	114.07±1.53[c]	138.70±2.59[d]
SENSORY PARAMETER				
Color & Appearance	7.35±0.56[b]	7.61±0.62[b]	7.42±0.45[b]	6.57±0.83[a]
Body & Texture	7.50±0.30[b]	7.65±0.52[b]	7.24±0.49[b]	6.36±0.84[b]
Flavor	7.41±0.41[b]	7.55±0.58[b]	7.17±0.41[b]	6.36±0.71[a]
Overall Actability	7.45±0.31[b]	7.62±0.50[b]	7.25±0.45[b]	6.36±0.81[a]

Means with Different superscript in a row differ significantly (p<0.05)

Table 5.5a Anova for physico-chemical and textural parameter

		Moisture		pH		Spreadability		Firmness		Color	
Source variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Levels of FR	3	16.61	4.63	0.034	12.69*	0.370	210.2*	1983.1	372.1*	66.03	81.61
Error	8	3.58		0.03		0.002		5.32		0.809	
Total	11										

**Significantly differ at levels of 5%*

Table 5.5b Anova for sensory parameter

		C & A		B & T		Flavor		OA	
Source of variation	df	MS	F	MS	F	MS	F	MS	F
Between the FR levels	3	3.158	7.792*	4.970	14.920*	4.271	14.152*	4.710	15.460*
Error	56	0.405		0.333		0.302		0.305	
Total	59								

**Significantly differ at levels of 5%*

of 5.32, 5.34 and 5.45 respectively. The pH values differed significantly (p<0.05) among four types of cheese.

The low fat cheese normally appears to be whiter than full fat cheese. This is mainly because of less fat content and more moisture content in cheese. It was observed that cheese with 10% FR attained maximum whiteness (209.7) followed 15% FR, control and 20% FR added cheese. The color value differed significantly among the samples. Lower value in both 15 and 20% FR added cheese was due to the presence higher quantity of WPC affecting adversely on the whitening of cheese as shown in Table 5.5. The spreadability of control and 10, 15 & 20% FR added cheese differed significantly. The spreadability of control was significantly highest (2.10 $N.s^{-1}$) followed by 10, 15 and 20% FR added cheese. This indicated that spread ability decreases with decrease of fat content in cheese. Spreadability of the product is also influenced by two factors like FR addition and moisture content. Firmness of the cheese differed significantly ($p<0.05$) among the samples. Firmness increased with increasing level of fat replacers. Cheese with 10% FR showing good spreadability with moderate firmness seems to be the best among three levels of fat replacers.

<u>5.3.1.2.2 Changes in sensory attributes</u>

The effect of 50% fat reduction with different fat replacer levels on color and appearances score of cheese shown in Table 5.5. The average color & appearances scores of control, 10%, 15% & 20% fat replacer addition were 7.35, 7.61, 7.42 and 6.57 respectively. The 10% FR addition sample had a better score compared to other sample. The average body & texture score of all sample shown in Table 5.3.2 The average B &T was found higher sensory score in 10% FR addition sample but not significantly different ($p>0.05$) in control and other samples. The average flavor score of sample shown in Table 5.3.2 Higher flavor score was found in control and 10% FR sample is 7.41 and 7.55 respectively , due to the fat content because fat are contributing the flavor of cheese. The average over all acceptability score of cheese shown in the Table 5.5. Acceptability of cheese is the summation of all color & appearances, body & texture and flavor. Since 10% FR addition sample had better color & appearance, flavor and body & texture score which resulted in higher score than other samples. Higher sensory score in the cheese was due to the good spreadability and high firmness.

In 50% fat reduced Mascarpone cheese, addition of 10% FR has produced best quality cheese among the three levels of FR addition.

5.3.1.3 Effect of 60% fat reduction with addition 10, 15 and 20 % fat replacer (w/w) on the physico-chemical, textural & sensory characteristics of Mascarpone cheese

The third fat reduction of 60% fat in the cheese was studied. To study 60% fat reduction in cheese, milk was standardized to 3-4% fat with 50% fat cream and was coagulated with 4% lactic acid solution at 80-82°C. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the from of pasteurized cream was added in the coagulated mass and mixed well to obtain 16% fat in the final product. It was kept in cold condition for overnight for further removal of whey. Coagulated mass was taken out from the cloth and weighed. Three mixtures of fat replacers were heat processed and cooled. Fat replacers of 10% (6% inulin, 2% WPC & 2% N-creamer), 15% (6% inulin, 4.5% WPC & 4.5%N-creamer) and 20% (6% inulin, 7% WPC & 7% N-creamer) were added in Mascarpone cheese of 16 % fat and mixed them homogeneously. Moisture content, pH, color, spreadability, firmness and sensory characteristics of the products obtained were compared. A control Mascarpone cheese was manufactured using 50% fat in milk and 50% fat incorporation in coagulum to obtain 40% fat in final Mascarpone cheese without addition of fat replacers.

5.3.1.3.1 Effect on physico-chemical and textural parameters

The influence of fat replacers in combination of the Inulin, WPC and N-creamer was studied in 3 combinations at three different levels of addition in 60% fat reduced Mascarpone cheese. The results are presented in Table 5.6. It was observed that with the increase of fat reduction level, moisture content in cheese increased. Again, with increase in FR levels, the moisture content in cheese decreased. It was observed that the moisture content in cheese with 10% FR was significantly maximum (60.44%) of amongest other levels except 15%. The pH has increased with the increase of FR levels in cheese. The control sample had pH of 5.24 whereas samples with 10, 15 and 20% FR showed pH of

5.41, 5.41 and 5.62 respectively. The pH values differed significantly except between 10 and 15% FR added cheeses. The whiteness of cheese increased significantly ($p<0.5$) with the increasing levels of FR addition than control. Whiteness improved with higher fat reduction in cheese due to the low fat content and higher moisture content. Highest color of 214 was observed in the cheese with 10% FR followed by15 and 20% FR added cheese. The color differed significantly except between 15 and 20% FR level. The spreadability of control and 10, 15 & 20% FR added cheeses was 2.11, 1.91, 1.64 and 1.40 $N.s^{-1}$ respectively. It has been observed that spreadability decreased with the increase in FR level or fat content. Spreadability of cheese differed significantly among the three levels of FR and control. Other hand firmness increased with the increased levels of FR addition in cheese. There was no significant reduction in the firmness between control and 10% FR added cheese whereas the different was significant among three levels of FR content. .

5.3.1.3.2 Changes in sensory parameters

The sensory scores of 60% fat reduced cheese with addition of different levels of fat replacer as presented in Table 5.6. In general the sensory scores of 10% FR added and control cheeses were closer to each other. The average color & appearances scores of control, 10%, 15% & 20% fat replacer addition were 7.46, 7.40, 7.29 and 6.82, respectively. The 10% FR added sample showed a better score compared to other samples. The average B &T score did not differ significantly ($p>0.05$) except with 20% FR added cheese. Higher flavor score both in control and 10% FR added was same of 7.56 followed by 7.28 and 6.73 in 15 and 20%FR added cheeses, respectively. High scores in both control of 10% FR added cheese compared to 15 & 20 % FR was due to the proportionally higher fat content. Since, over all acceptability scores of cheese is the summation of all color & appearances, body & texture and flavor scores, it was observed that 10% FR added cheese scored close to control and differences between them was not significant.

Addition of 10%FR in 60% fat reduced cheese also resulted in best quality of chhese among 3 levels of FR addition. Hence, it can be concluded that the among three levels of fat replacers, 10% FR addition was found to be best and similar to that of control sample in all aspects.

Table 5.6. Effect of 60% fat reduction with addition 10, 15 and 20 % fat replacer (w/w) on the physico-chemical, textural & sensory characteristics of Mascarpone cheese

PARAMETER	Control	10%	15%	20%
Moisture (%)	53.17±3.20[a]	60.44±0.63[b]	60.28±0.77[b]	58.19±0.49[b]
pH	5.24±0.05[a]	5.41±0.10[b]	5.416±0.07[b]	5.626±0.07[c]
Color (0-255)	197.96±2.30[a]	214.63±1.36[c]	209.23±1.10[b]	206.53±1.35[b]
Spreadability ($N.s^{-1}$)	2.11±0.14[d]	1.91±0.07[c]	1.64±0.06[b]	1.40±0.02[d]
Firmness(N)	70.02±5.01[a]	83.69±3.21[a]	103.5± 4.01[b]	127.95±2.52[c]
SENSORY PARAMETER				
Color & Appearance	7.46± 0.57[b]	7.40±0.57[b]	7.29±0.53[ab]	6.82±0.97[a]
Body & Texture	7.57±0.66[b]	7.50±0.57[b]	7.35±0.51[b]	6.82±0.89[a]
Flavor	7.56±0.50[b]	7.56±0.59[b]	7.28±0.51[b]	6.73±0.93[a]
Overall Actability	7.60±0.64[b]	7.58±0.60[b]	7.23±0.69[ab]	6.79±0.93[a]

Means with Different superscript in a row differ significantly (p<0.05)

Table 5.6a Anova for physico-chemical and textural parameter

		Moisture		pH		Spreadability		Firmness		Color	
Source variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Levels of FR	3	34.5	11.9*	0.07	11.6*	0.28	35.1*	1531.1	105.8*	145.0	58.8*
Error	8	2.88		0.006		0.00		14.46		2.55	
Total	11										

**Significantly differ at levels of 5%*

Table 5.6b Anova for sensory parameter

		C & A		B & T		Flavor		OA	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Between the FR levels	3	1.264	2.954	1.738	3.784	2.296	5.275*	2.166	4.047
Error	56	0.428		0.459		0.435		0.535	
Total	59								

**Significantly differ at levels of 5%*

5.3.1.4 Effect of 10 % fat replacer (w/w) on the physico-chemical, textural & sensory characteristics of 40, 50 and 60% fat reduced Mascarpone cheeses.

Among the three fat replacers of 10% (6% inulin, 2% WPC & 2% N-creamer), 15% (6% inulin, 4.5% WPC & 4.5%N-creamer) and 20% (6% inulin, 7% WPC & 7% N-creamer) in 40, 50 and 60% fat reduced cheese, 10 % fat replacer (6% inulin, 2% WPC & 2% N-creamer) was selected based on physical and sensory characteristics. For further study, effect of 10% fat replacer (6% inulin, 2% WPC & 2% N-creamer) on the physico-chemical, textural & sensory characteristics of 40, 50 and 60% fat reduced Mascarpone cheeses were studied.

Therefore, Mascarpone cheeses were made from the milk standardized to 3.0 -6.0% fat in milk with 50% fat cream and was coagulated with 4% lactic acid solution at 80-82^{0}C. After coagulation, the coagulated mass was cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the from of pasteurized cream was added in the coagulated mass and mixed well to obtain 24, 20 and 16% fat in the final product. It was kept in cold condition overnight for further removal of whey. Coagulated mass was taken out from the cloth and weighed. In the three types of reduced fat cheeses of 24, 20 and 16% fat, 10% fat replacer (6% inulin, 2% WPC & 2% N-creamer) were added and mixed them homogeneously. Moisture content, pH, color, spreadability, firmness and sensory characteristics of products obtained were compared. A control Mascarpone cheese was manufactured using 50% fat in milk and 50% fat incorporation in coagulum to obtain 40% fat in final Mascarpone cheese without addition of fat replacers.

<u>5.3.1.4.1 Effect on physico-chemical and textural Parameters</u>

It was observed that as level of fat reduction increased with 10% FR, moisture content increased (Table 5.7). Significantly ($p<0.05$) higher moisture content (60.67%) was observed in 60% fat reduced cheese. As explained earlier, with the increased level of fat reduction, pH increased. Similarly, colour/ whitness increased with the increased level of fat reduction in cheese. Colour value was significantly higher in 60% fat reduced cheese than control samples. Spreadability decreased, as expected, with the increased level of fat reduction in cheese. However, as the fat content reduced, moisture retention in cheese increased which had influenced spreadiability of cheese. Spreadability has inverse relation on firmness, hence firmness decreased with the increased level of fat reduction. Firmness reduction may due to increased level of moisture retention in cheeses with low fat.

5.3.1.4.2 Effect on sensory characteristics

The average sensory scores with standard error for color & appearance, body & texture, flavor and overall acceptability are presented in Table 5.7. The color appearance scores of cheese were 7.38, 6.76, 6.98 and 6.76 for control, 40, 50, 60% frat reduced cheeses, respectively. Though the color & appearance score did not show any significant ($P>0.05$) differences among 40, 50 & 60% fat reduced samples, however it was significant with control. The average body & texture scores of cheese were 7.52, 6.74, 7.11 and 7.74 for control, 40, 50, 60% fat reduced cheeses, respectively. Though the body & texture score showed significant ($P>0.05$) differences between 60 and 40, 50% fat reduced samples but difference was not significant between control and 60% fat reduced cheese. Good body and texture score may link with higher spreadability and higher moisture content in 60% fat reduced cheese. As observed, the average overall acceptability scores of control and 60% fat reduced cheese were very similar and did not differ significantly. Overall acceptability score showed significant ($P<0.05$) differences between the 60 and 40, 50% fat reduced samples. However, maximum score of 7.74 was observed in 60% fat reduced cheese..

So based on physico-chemical parameters, sensory characteristics and their statistically analysis, it was adjudged that 60% fat reduced Mascarpone cheese with addition of 10% fat replacer in three combinations resulted in a most acceptable low fat Mascarpone cheese.

Table 5.7 Effect of 10 % fat replacer (w/w) on the physico-chemical, textural & sensory characteristics of 40, 50 and 60% fat reduced Mascarpone cheeses

PARAMETER	Control	40%(10%FR)	50%(10%FR)	60%(10%FR)
Moisture (%)	54.58±1.89[a]	56.76±1.81[ab]	58.90±0.51[bc]	60.67±0.83[c]
pH	5.21±0.03[a]	5.30±0.04[b]	5.34±0.011[b]	5.53±0.05[c]
Color (0-255)	197.76±[a]	208.8±2.15[b]	210.5±1.09[b]	214.93±1.76[c]
Spreadability(N.s^{-1})	2.07±0.06[c]	1.64±0.07[a]	1.72±0.030[ab]	1.83±0.06[b]
Firmness (N)	79.50±4.51[a]	107.90±2.38[b]	121.17±4.97[c]	100.13±4.44[b]
SENSORY PARAMETER				
Color & Appearance	7.38 ±0.60[b]	6.76 ±0.45[a]	6.98± 0.48[a]	6.76 ±0.54[a]
Body & Texture	7.52±0.51[b]	6.74±0.58[a]	7.11±0.26[a]	7.74±0.51[b]
Flavor	7.58±0.48[c]	6.62±0.65[a]	7.02±0.49[b]	7.71±0.46[a]
Overall Actability	7.44±0.51[bc]	6.75±0.59[a]	7.11±0.43[ab]	7.74±0.49[c]

Means with Different superscript in a row differ significantly (p<0.05)

Table 5.7a Anova for physico-chemical and textural parameter

		Moisture		pH		Spreadability		Firmness		Color	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Levels of FR	3	20.89	10.67*	0.054	34.22*	0.104	28.72*	911.72	51.69*	160.12	58.79*
Error	8	1.95		0.002		0.004		17.63		2.723	
Total	11										

**Significantly differ at levels of 5%*

Table 5.7b Anova for sensory parameter

		C & A		B & T		Flavor		OA	
Source of variation	**df**	**MS**	**F**	**MS**	**F**	**MS**	**F**	**MS**	**F**
Levels of FR	3	2.368	9.884*	2.954	11.535*	3.830	13.54*	2.599	9.884*
Error	56	0.263		0.256		0.283		0.263	
Total	59								

**Significantly differ at levels of 5%*

5.4 PREPARATION OF STANDARDIZED LOW FAT FIBRE ENRICHED MASCARPONE CHEESE

Mascarpone cheese was manufactured from fresh cow milk. Mascarpone cheese was made from milk standardized to 3-4% fat with 50% fat cream and was coagulated with 4% lactic acid solution at 80-82^{0}C. After coagulation, the coagulated mass cooled at room temperature by natural cooling (30 min). Whey was drained out from the coagulated mass using muslin cloth. After maximum amount of whey draining, remaining 50% fat in the from of pasteurized cream was added in the coagulated mass and mixed well to obtain 16% fat in the final product. It was kept in cold condition overnight for further removal of whey. Coagulated mass was taken out from the cloth and weighed. Heat processed and cooled fat replacer of 10% (6% inulin, 2% WPC & 2% N-creamer) was added in Mascarpone cheese of 16% fat and mixed them homogeneously. Low fat Mascarpone cheese was packed in polypropylene cup and stored at 5-8°C. The outlines for the manufacture of low fat fibre enriched Mascarpone cheese is presented in Figure 5.1.

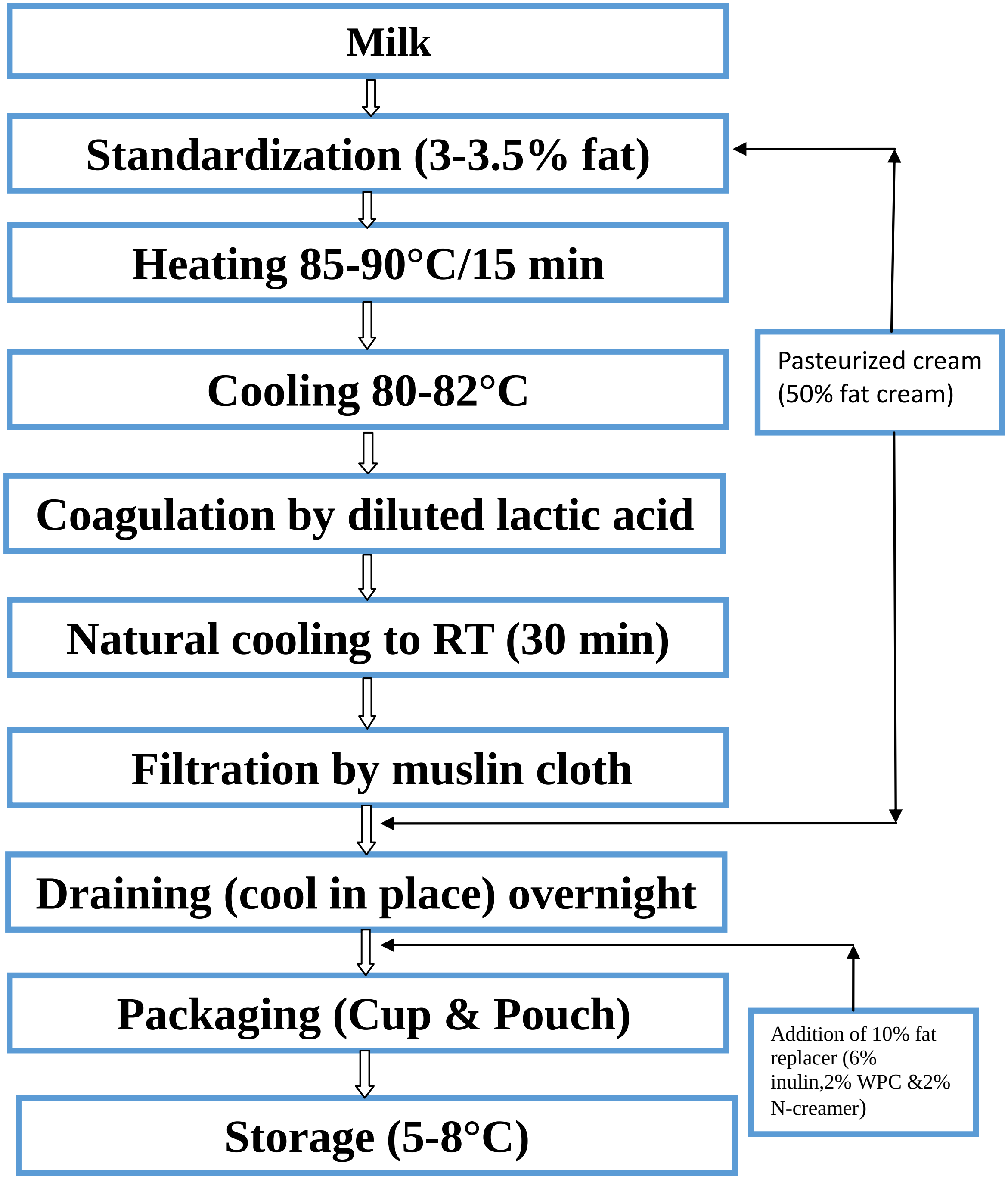

Figure 5.1: Flow diagram for the manufacture of low fat fibre enriched Mascarpone Cheese.

5.5 EVALUATION OF QUALITY CHARACTERISTICS OF THE DEVELOPED LOW FAT FIBRE ENRICHED MASCARPONE CHEESE

Low fat fibre enriched Mascarpone cheese was made as per method outlined in section 5.4. A control Mascarpone cheese was also manufactured using 50% fat in milk and 50% fat incorporation in coagulum to obtain 40% fat in final Mascarpone cheese without additives. Both the cheeses were packed and stored at 5-8°C and compared for physico chemical and sensory characteristics.

Table 5.8 Physico-chemical and textural properties of control and experimental low fat Mascarpone cheese

PHYSIC-CHEMICAL PARAMETER	CONTROL	EXPERIMENT SAMPLE
Total solids (%)	49.62±0.41	38.02±0.13
Protein (%)	7.5±0.07	8.42±0.08
Fat (%)	38.6±0.26	16.42±0.08
Ash (%)	0.76±0.05	1.53±0.22
Total Carbohydrate (%)	2.72±0.41	11.46±0.13
pH	5.21±0.01	5.48±0.01
Acidity (%LA)	0.21±0.01	0.18±0.008
Spreadability ($N.s^{-1}$)	2.03±0.03	1.84±0.03
Firmness (N)	85.7±3.46	103.1.87
Water activity	0.95±0.001	0.96±0.001

5.9 Sensory characteristics of control and experimental low fat Mascarpone cheese

Parameter	Control	Experimental
Color & Appearance	7.78±0.20	8.03±0.24
Body & Texture	7.65±0.21	7.98±0.11
Flavor	7.97±0.12	7.72±0.20
Overall acceptability	7.62±0.17	8.09±0.25

Average value of 5 trials

5.5.1 Physico-chemical and textural properties of control and experimental Mascarpone cheeses

The means values of physico-chemical characteristics of control and experimental Mascarpone cheese samples are presented in Table 5.8. TS content of experimental sample was lower than control because of the reduction of fat where moisture might have compensated fat. Protein content was slightly higher in experimental sample due to the incorporation of WPC as fat replacer. Fat content in experimental was lower than control sample. Higher ash content in experimental cheese may due to the incorporation protein and carbohydrate based fat replacer. Higher carbohydrate content in the experimental sample was, as expected, due to addition of carbohydrate based fat replacer. pH of experimental sample was more due to the incorporation inulin and other fat replacers. Marginally low acidity in experimental sample may due to the incorporation three fat replacers. The mean spreadability of control was slightly more may be due to higher content as fat has influence on the spreadability (2.03 $N.s^{-1}$). Since the firmness has inverse relation with spreadability, firmness value of experimental sample was slightly lower and close to the control sample. Water activity of experimental sample was slightly higher than control sample due to higher moister content.

5.5.2 Sensory characteristics of control and experimental Mascarpone cheeses

The sensory characteristics of control and optimized low fat fibre enriched Mascarpone cheese samples are presented in Table 5.9.

Average color and appearance scores of control and experimental samples were 7.78 and 8.03 respectively. Experimental cheese had better score because of improvement in white color due to low fat and high moisture. Carbohydrate based fat replacers might have functioned like fat by binding water, providing texture, mouth feel and opacity (Giese, 1996). Johnson (2000) and Harrigan (1989) have reported diverse functional properties of fat mimetic towards physico chemical attributes, desirable eating qualities of fat, viscosity, mouth feel and appearance.

Table 5.9 depicts better body & texture score of experimental sample than that of control because of presence of fat replacers and more moisture content with good spreadability as it was observed in physico chemical analysis. Reduction of the fat content in cheese also alters the main parameters of texture and rheology.

The average flavor score of cheese was 7.97 and 7.72 for control and experimental sample, respectively (Table 5.9). Control sample had better score because of higher fat in it which is responsible for flavor. Intense flavor in control Mascarpone cheese was observed because of more acidity and or low pH of the product. Low flavor intensity in experimental cheese is due to the utilization of fat mimetic for the production of reduced fat Mascarpone cheese. Similar observation has been reported in low fat Cheddar Cheese because of fat mimetic may have a negative effect on flavor (Drake *et al.*, 1996).

Acceptability of cheese is the summation of all color & appearances, body & texture and flavor. Experimental sample had better score due to better score for color and appearance as well as body and texture. Higher score for experimental sample was also due to good taste, mouthfeel and improvement in color of cheese. Better score may be also due to the differences in pH between the experimental sample and control.

5.6 SHELF LIFE STUDIES OF DEVELOPED LOW FAT, FIBRE ENRICHED MASCARPONE CHEESE

Control and developed low fat Mascarpone cheese samples were prepared as per the optimized method (section 5.3 & 5.4) and sample was packed 100 g in polyethylene pouches and polystyrene cups. All the samples were stored at refrigeration temperature (5-8°C). The samples were drawn at an interval of four days. Samples were first taken for microbial evaluation followed by sensory quality. The changes in pH, acidity and TBA of the sample were also measured. Mascarpone cheese samples were coded as: **CC-control in cup, CP- Control in pouch, SC- Sample in cup, SP- Sample in pouch.**

5.6.1 CHANGES IN VARIOUS PARAMETERS AT STORAGE TIME

5.6.1.1 Changes in sensory attributes

5.6.1.1a Color and appearances

The changes in score of color & appearance of stored Mascarpone cheeses are shown in fig. 5.2. It is evident from the figure that on zero day score of SC (8.1) and SP (8.1) was apparently more than CC (8) and CP (8). The sensory score of the Mascarpone Cheese decreased over the period of the storage. CC and CP were accepTable up to 7-8 days when stored under refrigeration. Thereafter an intense sour flavor developed and slight whey separation was found. This may be due to the microbial spoilage. But in case of SC and SP spoilage occured after 10 days. In all samples, whiteness of sample was decreased during storage. But in CC and CP sample, the decrease in whiteness was more compared to SC and SP samples. This may be due to discoloration caused by the microbial growth in the stored Mascarpone cheese (Cantoni *et al.*, 1997).

5.6.1.1b Body & Texture

Figure 5.3 shows the changes in body and texture score of CC, CP, SC and SP samples during storage under refrigeration. It was observed that the body and texture scores were decreased slightly with increase in storage time. With increase in the storage period, some whey separation was observed and also stickiness was found in the product. The score of CC and CP on 8th day were 6 and 6.5 respectively whereas in SC and SP the corresponding score were 6.6 and 7 on 12th day. It means that both the experimental samples showed better score and probably longer shelf life.

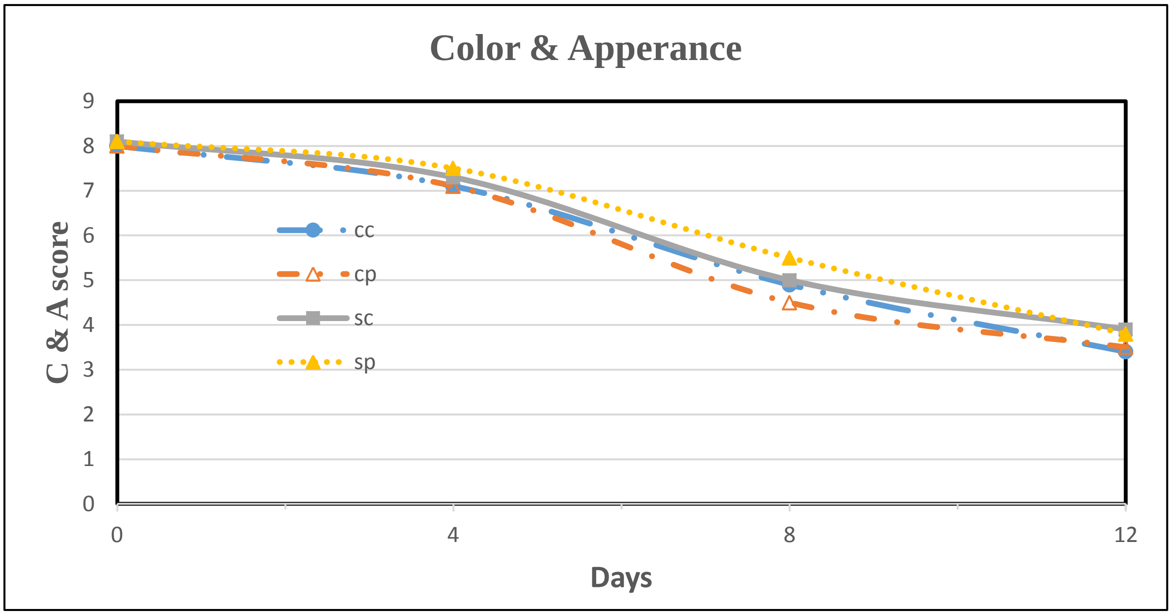

Fig. 5.2 Changes in color & appearance of Mascarpone cheese stored at 5-8^{0}C

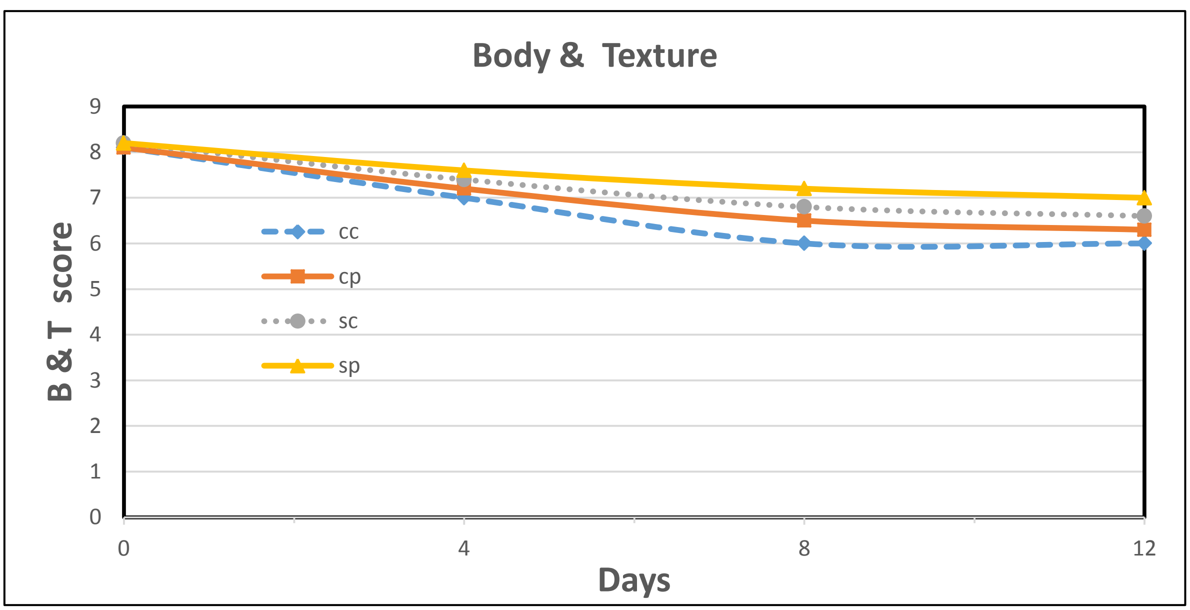

Fig. 5.3 Changes in body & texture of Mascarpone cheese stored at 5-8^{0}C

5.6.1.1c Flavor

The flavor score drastically reduced during storage period. The initial score were 8.3 for cup and 8.3 for CP which became 4.0 and 4.3 respectively after 8 days of storage under refrigeration as shown in figure 5.4. Similarly, for SC and SP the initial score of 8.5 and 8.5 became 4.5 and 4.9 respectively after 8 days storage. The fresh flavor of Mascarpone cheese was changed to acidic and sour flavor after 7-8 days in case of CC and CP whereas in case of SC SP after 8-9 days. Samples of CC and CP had higher fat, therefore fat oxidation might have taken place whereas SC and SP had low Fat content and thus low fat oxidation. The CC and CP samples had higher acidic taste compared to SC and SP samples because SC and SP sample contains lesser fat content and the fat replacers might have resisted microbial growth causing increase acidic taste.

5.6.1.1d Overall acceptability

The changes in overall acceptability score of cheese during storage is presented in Figure 5.5. The scores of fresh samples were almost same but it decreased as storage period progressed. The CC and CP samples had least acceptability score on 8th and 12th days as compared to the SC and SP samples. Since the flavor was varying at a faster rate, the overall acceptability of CC and CP showed a faster rate of decline compared to SC and SP. The scores of fresh samples were 8.16, 8.16, 8.27 & 8.27 which became 3.84, 3.67, 3.9 and 4.27 for CC, CP, SC and SP respectively at the end of 12th days. After 12 days storage the cheese becomes acidic and sour. It was found that the freshness was completely lost and the cheeses become unacceptable.

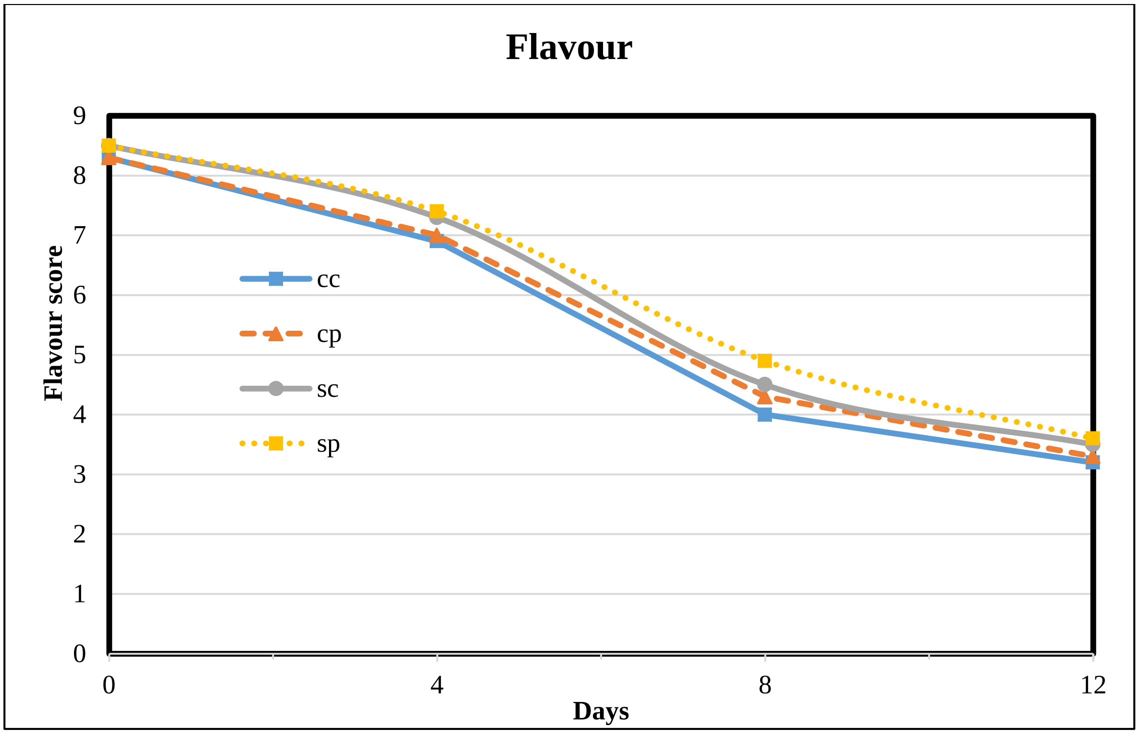

Fig. 5.4 Changes in flavor of Mascarpone cheese stored at 5-8^{0}C

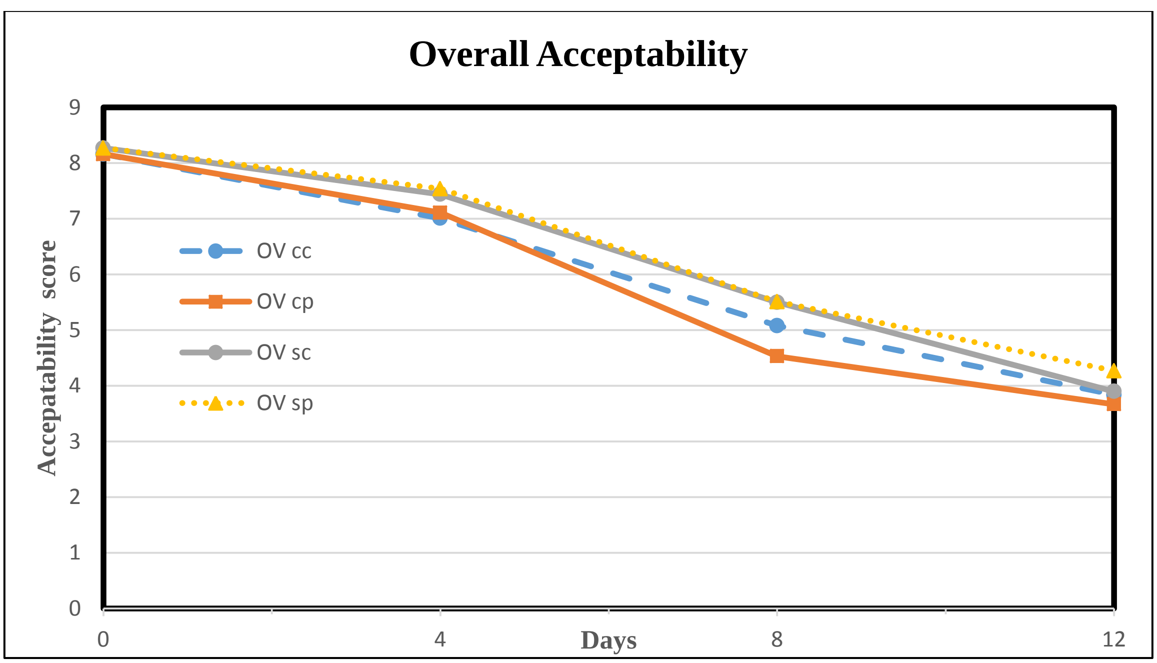

Fig. 5.5 Changes in overall acceptability of Mascarpone cheese stored at 5-8^{0}C

5.6.1.2 Changes in Chemical parameters

5.6.1.2a Changes in pH

Figure 5.6 depicts the changes occurred in pH during storage of Mascarpone cheese stored at 7±1°C. The pH of Mascarpone cheese decreased with the increased storage time due to activity of microorganism. The initial pH of CC and CP were 5.2 and 5.2 and it became 4.95 and 4.99 respectively, whereas the initial pH of SC and SP were 5.5 and 5.49 became 5.24 and 5.28 respectively, while after 12 day storage. The rate of decrease in pH of the cheese was observed to be lower in the pouch packed samples as compared to cup samples irrespective of samples. Experimental samples showed higher pH than control samples because of added fat replacers.

5.6.1.2b Changes in acidity

Figure 5.7 shows the changes in mean values of acidity in 4 samples of Mascarpone cheese stored under refrigeration. The acidity of both control samples were 0.14 % LA on zero day whereas on the 12^{th} day it was 0.30% in cup and 0.29% in pouch filled samples. Acidity of both fresh experimental samples was 0.12% LA and resulted in 0.25% in cup and 0.23 % in pouch packed samples on 12^{th} days of storage at refrigerator. The result showed that during storage, rate of acidity development in CC and CP cheese higher as compared to the acidity in SC and SP samples. Low acidity in both the experimental samples may be due addition of inulin and other fat replacers. Between two packaging materials, pouch packed samples showed lesser incremental increase compared to the cup packed samples during storage.

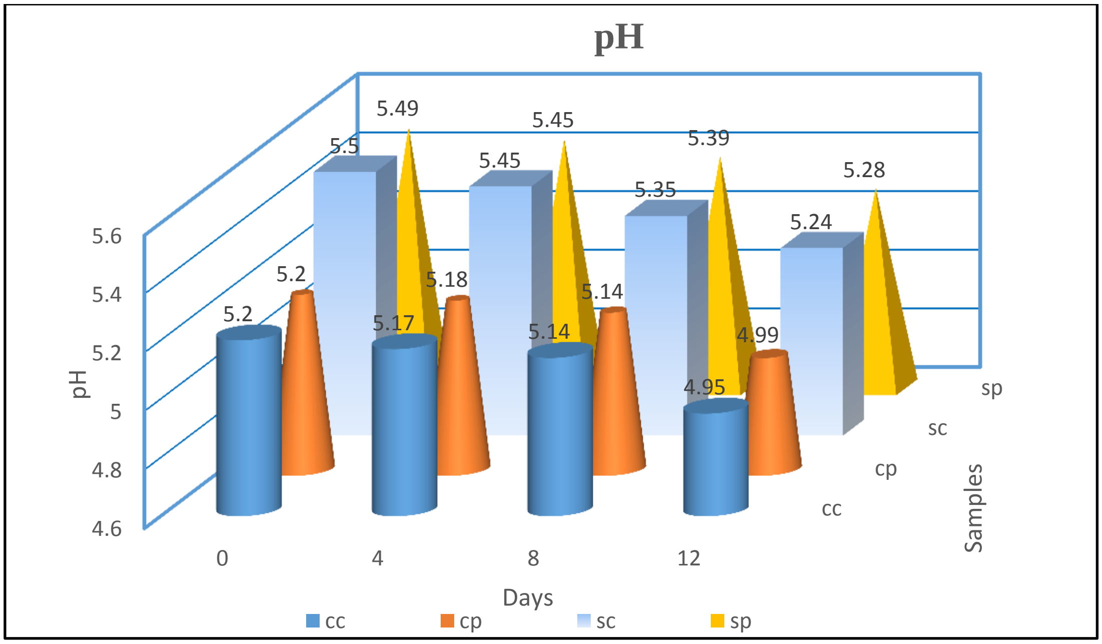

Fig 5.6 Changes in pH of Mascarpone cheese stored at 5-8^{0}C

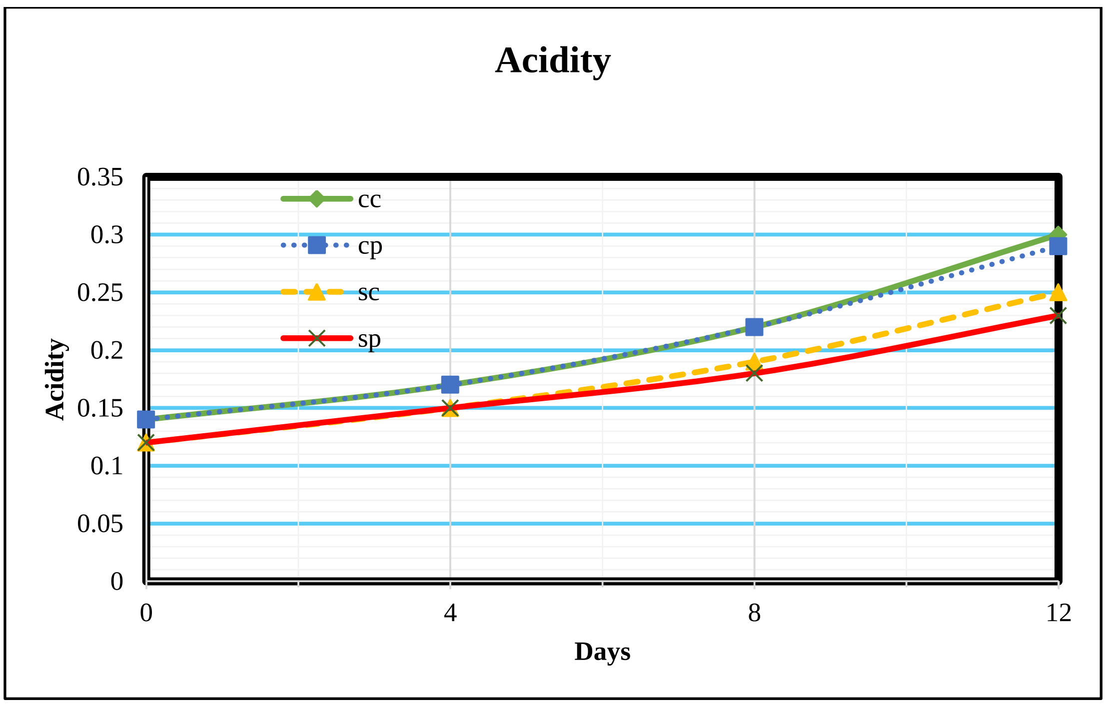

Fig 5.7 Changes in acidity of Mascarpone cheese stored at 5-8^{0}C

5.6.1.2c Changes in Thiobarbutric Acid (TBA)

TBA value is indication of the level of oxidation in product. It means that higher TBA value means higher will be the oxidation in product. Higher is the fat in product, more chances of fat oxidation. TBA value of product increased with increase in the number of storage days as evidence from figure 5.8. The average TBA value of the cheese in CC and CP were 0.065 on zero days but however in SC and SP samples it was 0.060 for both samples at same day whereas at 12th day CC and CP average value was 0.112 and 0.103 respectively and SC and SP has 0.084 and 0.077 respectively. This result indicated that higher rate of oxidation occurred in CC and CP due to the higher amount of fat as compare to the SC and SP. Pouch packed both control and experimental samples showed lower oxidation than the samples stored in cup for both cases.

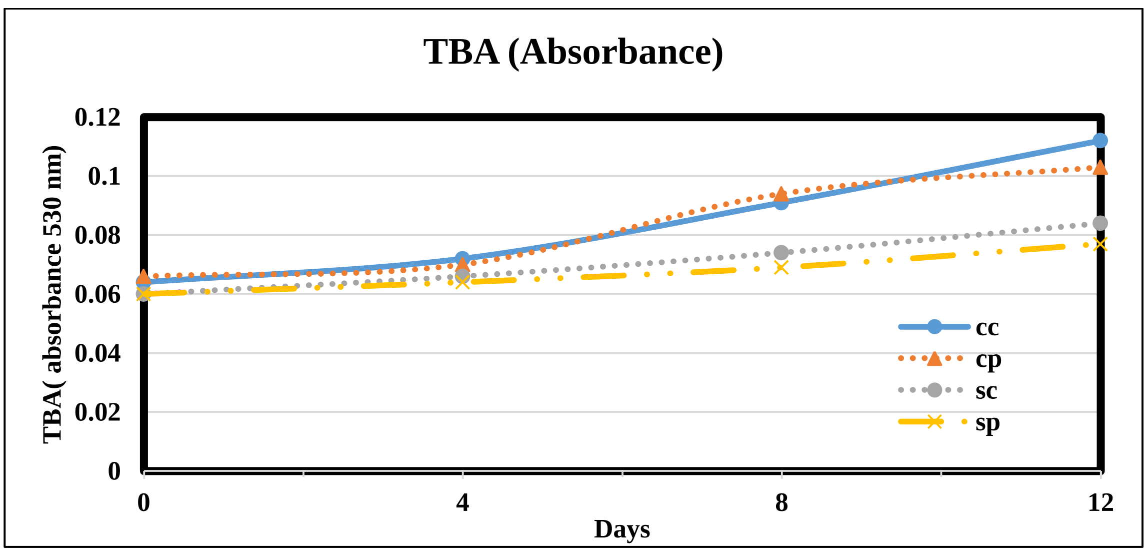

Fig 5.8 Changes in TBA of Mascarpone cheese stored at 5-8^{0}C

5.6.2. ACCEPTABILITY PREDICTION MODEL BASED ON SENSORY SCORE

Based on sensory scores, product acceptability which in turn of shelf life prediction of the product was calculated by fitting a regression equation where sensory score was dependent variable and both pH and TBA values were independent variables. With this prediction equation, one can evaluate the status of acceptability of the product if pH and TBA values are known during storage. The limitation of the equation is the pH range which should be between 5.2 to 5.5. The regression curve for two packaging materials and two types of cheeses is given in Figure 5.9

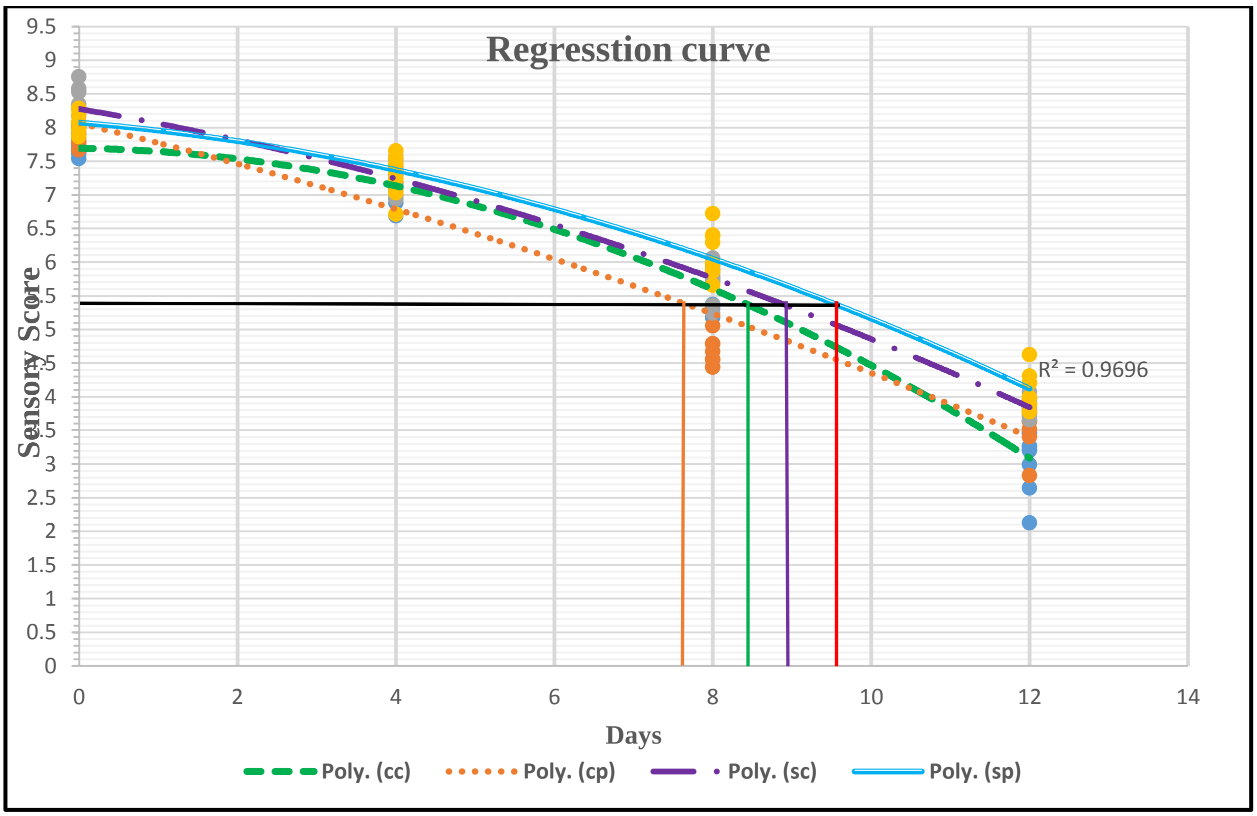

Fig 5.9: Regression curve for shelf life prediction

Regression equation: $Y = A + B_x + C_{x1}$

Whereas, Y = sensory score, A, B, C (constant)

$_x$, First variable and $x_{1,}$ Second variable

Regression equation For SP (Experimental Sample in Pouch)

A) pH contribution for changes in sensory score

Model	R Square	Un standardized coefficient	
1		Constant	-95.445
	0.895	pH (5.35-5.50)	18.831

Y= -95.445+ 18.831(pH)

B) TBA (Absorbance) contribution for changes in sensory score

Model	R Square	Un standardized coefficient	
1		Constant	21.641
	0.891	TBA	-224.731

Y= 21.641-224.731 (TBA)

C) TBA (Absorbance) and pH combined contribution for changes in sensory score

Model	R Square	Unstandardized Coeffiency	
1		Constant	-41.328
	0.895[a]	pH	0.181
	0.913[b]	TBA	-108.142

Y= -41.328 + 10.181(pH)-108.142(TBA)

5.7 MICROBIAL CHANGES

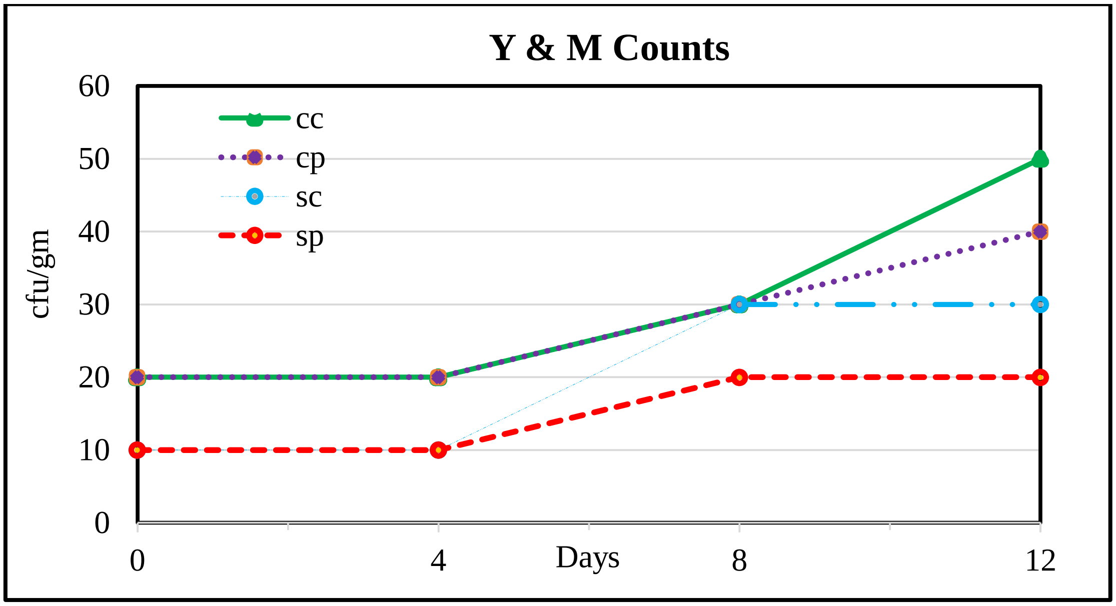

Fig: 5.10 Changes in Y & M counts in Mascarpone cheese samples stored at 5-8^{0}C

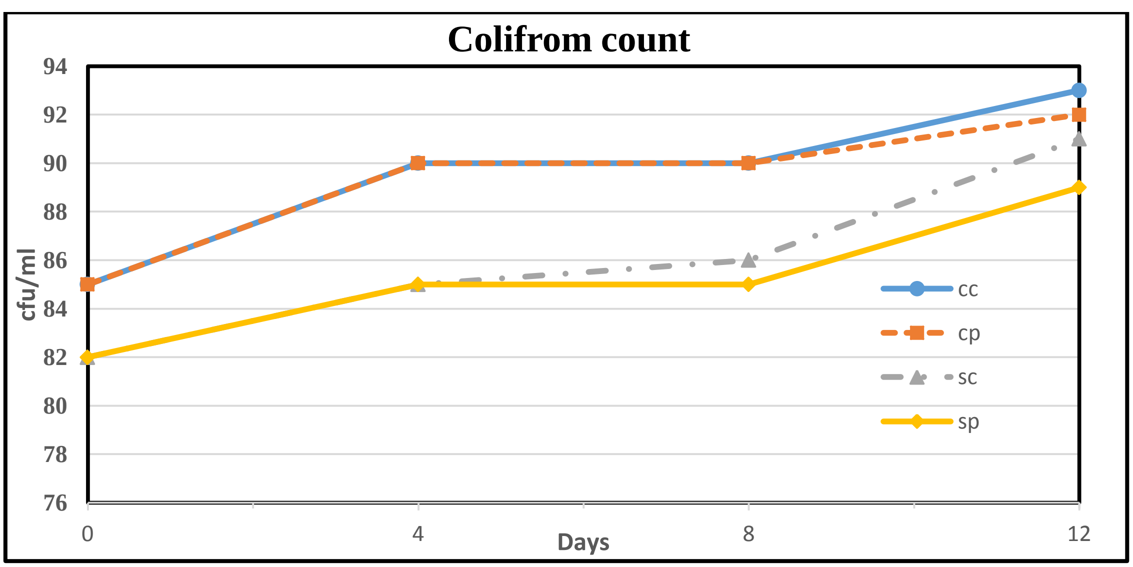

Fig: 5.11 Changes in Coliform count in Mascarpone cheese samples stored at 5-8^{0}C

5.7.1 Changes in Yeast& Mold counts

Fig 5.10 shows the yeast and mold counts (cfu/g) in cheese samples during storage at 5-8^{0}C. The initial counts were 20, 20, 10 and 10 cfu/g for CC, CP, SC and SP, respectively. Initial counts were found to be low in SC and SP compared to CC and CP samples because higher pH due to addition of fat replacer in SC and SP. All the counts increased with the increase in storage periods. It was observed that counts were more in cup packed samples than that of pouch packed samples. Initially, the rate of increase in counts was slow and almost constant till 8 days of storage. Thereafter, the rate of increase was more in both control samples but maximum was in cup packed control sample. The main spoilage of both the Mascarpone cheese was due to yeast and mould growth.

5.7.2 Changes in Coliform

The coliform count in both the Mascarpone cheeses increased during storage at 5-8^{0}C (Figure-5.11). The initial count of control and low fat fibre enriched Mascarpone cheeses were 85, 85 82 and 82 cfu/ g in CC, CP, SC and SP samples, respectively. Counts in both the samples increased during entire period of storage. Maximum growth of 95-100 cfu/g and minimum 80-85 cfu/g were observed in cup packed control and pouch packed low fat Mascarpone cheese at the end on 12th day of storage.

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Chapter- 6

Summary and Conclusion

6.0 SUMMARY & CONCLUSION

Mascarpone is high fat, soft varieties of Italian cheese. It is milky-white in color and easily spreadable. Its mild flavor and soft texture allow it to be used in savory and sweet dishes, making the possibilities nearly endless items in the kitchen. It is added to enhance flavor without overwhelming the original taste. It contains about 40% fat and does not contain fibre. Consumption of high fat is linked to enhance low density lipoprotein (LDL) and cholesterol level which in turn is linked to increase risk of atherosclerosis and cardio vascular diseases (CVD). Reduction of dietary fat is the primary goal of many consumers but low fat product suffers with flavour, textural attributes and high protein/ moisture content. Addition of suitable fat replacer can partially address these problems. Various types of fat replacers like inulin, WPC and N-creamer were used to achieve the desired textural characteristics as well as fibre enrichment in low-fat cheese. Intake of inulin as fibre provides many functional properties and physiological benefits. Therefore, an attempt was made to develop Mascarpone cheese with reduced fat and added fat replacer to enrich fibre which may satisfy health conscious consumers.

To optimise the method of preparation of Mascarpone cheese, milk fat in the form of 50% fat pasteurized cream was added at three different stages of coagulation. Out total calculated fat required to obtain 40% fat in cheese, i) 100% fat was added in milk before coagulation ii) 50% fat was added in milk and 50% fat was added in coagulum iii) 100 % fat was added in coagulum. Addition of 50% fat in milk and other 50 % fat in coagulum resulted in higher yield and higher fat recovery with good sensory score than its addition to milk before coagulation and in coagulum. Among three acids of acetic, citric and lactic acid, 4% diluted lactic acid coagulated cheese showed 2.0 - 2.5% more yield and lowest fat loss in whey (about 0.85%) and significantly more spreadability with low firmness. Overall sensory attribute was found to be best for the cheese made with lactic acid.

Inulin was added at different levels of 4, 6 & 8% in dry from and blended homogeneously with the coagulated mass. Moisture content and firmness of the cheese showed an inverse relation with the levels of inulin incorporation where as spreadability showed direct relation up to 6 % inulin incorporation. Inulin incorporation of 6% showed highest spreadability and lowest firmness. Cheese with 6% inulin scored significantly higher body and texture and overall acceptability scores indicating best resultant product. But the color & appearance score was non significant.

Mascarpone cheeses with 16, 20 and 24% fat (corresponding to 60, 50 & 40%fat reduction) and control with 40% fat were prepared. The influence of fat replacers in combination of the Inulin, WPC and N-creamer were studied by adding fat replacer in 3 combinations: 10% (6% inulin, 2% WPC & 2% N-creamer), 15% (6% inulin, 4.5% WPC & 4.5%N-creamer), 20% (6% inulin, 7% WPC & 7% N-creamer) into each level of reduced fat Mascarpone cheese on weight basis. Among the 10, 15 and 20% level of fat replacer addition in 40, 50 and 60 % fat reduced cheese, addition of 10% fat replacer in all reduced cheeses showed significantly higher spreadability and lower firmness than the addition of 15 and 20 % fat replacers. pH of experimental sample was higher than control and it increased with the increased level of fat replacer. Highest pH was observed in the reduced fat (16%) cheese added with 20% fat replacer. The results indicated that in general low fat cheese samples retained more moisture than full fat cheese. However, as the level of incorporation of combined fat replacer increased, the moisture content in the low fat cheese decreased. Fat reduced cheese with 10% fat replacer showed maximum whiteness than other samples. Based on physic-chemical and sensory observation, addition of 10 % fat replacer was found to the best in all the 3 levels of reduced fat cheese.

The trials were taken for optimization of fat reduction levels (40, 50 and 60%) with addition of 10% fat replacer (6% inulin, 2% WPC & 2% N-creamer). Among three reduced levels of 40,50 and 60% fat reduction, cheese with 16% fat (60% reduction) showed significantly higher moisture content (60.67%), higher pH (5.53) and maximum white colour (214.9) than other samples. The mean spreadability and firmness values of 60 % fat reduced cheese were closer to the full fat (40%) control Mascarpone cheese. Sensory scores also showed significantly higher colour and appearances score (7.38) and body and texture score (7.74) than other experimental samples but body and texture score was non significant with control sample score (7.52). Both low fat and fat replacer have negative effect on the flavour of cheese. Due to this reason, control (7.78) cheese had significantly higher flavour score than low fat experimental cheeses. Therefore, based on above observations, 60% fat reduction in cheese with addition of 10% fat replacer (6% inulin, 2% WPC& 2% N-creamer) was adjudged best.

The low fat and fibre enriched Mascarpone cheese was prepared and evaluated for its compositional, physico-chemical and sensory characteristics. Full fat control cheese was manufactured without any additives and compared. The physic-chemical characteristics of developed Mascarpone cheese were: total solids-38.02%, fat- 16.42 %, protein-8.42 %, carbohydrates -11.46 % and ash -1.53 %. pH was 5.4 whereas spreadability, firmness and water activity of developed sample were 1.84 ($N.s^{-1}$), 103 (N) and 0.96 respectively. The total solids and fat content were higher and protein, ash and carbohydrate contents were lower in control than that of developed low fat cheese. Sensory evaluation showed better overall acceptability for developed low fat cheese compared to control cheese.

Shelf life study of developed cheese was carried out by packing the products in polystyrene cup and polyethylene pouches and kept at 5-8^0C in refrigerator. Experimental sample packed in pouch showed higher shelf life than cup packed sample. Developed low fat cheese was good up to 9-10 days whereas control sample spoiled between 6-7 days. The pH decreased continuously whereas acidity and TBA increased in all the samples irrespective of packaging materials during entire period of storage. Between two packaging, spoilage was faster in cup packed samples as compared to pouch packed samples. A shelf life predication equation was developed based on sensory scores, pH and TBA values of the stored pouch packed samples. The predication equation is: Y = - 41.328 + 10.181(pH) - 108.142 (TBA) where variation of pH should be between 5.35 -5.50. Coliform count and yeast & mould counts increased in all the samples during storage. Further extension of shelf life may be possible with help of preservative and using other different types of packaging materials.

This study showed that reduction of fat in Mascarpone cheese with the addition of suitable fat replacer including inulin (fibre), whey protein concentrate and N creamer provides health benefits and reduces the risk for developing coronary heart disease, stroke, hypertension, diabetes, obesity and certain gastrointestinal diseases without any adverse effects on physico-chemical and sensory quality except flavour. This low fat, fibre enriched Mascarpone cheese may be a good substitute of butter.

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Annexure

ANNEXURE: 1

SENSORY SCORE CARD

DAIRY TECHNOLOGY SECTION

(9 POINT HEDONIC SCALE)

PRODUCT: **DATE:**

NAME OF THE JUDGE:

Characteristics	Sample					
	A	B	C	D	E	F
Color & appearance						
Flavour						
Body & Texture						
Overall Acceptance						

Remarks:

9-like extremely; 8-like very much; 7-like moderately: 6-like slightly; 5- neither like nor dislike; 4-dislike slightly; 3-dislike moderately; 2-dislike very much; 1-dislike9 extremely

ANNEXURE-III

Fig: Effect of fat addition at different stage on sensory attributes Mascarpone cheese

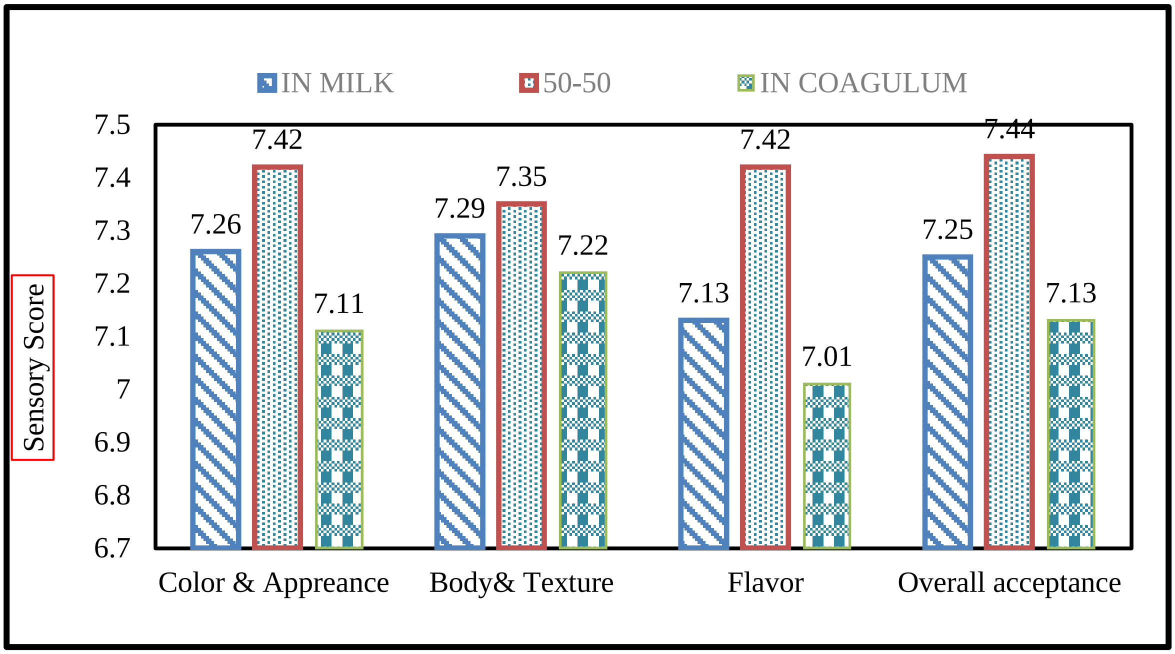


Fig: Effect of acids on sensory attributes Mascarpone cheese

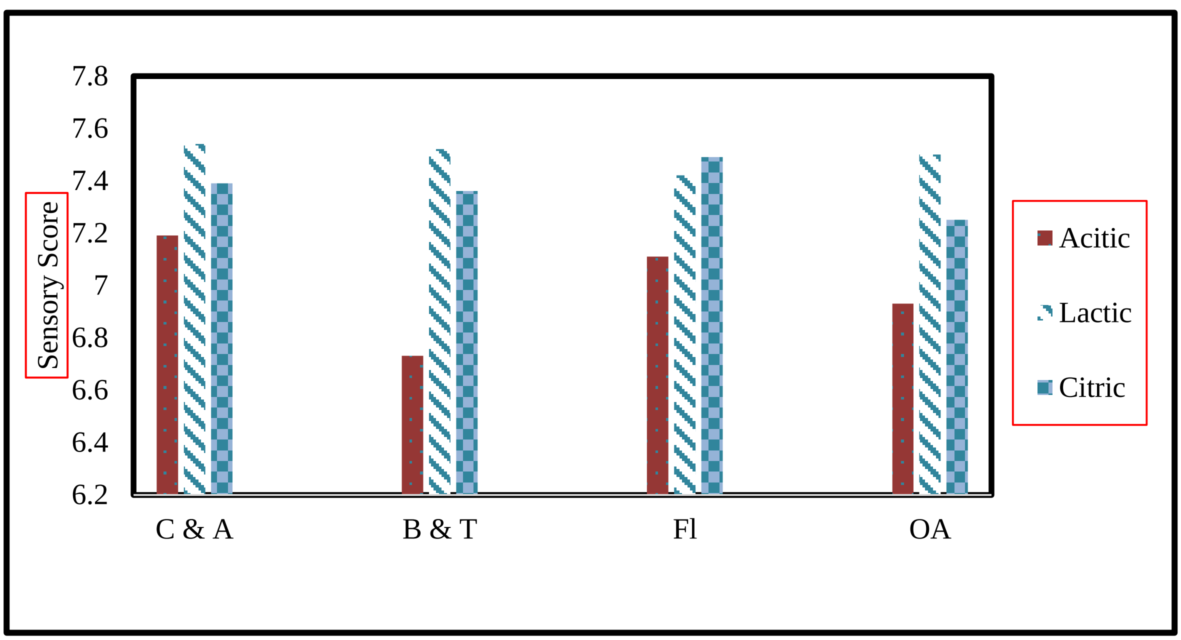
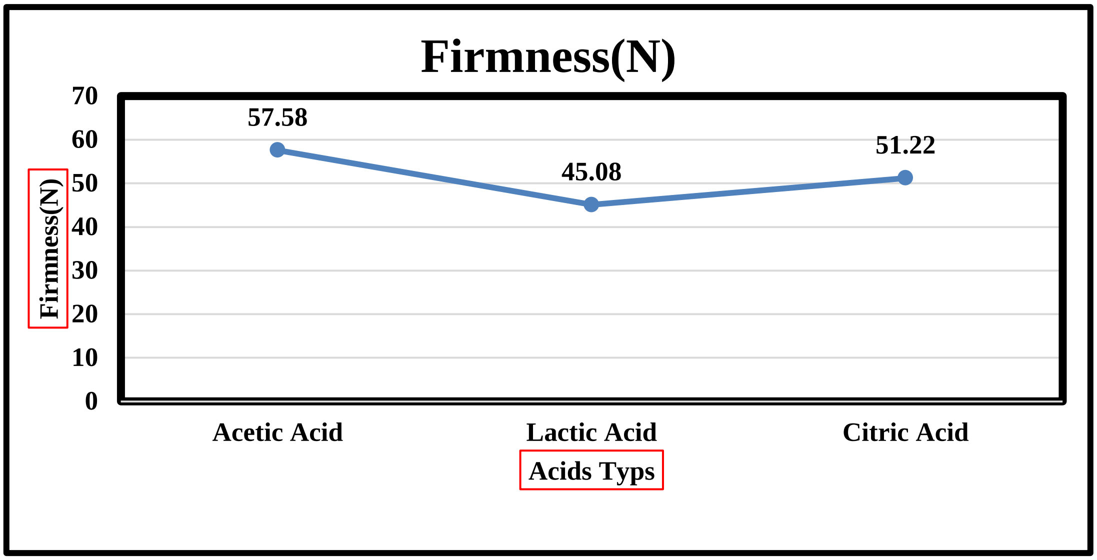


Fig: Effect of different acids used on the textural characteristics of Mascarpone cheese



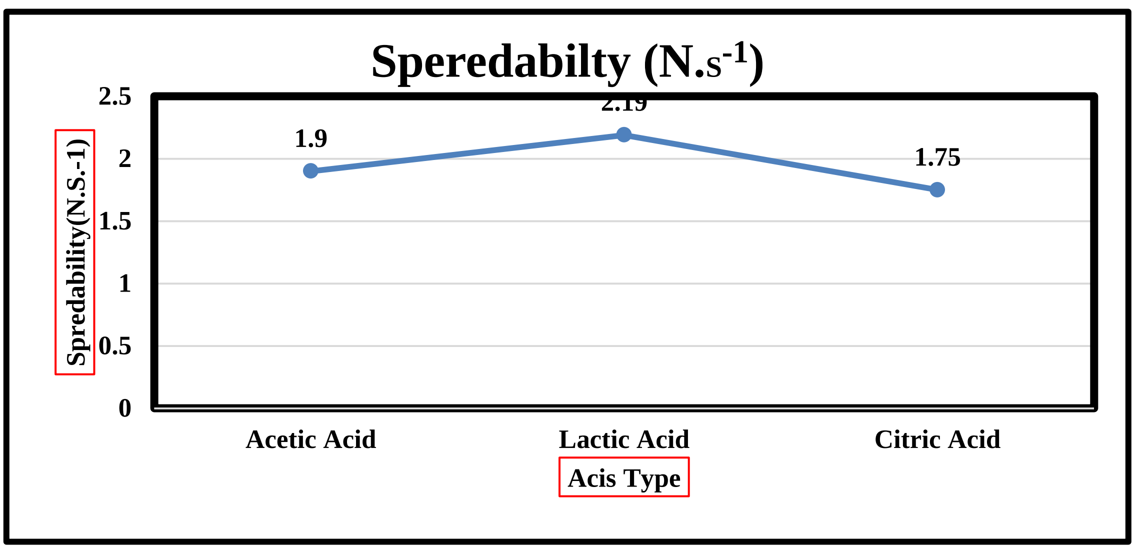
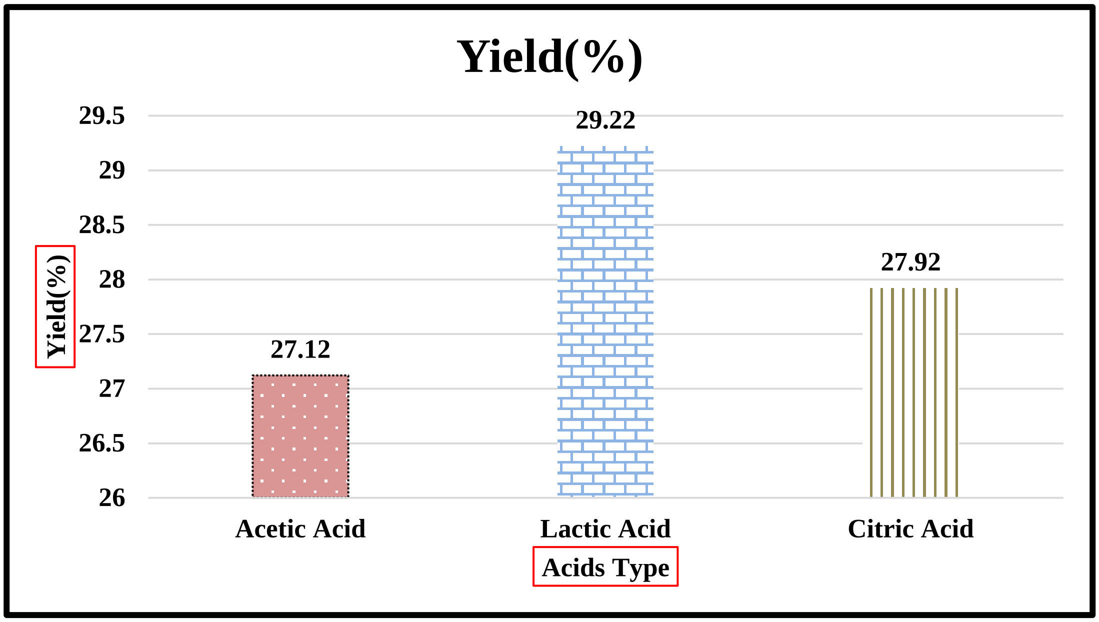


Fig: Effect of different acids used on the yield and fat loss in whey of Mascarpone cheese



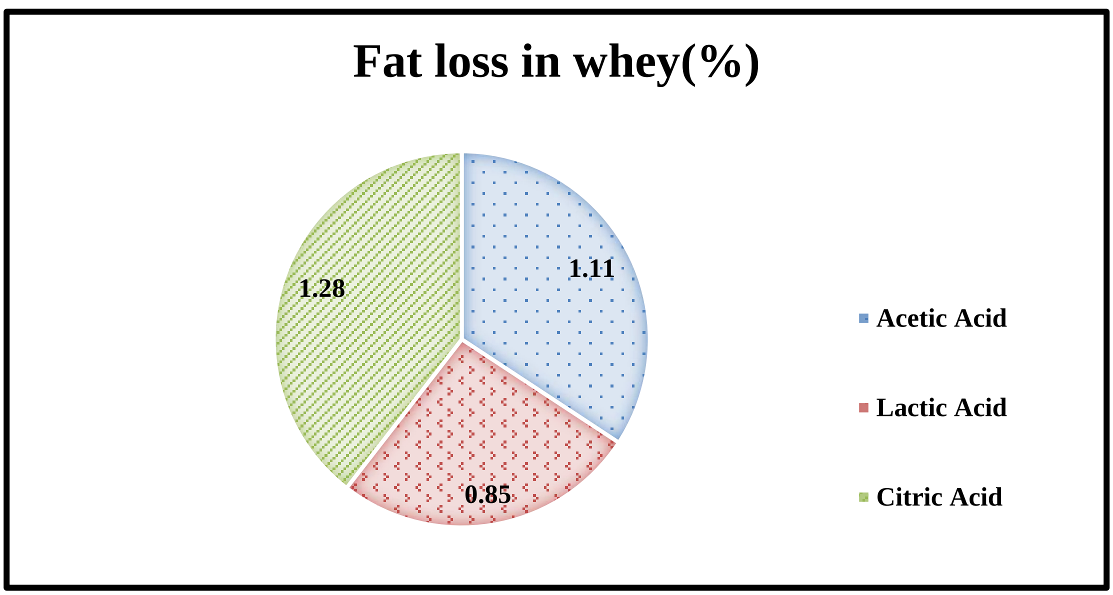


Fig: Effect of different levels of inulin incorporation on sensory attributes of Mascarpone cheese

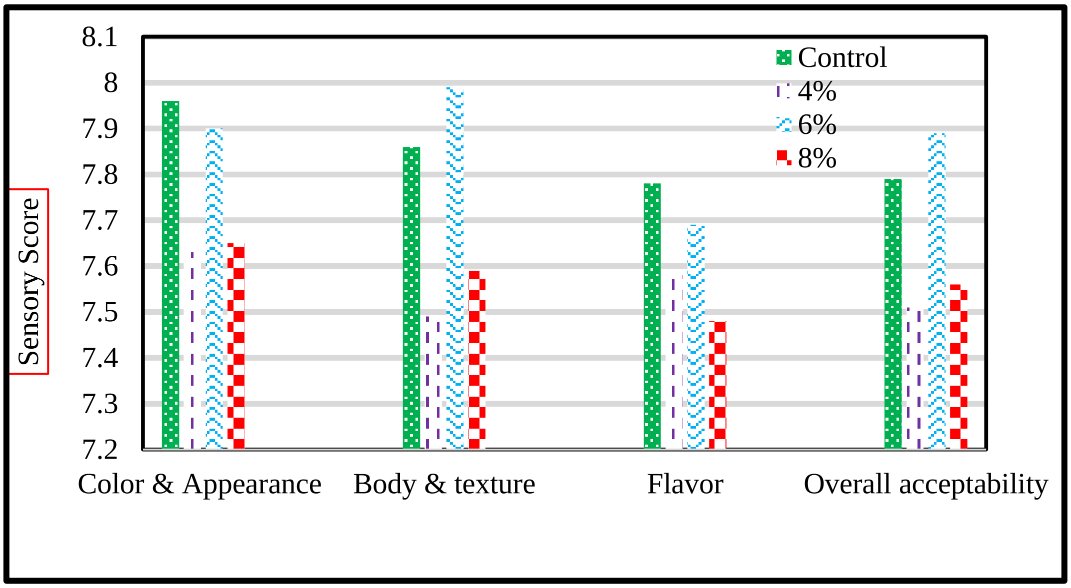
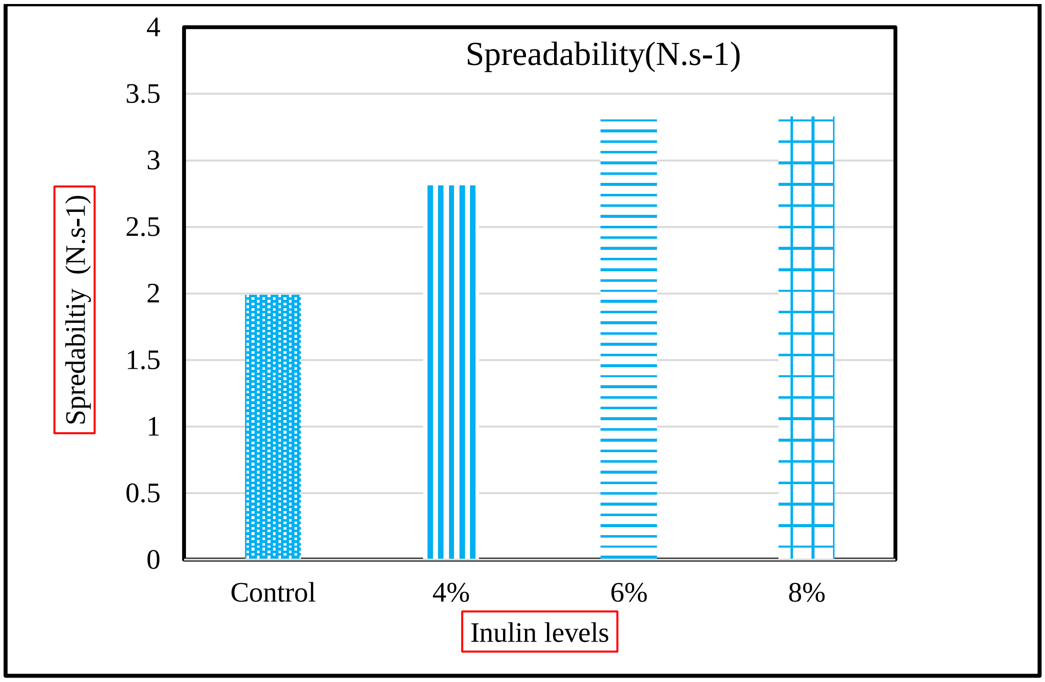


Fig: Effect of different levels of inulin used on the textural characteristics of Mascarpone Cheese



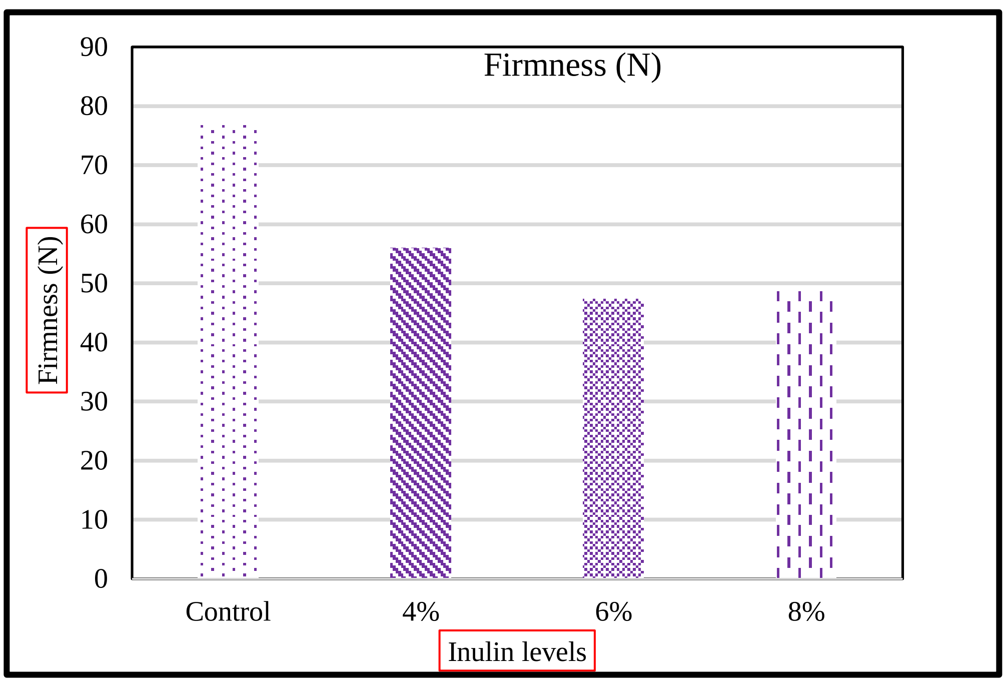
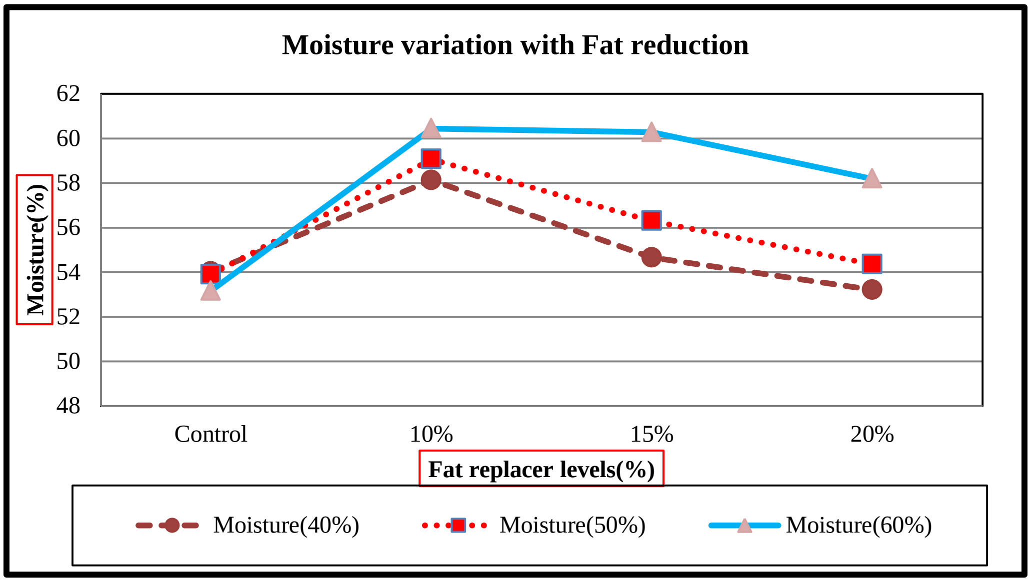


Fig: Effect of 40, 50 and 60% fat reduction with addition of 10,15 and 20% fat replacer on the physico-chemical characteristics of Mascarpone cheese



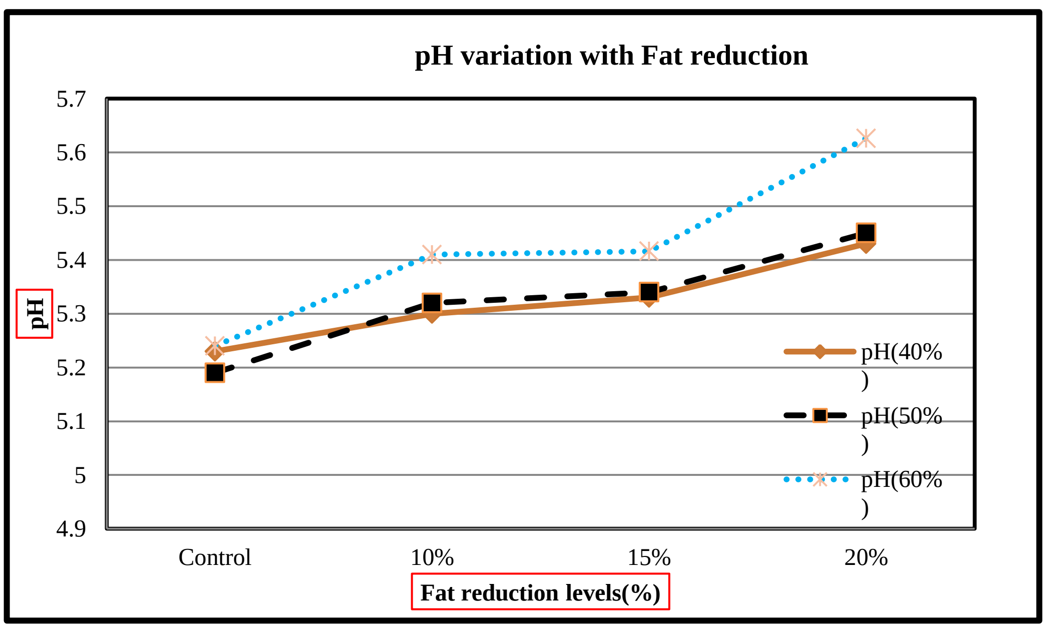
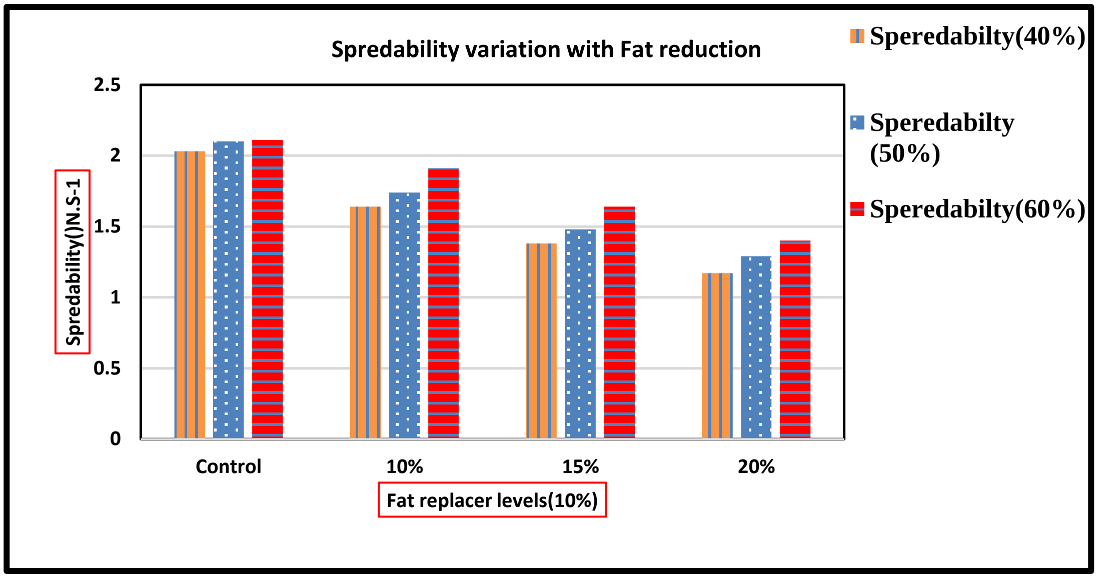


Fig: Effect of 40, 50 and 60% fat reduction with addition of 10, 15 and 20% fat replacer on the textural characteristics of Mascarpone cheese



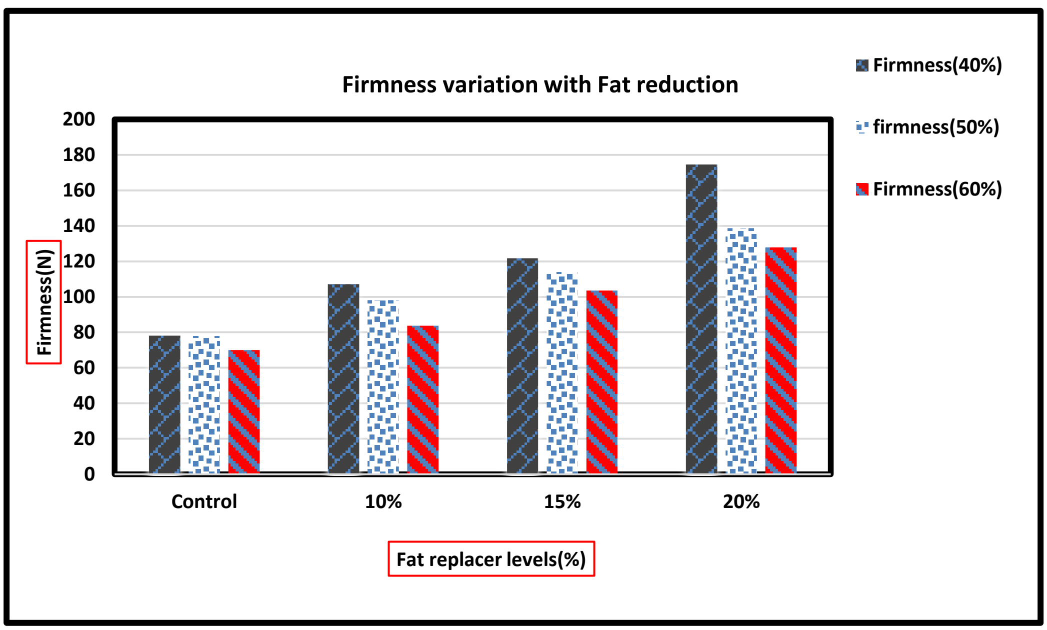
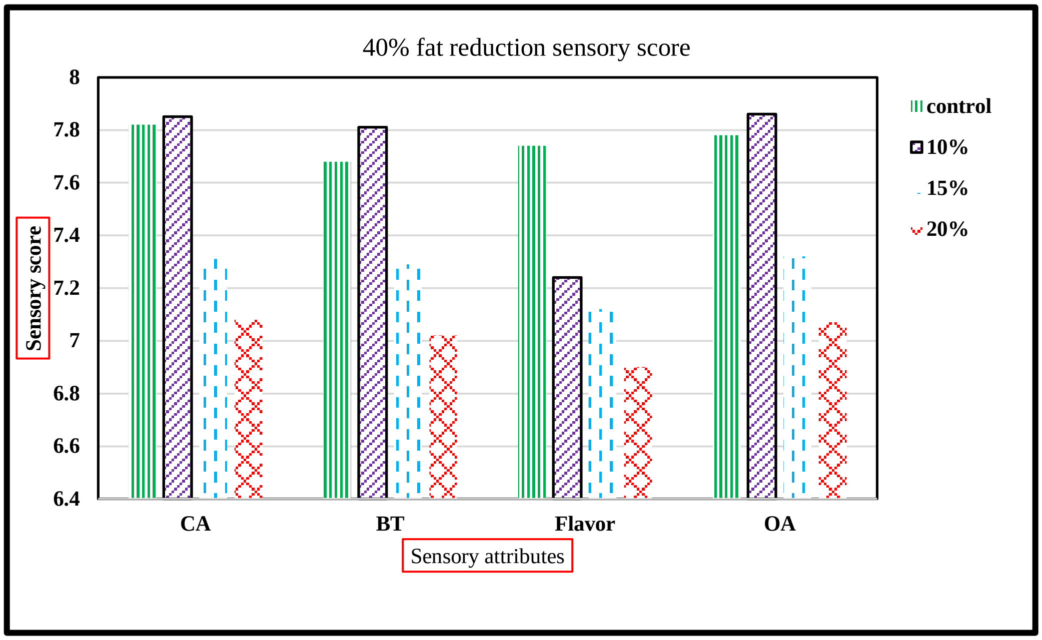
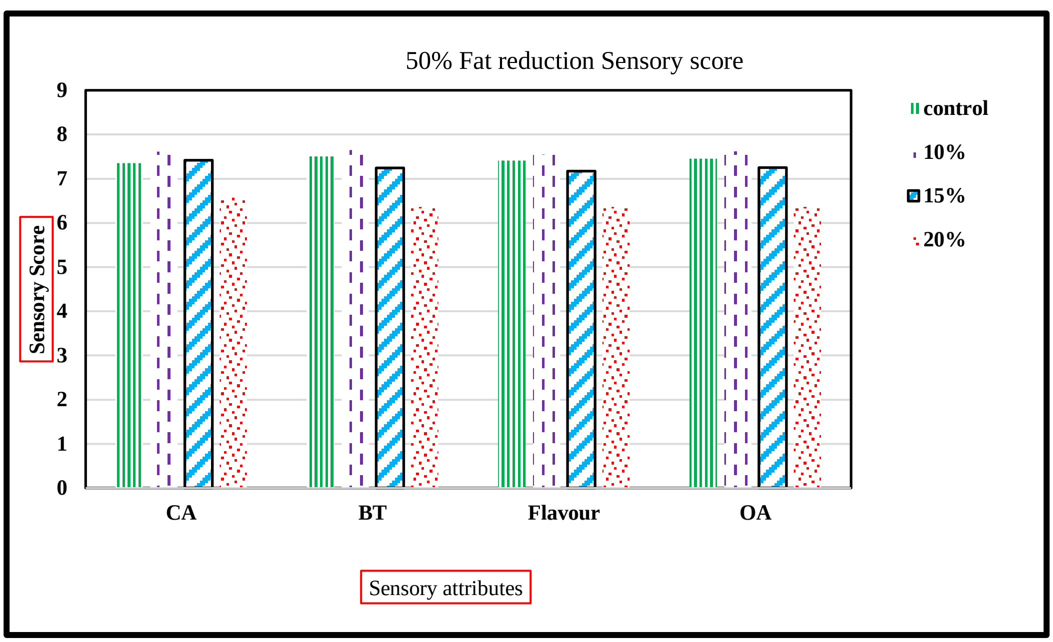
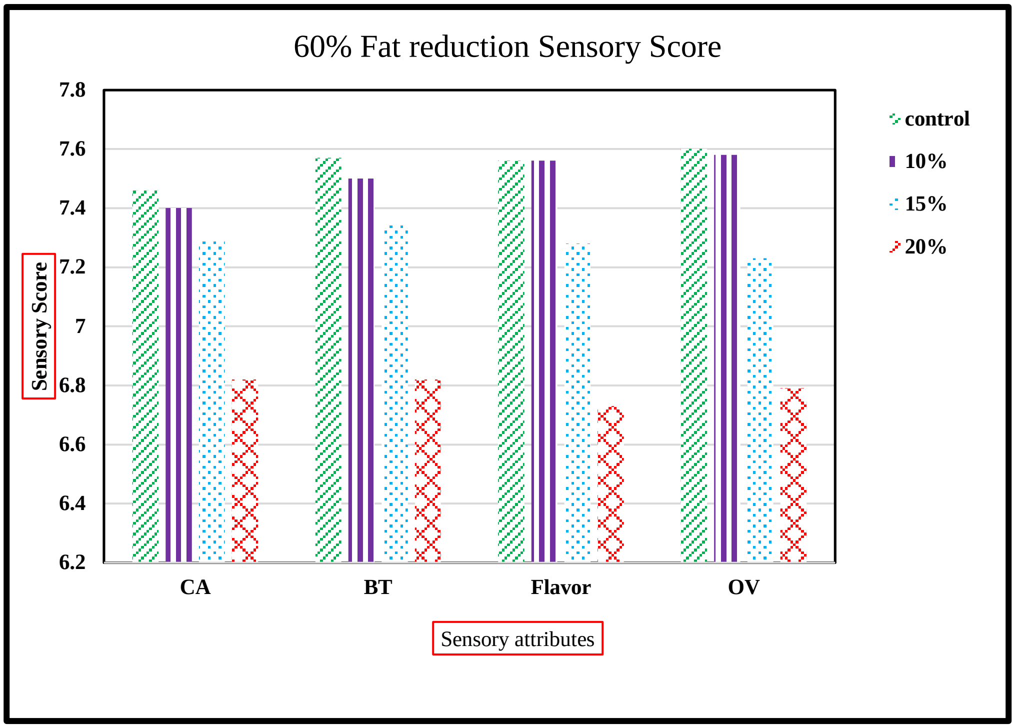


Fig: Effect of 10 % fat replacer (w/w) on the sensory characteristics of 40, 50 and 60% fat reduced Mascarpone cheeses






60% Fat reduction Sensory Score
7.8
7.6
7.4
7.2
7
6.8
6.6
6.4
6.2
Sensory Score
CA
BT
Flavor
OV
Sensory attributes
control
10%
15%
20%

ANNEXURE-III

Table: Microbiological Test of control in cup (CC), control in pouch(CP), experimental Sample in cup(SC) and experimental Sample in pouch(SP) stored at refrigeration temperature.

	Colifrom count (CFU/gm)				Yeast & Mould (CFU/gm)			
Days / Sample	0	4	8	12	0	4	8	12
CC	85	90	90	93	20	20	30	50
CC	85	90	90	92	20	20	30	40
SC	82	85	86	91	10	10	30	30
SP	82	85	85	89	10	10	20	20

Table: Change is pH acidity and TBA absorbance

	pH				**Acidity**				**TBA(Absorbance)**			
	0	4	8	12	0	4	8	12	0	4	8	12
CC	5.2	5.17	5.14	4.95	0.14	0.17	0.22	0.3	0.064	0.072	0.091	0.112
CP	5.2	5.18	5.14	4.99	0.14	0.17	0.22	0.29	0.066	0.07	0.094	0.103
SC	5.5	5.45	5.35	5.24	0.12	0.15	0.19	0.25	0.06	0.066	0.074	0.084
SP	5.49	5.45	5.39	5.28	0.12	0.15	0.18	0.23	0.06	0.064	0.069	0.077

Table: Change in sensory score of stored at refrigeration temperature.

	Storage Sample	**0**	**4**	**8**	**12**
Color & appearance	**CC**	**8**	**7.1**	**4.9**	**3.4**
	CP	**8**	**7.1**	**4.5**	**3.5**
	SC	**8.1**	**7.3**	**5**	**3.9**
	SP	**8.1**	**7.5**	**5.5**	**3.8**
Body & texture	**CC**	**8.1**	**7**	**4.5**	**3.3**
	CP	**8.1**	**7.2**	**4.6**	**3.4**
	SC	**8.2**	**7.4**	**4.8**	**3.7**
	SP	**8.2**	**7.6**	**5.2**	**3.8**
Flavour	**CC**	**8.3**	**6.9**	**4**	**3.2**
	CP	**8.3**	**7**	**4.3**	**3.3**
	SC	**8.5**	**7.3**	**4.5**	**3.5**
	SP	**8.5**	**7.4**	**4.9**	**3.6**
Overall acceptability	**CC**	**8.16**	**7.01**	**5.08**	**3.84**
	CP	**8.16**	**7.11**	**4.53**	**3.67**
	SC	**8.27**	**7.44**	**5.5**	**3.9**
	SP	**8.27**	**7.54**	**5.51**	**4.27**