EFFECT OF PORK SKIN COLLAGEN AS A FAT REPLACER IN LOW FAT FRANKFURTER

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DECLARATION

I hereby declare that this thesis, entitled **EFFECT OF PORK SKIN COLLAGEN AS A FAT REPLACER IN LOW FAT FRANKFURTER** is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

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Certified that this thesis, entitled **EFFECT OF PORK SKIN COLLAGEN AS A FAT REPLACER IN LOW FAT FRANKFURTER** is a record of research work done independently by **Dr. Selva Kumar. P** under my guidance and supervision and it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to him.

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Dedicated to
my
wife and son

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SELVA KUMAR. P.

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Introduction

INTRODUCTION

Urbanization, increased per capita income, improved standards of living, health consciousness and changing life style of people in developing countries led to increased demand for Ready-to-Eat and Ready-to-Cook meat products around the world, especially in developing countries. Sausages add variety and are versatile and popular products with good acceptability.

It is proved undoubtedly that high fat intake is associated with increased risk of obesity, colon cancer and saturated fat intake is associated with high cholestraemia, atherosclerosis and coronary heart disease (AHA, 1996; USDHHS, 1995). To prevent these health hazards various health organizations have promoted the reduction of fat and cholesterol intake in our diet. Consequent to the increased awareness of the adverse effects of excessive dietary fat, health conscious meat consumers are modifying their dietary habits to low fat diet. Hence the demand for low fat meat products is increasing in recent times.

At present emulsion type frankfurter sausages are generally manufactured with high fat levels of 20 to 30 per cent and fat contributes to nutritional, textural, sensory attributes and satiety.

By removing or lowering the fat in food products, many of the physico-chemical properties get altered leading to decrease in textural, sensory properties and overall acceptability of the processed meat products (Keeton, 1994). Although consumers prefer good food with minimal to no fat or low calories they also need their food to taste good. Hence improving overall palatability must assume utmost importance in any effort to reduce fat in reformulated meat products.

There is a great opportunity to develop low fat reformulated meat products like frankfurters with appropriate fat replacers and optimize their concentration to produce low fat meat products having better consumer acceptability and market value. Attempts have already been made to produce low fat pork sausages and beef sausages with good acceptability using fat replacers of plant based carbohydrates and proteins. But Claus and Hunt (1991) found that addition of plant proteins as fat replacers can impart undesirable characteristics like beany flavour with soy protein addition and grainy texture with fibre addition in the products. Low fat meat products with high water content have poor yield, texture and palatability. Collagen is used to improve water retention and fat emulsification and thereby improving the texture, flavour and succulence of the product. Pork skin collagen is an inexpensive adjunct to improve these traits. Pork skin connective tissue, a by-product of slaughter and fabrication of carcasses might be a potential water binder to replace fat in low fat or reduced fat meat products.

Hence the present study is carried out on low fat frankfurters with pork skin collagen as fat replacer. Labeling of the manufactured products with its nutritional facts and keeping quality has become a stipulation. Therefore, determination of the proximate composition, nutritional status and shelf life of the product is required for consumer acceptance and better marketing.

Hence the present study was under taken with the following objectives.

1. To develop a suitable formulary for low fat frankfurters with pork skin collagen as fat replacer.
2. To assess the effect of different levels of fat reduction and incorporation of pork skin collagen on the physico-chemical, textural, compositional and sensory qualities of the frankfurters.
3. To evaluate the shelf life under aerobic and vacuum packaging at -20^{o}C.

Review of Literature

REVIEW OF LITERATURE

The current study was conducted with the objectives of developing a low fat frankfurter with pork skin collagen (PSC) gel as a fat replacer and evaluating its physico-chemical, proximate composition, cooking and nutritional characteristics, organoleptic qualities and shelf life under aerobic (AP) and vacuum packaging (VP) at -20^{o}C. Search on available literature revealed that very little research has been carried out in the development of low fat frankfurters and a sizable volume of work has been carried out in other sausage products like beef, pork sausages and in beef patties with plant protein as fat replacers.

Therefore, literature on the rationale for development of low fat meat products, effect of fat reduction on sensory properties, development of low fat frankfurter, low fat frankfurter formulation, sensory, physical and cooking characteristics of meat products with varying fat levels and PSC gel as fat replacer and effect of aerobic and vacuum packaging systems of storage on various quality attributes of low fat meat products are reviewed.

2.1. RATIONALE FOR DEVELOPMENT OF LOW FAT MEAT PRODUCTS

High fat is associated with increased risk for obesity and colon cancer. Saturated fat intake is associated with high blood cholesterol and coronary heart disease. Saturated fatty acids and cholesterol are major dietary contributors to coronary heart disease and the Surgeon General's report on Nutrition and Health recommended diets low in cholesterol (AHA, 1996; USDHHS, 1995).

A survey conducted by Calorie Control Council found that 88 per cent of total adults reported consuming low fat, reduced fat or fat free foods and beverages (CCC, 1996).

World Health Organization has made recommendations to reduce daily fat intake so that it does not exceed 30 per cent of total calorie intake and to limit saturated fatty acids less than 10 per cent of total calorie intake, 6 to 10 per cent from poly unsaturated fatty acids, 10 to 15 per cent from mono unsaturated fatty acids less than 1 per cent from trans fatty acids and to limit cholesterol intake to 300 mg per day (WHO, 2003).

2.2. LOW FAT MEAT PRODUCTS

The definitions for nutrient claims as per Code of Federal Regulations (1995) indicate that total fat in low fat meat products should be $\leq$ 3g. If the fat per cent in the finished product is 10.41, 9.5, 4.5, 2.53 and 0.5-0.94, the product can be labeled as lite, lean, extra lean, low fat and fat free, respectively.

According to the Nutrition Labeling and Education Act of 1990, whole muscle beef products with no more than 10 per cent fat can be labeled as lean and products with less than 5 per cent fat as"extra lean" (Pearson and Gillet, 1997).

Allen *et al*. (1999) opined that traditional processed meat products have a relatively high fat content. Reduced fat products must show a 25% reduction of fat over traditional products.

Rheological behavior, pH, temperature alterations, meat particle size, mechanical action, fat distribution in the protein matrix, manufacturing procedures (design of machinery, etc.), process selection and properties (cooking, curing, smoking, drying, fermentation, etc,) and the end point characteristics are essential for fat reduction technology (Colmenero, 2000).

2.3. FUNCTIONS OF FAT IN MEAT PRODUCTS

Fat as a nutrient is useful for human health and development as well as for many physiological functions. Fat provides texture, mouthfeelness such as smoothness or creaminess and maintain moisture in foods and also carry, enhance and release the flavours of other food ingredients (Akoh, 1988).

Fat content has a basic effect on various physico-chemical and sensory characteristics such as flavour, mouth feel, juiciness, texture, handling, bite and heat transfer rate etc. The palatability and acceptability of meat products are directly related to the fat content (Pearson and Gillet, 1997).

2.4. TECHNOLOGICAL PROBLEMS WITH LOW FAT MEAT PRODUCTS

Production of low fat products through simple fat reduction substantially reduces product juiciness, tenderness flavour intensity and also making the product dry and rubbery (Pearson *et al.*, 1987).

The considerable influence the fat has on binding properties and texture caused problems in the preparation of reduced fat meat emulsion products (Cavestany *et al.*, 1994). Low fat (5 to 10 per cent) fresh or cooked or smoked sausages showed reduced cook yields, soft mushy interiors, rubbery skin formation, excessive purge in vacuum packages, shorter shelf life and changes in sensory qualities after cooking or reheating (Keeton, 1994).

Decreasing fat level from 30 to 5 per cent significantly ($P < 0.05$) increased the intensity of smokiness, spiciness and saltiness and reduced the overall acceptability of the flavour in low fat frankfurters (Hughes *et al.*, 1997).

Monahan and Troy (1997) reported that research into the relationship between fat level and quality in meat products has shown that the overall acceptability is affected by the fat level.

2.4.1. Flavour

Pearson (1997) pointed out that lack of flavour in low fat or reduced fat could be the direct result of reduction on flavour compounds or due to dilution and masking of flavour by additives that are added to replace fat and bind water. It is the loss of fat structure and texture in low fat products that makes them less flavourful.

The compounds responsible for species specific flavours in meat are due to the fat soluble components. Low fat meat products contain more water and as most of volatile aromatic components are oil soluble, the aroma is perceived as strong, harsh and unbalanced (Pearson and Gillet, 1997).

Hughes *et al.* (1998) reported that fat reduction increased the flavour intensity of the products probably by influencing the rate of release of flavour compounds.

2.4.2. Tenderness and Texture

Reducing fat levels to 10 per cent in meat products often resulted in cooked meat which was bland, dry with hard, rubbery and mealy texture (Keeton, 1994).

Andres *et al.* (2006) reported that as fat content increased more hard, gummy and cohesive product was obtained with higher chewiness, lower springiness and lower nutrient values in chicken sausages.

Nazeera (2007) reported that reducing fat levels significantly increased ($P < 0.05$) the shear force value in low fat restructured turkey meat loaves.

2.4.3. Juiciness

Huffman and Egbert (1990) reported improvements in juiciness with increased fat per cent from 5 to 20 in ground beef patties. Troutt *et al.* (1992b) opined that low fat (5 and 10 per cent) ground beef patties had lower juiciness and moisture release.

Hughes *et al.* (1998) reported increased overall flavour intensity and juiciness in low fat (5 and 12 per cent) frankfurter sausages.

2.4.4. Colour

Martin and Rogers (1991) explained that 50:50 beef-pork frankfurters have higher L^* values, lower a^* values and similar b^* values. Egbert *et al.* (1992) opined that colour stability of low fat ground beef product decreased with refrigerated storage time. Troutt *et al.* (1992a) revealed lower fat (5 and 10 per cent) patties had darker red colour.

Reducing the fat to lean ratio in meat products can increase product darkness and redness particularly if red meats are used and compensatory measures are not undertaken to modify colour (Keeton, 1994).

Hughes *et al.* (1998) opined that reducing fat level decreased the lightness and increased redness of the frankfurters although yellowness was not significantly affected.

Crehan *et al.* (2000) that explained reducing fat content from 30 to 5 per cent caused a significant decrease ($P < 0.05$) in L^*, b^* values and increase in a^* values in frankfurters.

2.4.5. Saltiness

Hughes *et al.* (1997) reported that decreasing fat content of pork frankfurters from 30 to 5 per cent resulted in increased intensities of smokiness, spiciness and saltiness.

Pearson and Gillet (1997) reported that it is critical to balance the salt and sugar levels to maintain flavour balance.

2.5. DEVELOPMENT OF LOW FAT FRANKFURTERS

One challenge of low fat meat production is finding ways to hold water and provide flavour, texture, and mouth feel and juiciness characteristics similar to those of full fat products (Eilert *et al.,* 1996).

Different processing strategies must be utilised to meet the low fat meat product market. Some strategies are identified to make low fat processed meats using leaner raw materials, replacement of fat with added water, tapioca starch, whey protein, gums and incorporation of connective tissue collagen (Cofrades *et al.,* 1997; Osburn *et al.,* 1997; Hughes *et al.,* 1998).

2.5.1. Meat emulsions

The quality attributes of low fat products depend upon the characteristics of the matrix formed which varies in accordance with the amount of protein and fat present (Bloukas and Paneras, 1993; Cavestany *et al.,* 1994).

2.5.2. Fat emulsification

Rust (1987) reported that myosin was a good emulsifier. Proteins like sarcoplasmic proteins and collagen can also emulsify fat particles.

Whiting (1988) reported that in order to coat fat particles, myofibrillar protein must first be extracted from the muscle using salt. An ionic strength of at least 0.6M is needed to prevent actomyosin aggregation before heating.

2.5.3. Protein gelation and fat stabilisation

Rust (1987) found cooking causes myofibrillar proteins to form a gel that holds the fat particle inside the protein shell while sarcoplasmic proteins form a weak gel that is less functional in retaining the fat particle.

Carballo *et al.* (1995) reported that an increase in the protein content during heating leads to the formation of much more stable gel matrix which permitted a smaller release of water and fat and thus producing lower total expressible fluid and purge loss values.

2.5.4. Water binding

Cavestany *et al.* (1994) reported decreasing fat by increasing water exhibited poorer binding properties in low fat products.

Carballo *et al.* (1995) reported that greater the proportion of protein and fat, the lower was total expressible fluid and smaller was the amount of liquid that separated off during storage.

2.6. FRANKFURTER FORMULATION

Frankfurters are comminuted semi solid sausages which are prepared from one or more kinds of raw skeletal meat and/or poultry meat. They shall contain fat and added water not more than 40 per cent collectively. They may be either smoked or unsmoked (Pearson and Gillet, 1997).

The maximum fat level for frankfurters was set at 30 per cent and added water at 10 per cent (Pearson and Gillet, 1997).

2.6.1. Meat and Nonmeat Ingredients

For ease of formulation pure lard may be used for additional fat in sausage manufacture (Brown and Ledward, 1987).

Acceptable frankfurter could be manufactured with a minimum of 1.3 per cent salt. As salt increased hardness, juiciness, saltiness and flavuor increased (Matulis *et al.*, 1995).

Sugar is used in meat products mainly to provide flavour or to mask the harshness of the salt. Sugars are used at levels from 0.5 to 2.0 per cent of meat. A variety of sugars such as sucrose, corn syrup and solids, dextrose and sorbitol are used (Pearson and Gillet, 1997).

Addition of pyrophosphates or diphosphates in emulsion products improves the water holding capacity, emulsification and protein extraction (Pearson and Gillet, 1997).

Meat inspection regulations specify that no more than 120 ppm of nitrite and 550 ppm of ascorbate be added as cure in emulsion type products (Pearson and Gillet, 1997; Ranken, 2000).

Finely ground spice can easily and completely be dispersed throughout the product than whole spices. When formulating a blend, it is best to start with low levels and to build the flavour gradually (Coggins, 2001).

Kilic and Richards (2003) studied the effect of pro-oxidative and anti oxidative factors in poultry doner kebab and reported that sodium ascorbate and vacuum packaging inhibited lipid oxidation with sodium ascorbate being the more effective antioxidant treatment.

2.6.1.1. Fat Replacers

2.6.1.1.1. Water

Currently USDA- FSIS (1990) allows sausages to contain a total of 40 per cent added water and fat and fat can only comprise a maximum of 30 per cent of the final product

Reducing fat without a concurrent addition of water increased the product cost due to higher lean content (Gregg *et al.*, 1993).

Reduction of fat in meat emulsion is generally accompanied by an increase in the water content and this alters the nature of system affecting texture, binding properties, colour etc (Cavestany *et al.*, 1994).

2.6.1.1.2. Collagen

Puolanne and Ruusunen (1981) reported that connective tissue may be important in the preparation of sausage not only with regard to water binding but also improving the firmness of cold sausage.

Commercial British sausages show about 35 per cent of the protein in the form of collagen and similar levels are indicated with other products containing pork rind, gelatin or shin beef (Hannan, 1984).

Collagen sources used in meat products manufacture include pork rinds, hide sections, gristles and edible bone collagen. Collagen hydrolysates are also used including gelatin from pork skin, hide and bone (Jobling, 1984).

All poorly digested residues whether of plant or animal origin should be considered as important dietary components and hence collagen is an important dietary component (Shrimpton, 1984).

Kenney *et al.* (1992) observed that raw and preheated connective tissues were useful in increasing tensile strength when added at 10 per cent of the formulations however, at 5 per cent added level only raw connective tissue was effective in increasing tensile strength. Addition of 1 per cent gelatin increased tensile strength but reduced cook yields.

Eilert and Mandigo (1993) found that heating collagen for 10 to 15 minutes at 48^{o}C-52^{o}C was adequate to separate soluble collagen in thermally processed products like frankfurters.

Osburn and Mandigo (1998) reported that chicken connective tissue gel formation might be enhanced by heating chicken connective tissue to 60^{o}C or by reducing its fat content in reduced fat bologna and gels with high hydration values (> 1.04) may bind additional added water (>200 per cent).

Schilling *et al.* (2003) reported that collagen inclusion increased water binding and decreased cooking loss values in boneless cured ham by acting synergistically with myofibrillar proteins.

2.6.1.1.3. Collagen and Connective tissue in meat emulsions

Rust (1987) recommended that collagen should be limited to 25 per cent of the total protein content in a sausage.

Eilert *et al.* (1993) reported that inclusion of higher levels of modified connective tissue in high fat formulations of frankfurters increased stability and thermal processing yield also increased proportional to that. Modified connective tissue collagen has useful functional characteristics such as water and fat binding.

Collagen proteins have the ability to coat fat particles. However, since collagen is not heat stable, the proteins shell formed by collagen can melt away during thermal processing and form gelation, thus freeing the fat particle (Pearson and Gillet, 1997).

2.6.1.1.4. Beef Connective Tissue and Gel Incorporation

Satterlee *et al.* (1973) revealed that enzymatically hydrolysed beef and pork skin used in batters found to have improved water and fat holding capacity and improved stability during cooking.

Powdered or preheated powdered connective tissue at levels of 5 per cent or 10 per cent in restructured steaks increased tensile strength and cook yield and 10 per cent level reduced sensory cohesiveness, juiciness and beefiness (Kenney *et al.,* 1992).

Sausages mixed with precooked tendon were more desirable in texture, flavor and acceptability than sausages with raw tendon (Saddler and Young, 1993). Eilert *et al.* (1993) reported that increasing the amount of connective tissue increased batter pH, emulsification temperature and peak force and energy to extrude.

Meullenet *et al.* (1994) reported that sensory attributes of chicken frankfurters with 2 per cent collagen fibres and about 20 per cent added water was comparable with reference hotdogs made with 100 per cent mechanically deboned poultry or beef and fat (50:50) each with 10 per cent added water.

2.6.1.1.5. Pork Skin and Gel Incorporation

Heating at 70^{o}C for 30 min was sufficient to enhance the water binding ability of pork skin connective tissue and more water was bound at 70^{o}C than at 50^{o}C. Pork skin could hold 3 g of water per g of skin. Pork skin gels, which effectively bind water in processed meat, can be formed by heating pork skin and water. Pork skin gels improve water holding capacity in processed meat and can also be used to alter sausage texture and colour. The pH of pork skin gels ranged from 7.42 to 7.69 as the amount of water in the gel increased from 100 to 600 per cent of the weight of the pork skin respectively. (Osburn *et al.,* 1997).

Gels made with < 300 per cent added water had melting point and cook stabilities that were suitable for addition into processed meats Osburn *et al.* (1997).

Consumer panelists rated bologna containing 30 per cent addition of a 500 per cent added water pork skin gel as 5 on an 8 point scale for overall acceptability and juiciness (Osburn *et al.,* 1997).

Osburn and Mandigo, (1998) reported that the properties of gels made from chicken skin were very similar to those of pork skin gels. Both beef and chicken skin gels have potential for use as water binders and texture modifiers in low fat comminuted sausages.

Schnell (1999) reported that pork skin collagen gel incorporation significantly increased ($P < 0.05$) the water holding capacity in low fat bologna.

Prabhu *et al.* (2004) Incorporation of pork collagen protein significantly increased ($P < 0.05$) cook yields, L^* b^* values and reduced the purge loss values in low fat frankfurters and ham.

2.6.1.2. Binders and Extenders

In order to bind water during cooking and reduce purge loss during storage, the starch selected for use in reduced fat emulsion products should not increase batter viscosity and should become functional during cooking process (Kendall and Mitolo, 1993).

Rogers *et al.* (1996) evaluated modified starches to determine their effects on the characteristics of fat free bologna and reported that all starch containing product displayed less purge.

The ability to gel and hydrate at normal processing temperatures, low sweetness and reducing characteristics had made food starches more accepted and more commonly used in processed meat and poultry items (Eilert and Mandigo, 1997).

Products identified as meat loaves are restricted to have 3.5 per cent of extender materials as allowed in other sausage products (Pearson and Gillet, 1997). Binders are used to tie up the water during processing and prevent purge during storage and improve sliceability (Smith, 1997).

2.6.2. Processing

2.6.2.1. Final Internal Processing Temperature

Sadler and Young (1993) adopted a final internal temperature of 80^{o}C in water bath to study the effect of pre heated tendon in fine emulsion sausages.

Effects produced in meat products by variation of fat and protein contents are much influenced by heat treatment (Carballo *et al.,* 1995).

Heating rate affects the texture of frankfurters regardless of fat content and slower heating rate produces a more fully formed gel net work and could be manipulated to induce certain textural properties in low fat frankfurters (Cofrades *et al.,* 1997).

Pork collagen added frankfurters were cooked to a final internal temperature of 72^{o}C (Prabhu *et al.,* 2004).

2.7. QUALITY ATTRIBUTES OF LOW FAT MEAT PRODUCTS WITH COLLAGEN

2.7.1. pH

Osburn *et al*. (1997) in pork skin connective tissue gel utilization in reduced fat bologna reported that the pH ranged from 6.87 in uncooked batter to 7.50 in cooked bologna.

Lin and Chuang (1999) studied the physico-chemical properties and shelf life of low fat Chinese style sausage and observed stable pH values of 6.2 for low fat sausage during storage at 4^{o}C.

Klettner *et al*. (2003) observed good correlation between weight loss due to roasting and pH in pork, beef and turkey meat and reported lower weight loss for meat with higher pH.

2.7.2. Emulsion Stability

Satterlee *et al*. (1973) reported that addition of collagen causing increased emulsion stability by increasing water binding and fat emulsification.

Eilert *et al*. (1993) who reported decreased emulsion stability in 24 per cent fat than 8 per cent fat in meat batters manufactured with modified beef connective tissue.

Schnell (1999) reported a positive correlation between pH and emulsion stability and expressible moisture in low fat bologna with pre gelatinized pork skin collagen.

Martinez *et al*. (2004) reported that 20 per cent fat pork frankfurter had significantly lower emulsion stability values than 9 per cent.

2.7.3. Cooking Characteristics

In bologna sausages greater the fat content lower were the total expressible fluid and expressible moisture and greater was the amount of fat released (Cavestany *et al.*, 1994).

Cooking loss was significantly lower in high fat than in low fat bologna sausages (Carballo *et al.*, 1996). Eilert *et al.* (1996) reported an increase in the processing yield of 10 per cent fat frankfurters when beef connective tissue was added at 20 per cent of the formulation.

Sheard *et al.* (1998) reported that in high fat content sausages and burgers, the fat loss was much more than in low fat products regardless of the method of cooking followed.

Incorporation of pork skin collagen at 1 per cent or above significantly increased ($P < 0.05$) cooked yields in frankfurters (Schnell, 1999).

Prabhu *et al*. (2004) reported cook yield increased when pork collagen content was increased from 0 to 3 per cent in emulsified and whole muscle meat products.

Serdaroglu and Degirmencioglu (2004) reported that the cook yield was lowest for 20 per cent fat than 10 and 5 per cent fat in Turkish meat balls and attributed it to the excess fat separation and water release during electric grilling.

2.7.4. Dimensional Shrinkage

El-Magoli *et al.* (1996) reported that in low fat ground beef patties, reduction of fat level from 22 to 11 per cent caused reduced shrinkage.

Troy *et al.* (1999) reported that all treatments of beef burger had a reduction in diameter. The full fat control shrunk the most due to high loss in fat and moisture during cooking.

Turkish meat balls formulated with 20 per cent fat had the highest reduction in diameter (Serdaroglu and Degirmencioglu, 2004).

2.7.5. Water Holding Capacity

Webster *et al.* (1982) reported the ability of collagen to increase the water holding capacity in processed products and attributed this to the collagen functionality of increased water binding and hydration.

Colmenero *et al.* (1996) reported that high fat sausages exhibited better water holding capacity than low fat and reported that greater the fat content, lower the total expressible moisture and greater was the amount of fat released in bologna sausages.

Hughes *et al.* (1997) reported that decreasing fat content of frankfurters from 30 to 5 per cent decreased the water holding capacity.

Osburn *et al*. (1997) in reduced fat bologna with pork skin collagen and Osburn and Mandigo (1998) in reduced fat bologna with pre heated chicken connective tissue reported that collagen caused improved water holding capacity of the respective products.

Bologna with pre-gelatinsed pork skin and water at 5 and 25 per cent respectively had reduced purge (Schnell, 1999). Low fat sausages had higher expressible moisture than those of regular fat controls (Yoo *et al.* 2007).

2.7.6. Texture Analysis

Arganosa *et al.* (1987) reported that collagen yielded a lower mean shear value at 20 per cent fat replacement in pork sausages. Troutt *et al.* (1992b) reported that Warner-Bratzler Shear and Lee Kramer Shear forces decreased as fat level increased in beef patties.

Meullenet *et al*. (1994) reported that increasing the level of collagen fibres to 5 per cent had lower shear stress values in low fat high added water chicken frankfurters.

El-Magoli *et al.* (1996) reported that Texture Profile Analysis showed an increase in chewiness for low fat sample over the high fat control, while hardness and springiness remained unaffected in ground beef patties.

Osburn *et al*. (1997) and Osburn and Mandigo (1998), observed improved water binding and gelling ability of collagen might have improved the texture by diluting the stronger binding myofibrillar protein in low fat formulations.

Low fat beef burgers were analysed using the Instron Universal Testing machine and highest shear force values were recorded in low fat controls (Troy *et al*., 1999). Yoo *et al.* (2007) reported decreased texture profile analysis values with increased added fat level in sausages.

2.7.7. Colour

Arganosa *et al.* (1987) reported that replacing the fat portion of the pork sausage with collagen did not affect the L^* and a^* values of the uncooked patties however, the b^* values were significantly lower at 15 per cent and 20 per cent levels of collagen.

Young *et al.* (1991) concluded that 15 and 20 per cent fat chicken patties had higher L^* and b^* values though a^* values were not affected.

Increasing the per cent of gel incorporation into bologna increased L^* and decreased cured colour ratio and a^* value (Osburn *et al.*, 1997). Jo *et al.* (1999) observed hunter L^* value of pork sausages increased with increase in fat content and storage than low fat pork sausages.

Abiola and Adegbaju (2001) reported that low fat pork sausages were darker than high fat pork sausages. Addition of light coloured collagen diluted the dark colour of low fat products (Schilling *et al*., 2003). Prabhu *et al*. (2004) who reported that use of collagen increased L^* and b^* values in frankfurters

2.7.8. Proximate Composition and Nutritional Quality

Sheard *et al.* (1998) studied the chemical composition and energy content of sausages and reported that average fat content before and after cooking were 22 g and 17 g and average energy content before and after cooking were 1215 KJ and 1016 KJ for the sausages.

If a serving of food supplies at least 10 per cent of the individual's RDA of a nutrient, it is considered as a good source of that nutrient. The RDAs are the amounts of essential nutrients considered adequate to meet the needs of healthy individuals. The RDAs are used by nutritionists and dietitians and as the basis for most public health programmes (Lupton and Cross, 1999).

Abiola and Adegbaju (2001) reported 53.51 per cent moisture, 24.66 percent fat, 12.41 per cent protein and 4.48 per cent ash in pork sausages. Andres *et al.* (2006) reported that chicken sausages contain 76.01 per cent moisture, 14.34 per cent protein, 0.61 per cent lipids and 2.74 per cent ash.

2.7.9. Sensory Analysis

Addition of 20 per cent beef connective tissue improved processing yields and decreased cohesiveness of 10 per cent fat 30 per cent added water frankfurters (Eilert *et al*., 1996).

High fat frankfurters were harder and chewier than low fat frankfurters. Low fat high added water frankfurters were springier and more cohesive (Cofrades *et al.*, 1997).

Decreasing fat content of frankfurters from 30 to 5 per cent reduced the overall acceptability, flavour and caused darker and redder coloration of the reduced fat frankfurters (Hughes *et al.*, 1997).

Traditional Greek sausages with 20 per cent fat had the highest score for all sensory attributes and sausages with 30 per cent fat were very light yellow, soft and too fatty when grilled, where as those with 10 per cent fat were dark, very hard after grilling (Papadima and Bloukas, 1999).

Sensory analysis parameters of low fat meat products including colour, flavour, chewiness, juiciness, saltiness, spiciness, smokiness, elasticity, firmness, coarseness, greasiness, overall acceptability etc and consumer evaluation are performed depending upon alteration factors in fat reduction process (Colmenero, 2000).

Serdaroglu and Degirmencioglu (2004) reported that 20 per cent fat meat balls have more overall acceptability.

2.8. PACKAGING AND STORAGE

The meat product packages function to provide protection against damage, physical and chemical changes and further microbial contamination during all subsequent storage, handling and merchandising. The packaging material must be moisture proof in order to prevent product dehydration and surface discoloration and must not impart any off odour and off flavour to the product and should retain the natural flavours and odours that are inherent to the product (Forrest *et al.*, 1975).

Chunked and formed products are normally either cured and heat processed or fresh frozen. Cured products include sausages and loaves. The four major problems related to the packaging of restructured meat products are microbial spoilage, change in colour, lipid oxidation and moisture loss. Selection of a packaging system can significantly influence the oxidative stability of restructured products and therefore, headspace control techniques (vacuum, gas flush, shrink and skin packaging) are extensively used to control oxidative rancidity (Harte, 1987).

2.8.1. Packaging Materials

Polyethylene bags are commonly used as packaging material due to their low cost and convenience. The use of PE bags may result in reduced shelf life of meat products because of increased fat oxidation and moisture absorption rate in comparison to products in vacuum bags, which are more costly (Almeida-Dominguez *et al*., 1992). Packaging material used for meat products are usually plastics in which polymers with good O_2 barrier properties (polyamide) are incorporated with polymers with good humidity barrier and sealing properties such as polyethylene (PE) and polypropylene (Gedde, 1999).

Polyamides (nylons) have relatively high melting points and low gas permeability, but they will absorb moisture and lose strength when exposed to moisture. Vacuum and modified atmosphere packaging has been used to extend the shelf life of packaged meat for several decades. Processed meat is typically packaged in heat shrinkable films. Either nylon or polyethylene terephthalate based film with a heat sealable layer (ionomer-surlyn or ethylene vinyl acetate) is used for processed poultry meat (Dawson, 2001).

2.8.2. Packaging Systems and Storage Stability on the Quality of Low Fat Meat Products

2.8.2.1. Purge

Colmenero *et al.* (1996) observed freezing and frozen storage can cause increased purge loss in low fat bologna sausages due to loss of binding properties and the inability to restrain water and total expressible fluid which is more pronounced in the lower fat levels in bologna sausages. They also observed that high fat content can decrease vacuum purge that occur during storage in bologna sausages.

Incorporation of pork skin collagen at 1 per cent or above significantly decreased ($P < 0.05$) purge loss in frankfurters (Schnell, 1999). Pork collagen effectively controlled purge in both the frankfurters and restructured ham after 4 and 8 weeks of refrigerated storage (Prabhu *et al.*, 2004).

2.8.2.2. Lipid Oxidation

St. John *et al.* (1986) reported that frankfurters made using pork fat at 30 per cent and 25 per cent level showed TBA values less than 1.0 for rancidity after 6 weeks under vacuum packaging.

Arganosa *et al.* (1987) reported that increasing the levels of food grade collagen significantly decreased the TBA values in pork sausages.

Lipid oxidation is one of the primary causes of deterioration in the quality attributes of meat and meat products on storage leading to the development of off flavour, loss of colour and texture, and decrease in nutritive value (Johns *et al*., 1989).

Brewer *et al.* (1992) reported that time in frozen storage increased off flavour and TBA value in ground beef patties. Low ground beef patties had greater lipid oxidation stability (Bullock *et al.*, 1994).

Morrisey *et al*. (1994) and Buckley *et al*. (1995) reported that lipid oxidation leads to discolouration, drip loss, off odour and off flavour development and the production of potential toxic compounds.

TBARS values for both vacuum packaging and modified atmospheric packaging in Chinese style sausages increased significantly with storage time, VP resulted in greater amount of oxidation than MAP (Wang *et al.*, 1995).

Jo *et al.* (1999) observed that TBARS values of cooked pork sausages increased with increase in fat content regardless of storage, irradiation and packaging types.

Kao and Lin (2006) observed a very slight increasing trend in TBARS values of reduced fat pork frankfurters during chiller storage. Frankfurters with 28 per cent fat added were generally higher in TBARS than reduced fat treatments at any storage period and attributed this to high fat content.

2.8.2.3. Sensory Qualities

Bullock *et al*. (1994) reported that after 24 weeks storage at –20^{o}C, the sensory attributes of low fat ground beef patties significantly decreased from the initial evaluation period. The greatest change in lean occurred between week 12th and 24th week.

Ho *et al. (*1995) reported that the juiciness scores for reduced fat pork sausage patties packaged in polyethylene declined faster than those packaged in vacuum pouches.

Schnell (1999) observed incorporation of pork skin collagen at 1 per cent or above resulted in higher juiciness and lower resistance to bite in frankfurters.

2.8.2.4. Nutritional Quality

Smith and Alvarez (1988) studied the stability of vacuum cook-in-bag turkey breast roll during refrigerated storage and observed no significant change in proximate composition during 87 days of storage at 4^{o} C.

Wu and Sheldon (1988) reported that turkey beast tissue contained 2 per cent lipid which remained constant over 4 day's storage at 4^{o}C.

Papadima and Bloukas (1999) studied the effect of fat level and storage conditions on quality characteristics of traditional Greek sausages and reported that storage conditions of 3^{o}C to 7^{o}C and 65 to 75 per cent relative humidity for 7 days had no effect on composition.

Materials and Methods

MATERIALS AND METHODS

The present study was carried out to develop a suitable formulary for the production of low fat frankfurter (LFF) using pork skin collagen (PSC) gel as a fat replacer and to assess its physico-chemical, nutritional and sensory qualities. The keeping quality of the developed low fat frankfurter under aerobic and vacuum packaging systems at -20 ^{o}C was also investigated.

3.1. DEVELOPMENT OF LOW FAT FRANKFURTERS

3.1.1. Raw Materials

3.1.1.1. Beef, pork and lard

Fresh hot deboned lean beef and pork trimmings were collected from Holstein Friesian cross breed bulls of 2 to 5 years age and Large White Yorkshire pigs weighing 60 to 90 kg humanely slaughtered in the Department of Livestock Products Technology, College of Veterinary & Animal Sciences, Mannuthy. Both were reared under good management conditions. External fat, blood clots, tendons and visible connective tissue, bones and cartilage, if any, were removed from trimmings. The meat samples were randomly analysed for fat content. The beef trimmings with a mean fat content of 1.69 per cent and pork trimmings with 2.27 per cent were assorted, stored at 0-4^{o}C for 48 h for conditioning and then kept frozen at -20 ^{o}C till used for the preparation of frankfurter. Lard was prepared from fresh back fat of the same pigs and stored at -20 ^{o}C till further use.

3.1.1.2. Pork skin collagen gel preparation

Pork skin of sound quality was harvested from ham and belly regions of Large White Yorkshire pigs of 60 to 90 kg. Pork skin was mechanically modified by completely removing the subcutaneous fat and cut into pieces of 2.5 cm × 2.5 cm size and was frozen at -20 ^{o}C till subsequent use for gel preparation. Pork Skin Collagen

(PSC) gel was prepared as outlined by Osburn *et al.* (1997) with slight modification. Appropriate amounts of pork skin and water (1:2 w/v) were moist heated at 80ºC for 45 min instead of 70ºC for 30 min. The gel was cooled to room temperature and homogenized in a domestic mixer (Sumeet, Mumbai). The prepared gel was stored at -20 ºC till the preparation of frankfurter.

3.1.1.3. Non meat ingredients

Good quality freshly ground anise, black pepper and cinnamon were used. Other ingredients were, salt, sugar, sodium nitrite, sodium ascorbate, sodium tripolyphosphate, refined wheat flour, onion, garlic, fresh ginger and ice.

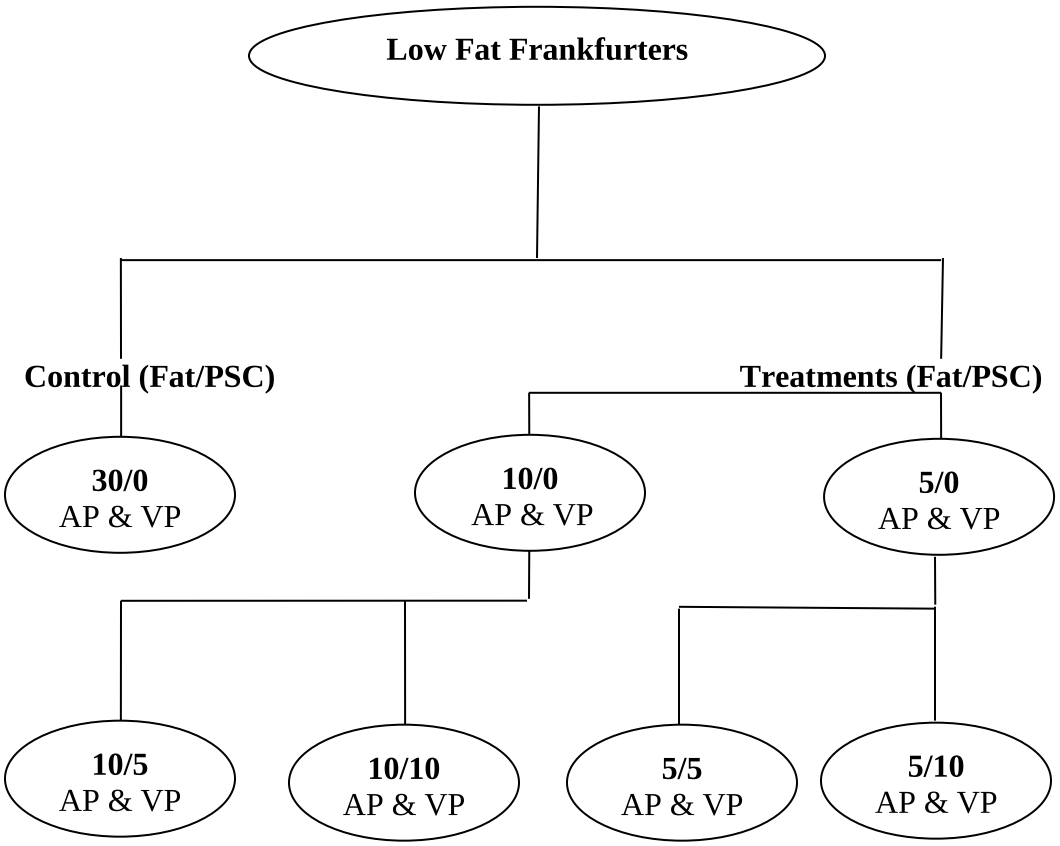
3.2. LOW FAT FRANKFURTER FORMULATION AND MANUFACTURE

Frankfurters were formulated in three different fat levels of 30, 10 and 5 per cent and two levels of PSC, 5 and 10 per cent, according to the scheme presented in Fig 1. Seven formularies for frankfurters with varying levels of fat with or without the addition of PSC are presented in Table 1.

Lard was added in all formulations in such a way that the final fat content of the batter did not exceed 30, 10, and 5, respectively. The low fat formulations with 10 and 5 per cent fat were again divided into three parts each, viz., 1) with out any PSC, 2) with 5 per cent PSC and 3) with 10 per cent PSC. Thus, the one control and six treatment formulations prepared were:

Control	1) 30% full fat with out PSC (30/0)
Treatments	2) 10% low fat with out PSC (10/0)
	3) 5% low fat with out PSC (5/0)
	4) 10% low fat with 5% PSC (10/5)
	5) 10% low fat with 10% PSC (10/10)
	6) 5% low fat with 5% PSC (5/5)
	7) 5% low fat with 10% PSC (5/10)

Fig. 1. Schematic representation of the experiment



30/0: 30% Full Fat; **10/0**: 10% Low Fat; **5/0**: 5% Low Fat; **10/5**: 10% Fat with 5% Pork Skin Collagen (PSC); **10/10**: 10% Fat with 10% PSC; **5/5**: 5% Fat with 5% PSC; **5/10**: 5% Fat with 10% PSC; **PSC:** Pork Skin Collagen

AP: Atmospheric Packaging; **VP**: Vacuum Packaging.

Table 1. Formulary for frankfurters with varying levels of fat with/without pork skin collagen

Ingredients	**Formulations (Fat/PSC)**						
	30/0	**10/0**	**5/0**	**10/5**	**10/10**	**5/5**	**5/10**
Lean Beef trimmings, g	300	300	300	300	300	300	300
Lean Pork trimmings, g	300	300	300	300	300	300	300
Lard, g	290	90	40	90	90	40	40
Ice Flakes, g	110	310	360	310	310	360	360
Salt, g	15	15	15	15	15	15	15
Sugar, g	3	3	3	3	3	3	3
Sodium Nitrite, ppm	100	100	100	100	100	100	100
Sodium Ascorbate, ppm	250	250	250	250	250	250	250
Sodium Tripolyphosphate, ppm	5	5	5	5	5	5	5
Refined Wheat Flour, g	30	30	30	30	30	30	30
Anise, g	1.2	1.2	1.2	1.2	1.2	1.2	1.2
Cinnamon, g	1.2	1.2	1.2	1.2	1.2	1.2	1.2
Black Pepper, g	5	5	5	5	5	5	5
Onion Paste, g	40	40	40	40	40	40	40
Garlic Paste, g	30	30	30	30	30	30	30
Ginger Paste, g	10	10	10	10	10	10	10
PSC, g	-	-	-	50	100	50	100

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC

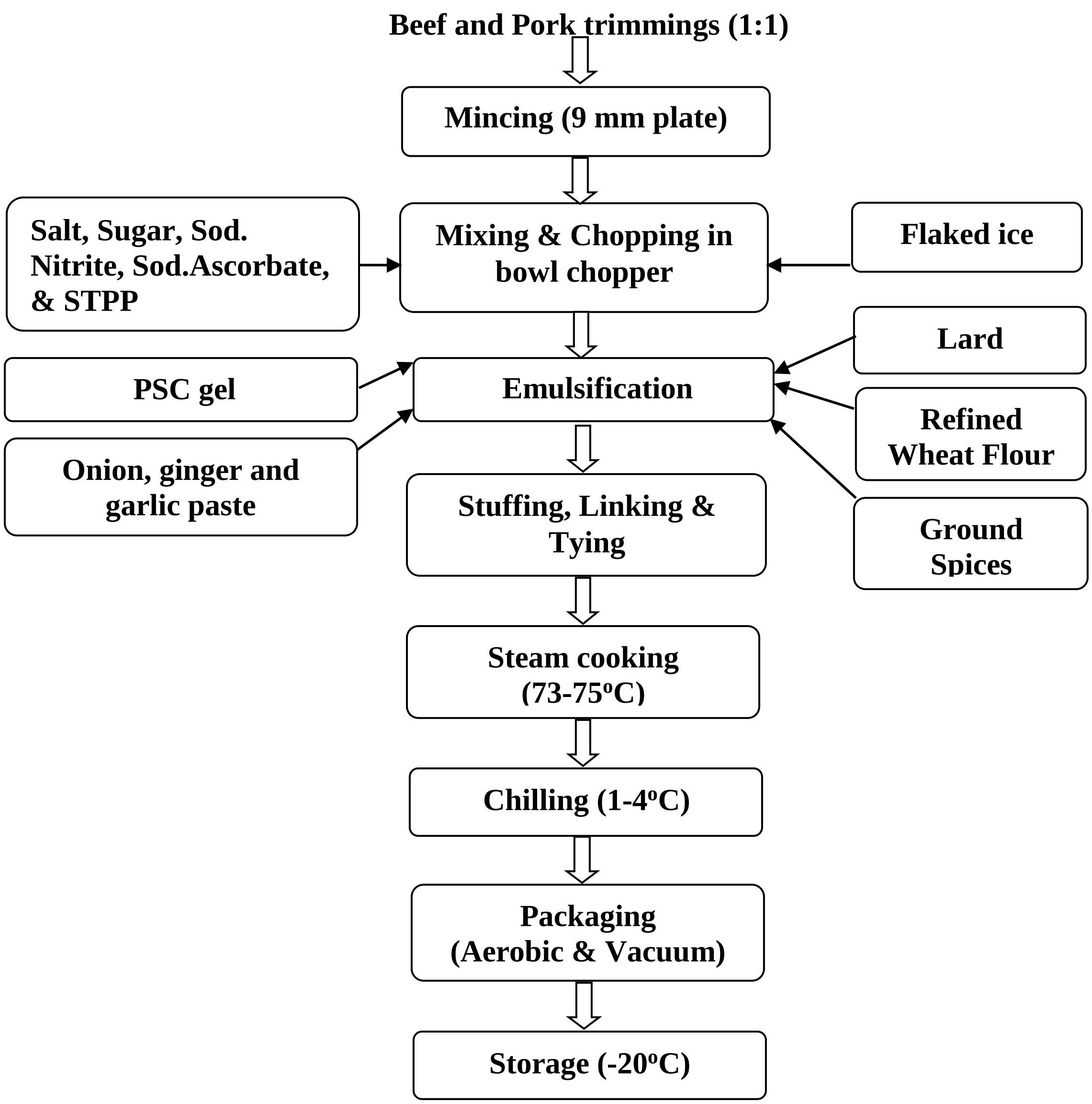
The steps in the preparation of low fat frankfurter are illustrated in the Flow Diagram (Fig.2). Beef and pork trimmings stored at -20^{o}C were thawed under refrigeration and ground separately through a 9 mm grinder plate in a meat mincer (MADO Primus Model MEW 613, Germany). The ground beef and pork trimmings were mixed with salt, sugar, sodium nitrite, sodium ascorbate and sodium tripolyphosphate (STPP) along with ice flakes in a bowl chopper (MADO GARANT, Germany). Lard and PSC were added with the mix and further chopped to make the meat emulsion. The ground spices, refined wheat flour, onion garlic and ginger pastes were also added to the emulsion in the chopper. The temperature of the batter was maintained between 10 – 12^{o}C.

The seven emulsions were stuffed into 18 - 20 mm sheep casing using a manually operated sausage stuffer, twist linked and tied manually. Sausages were steam cooked under atmospheric pressure to a core temperature of 73 - 75^{o}C, measured using a meat thermometer (Oaktank, China) and chilled to 1- 4^{o}C.

All the sausages were packaged under two different systems of packaging, viz., Aerobic Packaging (AP) in high density poly ethylene pouches of 200 µ and Vacuum Packaging (VP) in polyamide/polyethylene pouches (oxygen transmission rate: 208 cc/m^{2}/24hr, water transmission rate: 5g/cc/24hr at 38^{o}C and relative humidity 90 per cent) using a single chamber vacuum packaging machine (Sevana, Kochi, India) and stored at -20^{o}C for further studies.

Various physico-chemical parameters such as pH, emulsion stability (ES), cook yield (CY), cook loss (CL), dimensional shrinkage (DS), water holding capacity (WHC), Warner-Bratzler Shear Force Value (WBSFV), colour by Hunter L^{*}, a^{*}, b^{*} values, proximate composition, nutritional value, purge loss (PL), 2-Thiobarbituric Acid Reactive Substances (TBARS) values and organoleptic qualities were studied on the day of production of frankfurters and shelf life , moisture, protein and fat contents were assessed again on day 15, 30, 45, 60 and 75 of storage at -20^{o}C. The experiment was replicated six times.

Fig. 2. Flow diagram for low fat frankfurter preparation



3.3. QUALITY EVALUATION OF LOW FAT FRANKFURTERS

3.3.1. Physico-chemical and Nutritional Characteristics

3.3.1.1. pH

The pH of the frankfurter mixes from all the treatments and control, before and after cooking was determined using a combined electrode digital pH meter (µ pH system 362, Systronics, India).

3.3.1.2. Emulsion Stability

The emulsion stability was determined by the method of Baliga and Madaiah (1970). Twenty five grams of meat emulsion was taken in a polythene bag and heated in a thermostatically controlled water bath at 80^{o}C for 20 min. Then the exudates were drained and the cooked mass was blotted with a tissue paper, cooled and weighed. The percentage of cooked mass was expressed as emulsion stability.

3.3.1.3. Cooking Characteristics

Weights of frankfurters before and after cooking from all formulations were recorded. The cook yield and cook loss were calculated according to Murphy *et al.* (1975). The diameter of the sausage links was measured using Vernier Calipers in mm and the mean of five readings of the outer diameter of five links were taken before and after cooking.

Cook yield percentage (CY) = (weight of frankfurter after cooking / weight of frankfurter before cooking) x 100.

Cook loss percentage (CL) = 100 - cook yield.

Dimensional Shrinkage, percentage (DS) = (Diameter before cooking- Diameter after cooking / Diameter before cooking) x 100

3.3.1.4. Water Holding Capacity

Samples of 10 g from each sausage formulation were placed in glass jars and heated to 90^{o}C for 10 min in a thermostatically controlled water bath. Samples were carefully removed from the jars and cooled to 4^{o}C for 20 min, wrapped in fine cheese cloth and placed in 50 ml polycarbonate centrifuge tubes packed with cotton wool at the bottom. After centrifugation at 9000 x g Relative Centrifugal Force (RCF) for 10 min, samples were weighed again and the WHC (%) was calculated as follows (Liangi and Chen, 1991). WHC (%) = 1- (B-A/M) x 100

Where,

B = Weight of sample before heating.

A = Weight of sample after heating and centrifuging.

M = Total moisture content of the sample.

3.3.1.5. Warner-Bratzler Shear Force

The WBSF values of frankfurters from all formulations were recorded as per the method of Trindade *et al.* (2005) with a slight modification. Frankfurters after cooling over night to 1-4^{o}C were cut into 20 mm high and 18 mm dia cylinders instead of 13 mm dia and compressed in their round surface using Warner-Bratzler shear having a cross head speed of 200mm/min attached to a Universal Testing Machine (Shimadzu Texture Analyser Model EZ Shimadzu Corporation, Kyoto, Japan.). For each sample, five observations were recorded to obtain the mean value of shear force in kgf.

3.3.1.6. Colour

Colour of the sausage samples was determined objectively using Hunter Lab mini scan XE Plus Spectrophotometer (Hunter Lab, Virginia, USA) with diffuse illumination. The instrument was set to measure Hunter L^*, a^* and b^* using illuminant 45/0 and 10^{o} standard observer with an aperture size of 2.54 cm. It was calibrated using black and white tiles and colorimeter score recorded with ‘L’ of black equals 0 and ‘L’ of white equals 100, ‘a’ of lower numbers equals more green (less red), higher numbers equals more red (less green) and ‘b’ of lower numbers equals more blue (less yellow), higher numbers equals yellow (less blue) (Page *et al.*, 2001). The colour coordinates L^* (lightness), a^* (redness), b^* (yellowness) of the samples was measured thrice and mean values were taken.

3.3.1.7. Proximate Composition

The proximate composition of beef and pork trimmings, combination of these two in 1:1 proportion, PSC gel and each formulation of uncooked and cooked frankfurter was determined by the standard procedure of AOAC (1990) and the values were expressed in g per 100g of sample on as is basis. Analyses were conducted in duplicate. Moisture was determined by weight loss after 16 h drying in a hot air oven at 105^{o}C.

The fat content was determined in moisture free samples by an ether extraction procedure in an Automatic Solvent Extraction System (SOX plus, Model SCS 6, Pelican Equipments, Chennai, India). Moisture free fat free samples were used to estimate the protein and ash content. The protein content was determined by Block Digestion Method (KEL Plus, Model KES 6L, Pelican Equipments, Chennai, India.). Ash was determined by weight loss after 2½ h drying in a muffle furnace at 600^{o}C. The amount of carbohydrate was calculated as 100 minus sum of the percentage of moisture, protein, fat and ash. Moisture, fat and protein contents of frankfurter were determined on day 0, 15, 30, 45, 60 and 75 at -20^{o}C storage. The proximate composition was expressed in as-is-basis.

Effect of cooking on proximate composition of LFF of different formulations was studied by comparing the same parameters obtained before and after cooking.

3.3.1.8. Nutritional Value

Calorific Value

Total calories and calories from fat, protein and carbohydrate of each treatment of frankfurter were determined as per FAO (2002).

Calories from fat = fat per cent x 9

Calories from protein = protein per cent x 4

Calories from carbohydrate = carbohydrate per cent x 4

Total calories = sum of calories from fat, protein and carbohydrate.

Per cent Recommended Daily Allowance (RDA) for calories from fat, protein, and carbohydrate was calculated based on a 2200 kcal diet (ICMR, 1990).

Per Cent Daily Value of Protein in frankfurters

Per cent daily value of protein of different formulations of LFF was calculated and expressed as percentage of RDA using the following formula.

Per cent daily value of protein in frankfurter = per cent protein in frankfurter/ RDA of the protein. RDA of protein was taken as 60g (ICMR, 1990).

3.3.1.9. Purge Loss

Determination of purge (moisture) loss consisted of weighing each type of frankfurter sausage from frozen storage on day 15, 30, 45, 60 and 75. Two sausage links were taken out from the package and carefully blotted with tissue paper to eliminate any liquid on the surface of links and weighed. The initial weight of the links was measured in the beginning of the experiment. The difference in weight expressed as percentage of initial weight was reported as purge loss.

3.3.1.10. 2-Thiobarbituric Acid Reactive Substances Value

TBARS value in frankfurters was determined by the extraction method of Witte *et al.* (1970) with a slight modification. The extraction supernatant was centrifuged (Eltek Research Centrifuge TC 8100) at 6000 rpm for 5 min instead of filtration. Absorbance was measured at 530nm (Systronics-119, UV-Visible Spectrophotometer, Ahmedabad, India) against blank containing 5ml of distilled water and 5 ml TBA reagent. TBARS value, expressed as mg malonaldehyde per kg of frankfurter was calculated by multiplying the absorbance with a factor of 5.2. The TBARS value of frankfurters were measured on day 0, 15, 30, 45, 60 and 75 of storage at -20^{o}C under aerobic and vacuum packaging.

3.3.2. Sensory Evaluation

The sensory panel evaluation of the organoleptic qualities of LFF was conducted by a semi trained panel consisted of seven panelists using the score card as in Table 2 (AMSA, 1983).

The frankfurters after refrigerated thawing were fried in refined sun flower oil in a frying pan for 3 minutes and served hot. Panelists were then asked to evaluate the frankfurters and record a score for the samples on an eight point Hedonic scale for appearance and colour, flavour, texture, saltiness, juiciness, mouth coating and overall acceptability. The sensory evaluation was conducted on day 0, 15, 30, 45, 60, and 75 of storage at -20^{o}C, respectively.

3.4. COST OF PRODUCTION

The cost of production of the different frankfurter formulations was calculated from the cost of meat, other ingredients and processing.

3.5. STATISTICAL ANALYSIS

Data obtained were analysed by one way Analysis of Variance, Repeated Measures Analysis of Variance, Student's - *t* test, Kruskal-Wallis test and Mann-Whitney- U test using SPSS soft ware (Snedecor and Cochran, 1994).

Table 2. Score card for the organoleptic evaluation of low fat frankfurters

Panelist: ………………Date:……………….Expt:…………Session No:…………..

Attributes	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6	Sample 7	Sample 8	Sample 9	Sample 10	Sample 11	Sample 12	Sample 13	Sample 14
Appearance & Colour														
Flavour														
Texture														
Saltiness														
Juiciness														
Mouth Coating														
Overall Acceptability														

CODES

Appearance and Colour

8 Excellent
7 Very good
6 Good
5 Fair
4 Slightly poor
3 Moderately poor
2 Very poor
1 Extremely poor

Flavour

8 Extremely intense
7 Very intense
6 Moderately intense
5 Slightly intense
4 Slightly bland
3 Moderately bland
2 Very bland
1 Extremely bland

Texture

8 Extremely desirable
7 Very desirable
6 Moderately desirable
5 Slightly desirable
4 Slightly undesirable
3 Moderately undesirable
2 Very undesirable
1 Extremely undesirable

Saltiness

8 Extremely desirable
7 Very desirable
6 Moderately desirable
5 Slightly desirable
4 Slightly undesirable
3 Moderately undesirable
2 Very undesirable
1 Extremely undesirable

Juiciness

8 Extremely juicy
7 Very juicy
6 Moderately juicy
5 Slightly juicy
4 Slightly dry
3 Moderately dry
2 Very dry
1 Extremely dry

Mouth Coating

8 None
7 Practically nil
6 Traces
5 Slight
4 Moderate
3 Slightly abundant
2 Moderately abundant
1 Abundant

Overall Acceptability

8 Extremely acceptable
7 Very acceptable
6 Moderately acceptable
5 Slightly acceptable
4 Slightly unacceptable
3 Moderately unacceptable
2 Very unacceptable
1 Extremely unacceptable

Comments if any:

Signature of the Panelist

Results

RESULTS

Frankfurters were formulated with three different levels of fat at 30, 10 and 5 per cent and two levels of the fat replacer Pork Skin Collagen (PSC) gel at 5 and 10 per cent with a view to developing the most suitable low fat frankfurter. One full fat control with out PSC (30/0), and six treatments, viz., 10 and 5 per cent low fat frankfurters with out PSC (10/0, 5/0), 10 and 5 per cent low fat with 5 per cent PSC (10/5, 5/5), 10 and 5 per cent low fat with 10 per cent PSC (10/10, 5/10) were prepared. Various physico-chemical characteristics, proximate composition, nutritive value, organoleptic qualities and the shelf life studies by assessing the purge loss, 2-TBARS value, organoleptic qualities and moisture, protein and fat contents of frankfurters were studied again on d 15, 30, 45, 60 and 75 of storage at -20^{o}C under aerobic packaging (AP) and vacuum packaging (VP) systems. The results obtained are narrated and supported by Tables and Figures in this chapter.

4.1. PHYSICAL CHARACTERISTICS OF FRANKFURTERS

4.1.1. pH

The pH values of different formulations of LFF batter before cooking and of the products after cooking are shown in Table 3 and the trend in pH on cooking is presented in Fig. 3.

The pH of different formulations of LFF, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 before cooking were 6.12 ± 0.05, 6.20 ± 0.03, 6.13 ± 0.04, 6.26 ± 0.02, 6.31 ± 0.04, 6.28 ± 0.04 and 6.38 ± 0.02, respectively. The pH of frankfurters after cooking were 6.29 ± 0.02, 6.40 ± 0.05, 6.27 ± 0.02, 6.46 ± 0.03, 6.49 ± 0.03, 6.47 ± 0.04and 6.55 ± 0.01, respectively.

Table 3. pH, cooking characteristics ES, WHC and WBSFV of different formulations of LFF

Parameters	Formulations (Fat/PSC)						
	30/0	**10/0**	**5/0**	**10/5**	**10/10**	**5/5**	**5/10**
pH Batter	6.12^{a} ± 0.05	6.20^{ab} ± 0.03	6.13^{a} ± 0.04	6.26^{bc} ± 0.02	6.31^{cd} ± 0.04	6.28^{bc} ± 0.04	6.38^{d} ±0.02
pH Product	6.29^{a} ± 0.02	6.40^{b} ± 0.05	6.27^{a} ± 0.02	6.46^{bc} ± 0.03	6.49^{bc} ± 0.03	6.47^{bc} ± 0.04	6.55^{c} ± 0.01
ES (%)	77.48^{a} ± 0.28	87.22^{b} ± 0.18	86.61^{b} ± 0.36	87.27^{b} ± 0.2	87.40^{b} ± 0.19	86.66^{b} ± 0.39	86.74^{b} ± 0.42
CY (%)	90.56^{a} ± 0.56	96.05^{b} ± 0.10	95.51^{b} ± 0.12	96.13^{b} ± 0.09	96.28^{b} ± 0.08	95.56^{b} ± 0.12	95.56^{b} ± 0.16
CL (%)	9.44^{a} ± 0.56	3.95^{b} ± 0.10	4.42^{b} ± 0.11	3.87^{b} ± 0.09	3.73^{b} ± 0.08	4.44^{b} ± 0.12	4.32^{b} ± 0.15
DS (%)	3.95^{a} ± 0.03	1.88^{b} ± 0.02	2.09^{b} ± 0.13	1.86^{b} ± 0.02	1.83^{b} ± 0.02	2.06^{b} ± 0.13	2.04^{b} ± 0.12
WHC (%)	96.34^{f} ± 0.18	94.84^{bc} ± 0.03	93.72^{a} ± 0.01	95.31^{de} ± 0.05	95.57^{e} ± 0.09	94.56^{b} ± 0.20	95.00^{cd} ± 0.24
WBSFV (kgf)	0.51^{a} ± 0.02	1.58^{b} ± 0.01	1.82^{c} ± 0.01	1.41^{d} ± 0.02	1.12^{e} ± 0.01	1.20^{f} ± 0.01	1.14^{e} ± 0.02

Means bearing same alphabets in the same row do not indicate significant difference (P< 0.05).

PSC- Pork Skin Collagen;**30/0 -** 30% full fat; **10/0 -**10% low fat; **5/0 -** 5% low fat; **10/5 -** 10% fat with 5% PSC; **10/10 -**10% fat with 10% PSC; **5/5** 5% fat with 5% PSC; **5/10 -**5% fat with 10% PSC; **ES** –Emulsion Stability; **CY** – Cook Yield; **CL-**Cook Loss; **DS-**Dimensional Shrinkage; **WHC-** Water Holding Capacity; **WBSFV-** Warner-Bratzler Shear Force Value. **LFF-** Low fat frankfurter.

Table 4. Hunter Colour values of different formulations of LFF

Parameter	Formulations(Fat/PSC)						
	30/0	**10/0**	**5/0**	**10/5**	**10/10**	**5/5**	**5/10**
L^*	62.99^{a} ±0.01	61.43^{b} ±0.13	56.04^{c} ±0.19	60.80^{d} ±0.05	62.99^{a} ±0.23	57.24^{e} ±0.02	57.85^{f} ±0.16
a^*	5.62^{a} ± 0.02	5.80^{c} ± 0.01	5.93^{e} ± 0.01	5.83^{cd} ± 0.02	5.70^{b} ± 0.03	5.90^{e} ± 0.22	5.88^{de} ± 0.02
b^*	18.18^{a} ± 0.04	16.05^{b} ± 0.05	14.85^{c} ± 0.11	15.34^{d} ± 0.04	17.51^{e} ± 0.11	15.06^{f} ± 0.01	15.52^{d} ± 0.05

Means bearing same alphabets in the same row do not indicate significant difference ($P < 0.05$).

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** – 10% fat with 10% PSC; 5/**5** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-**Low fat frankfurter. **L^*-** Lightness; **a^*-** redness; **b^*-** yellowness

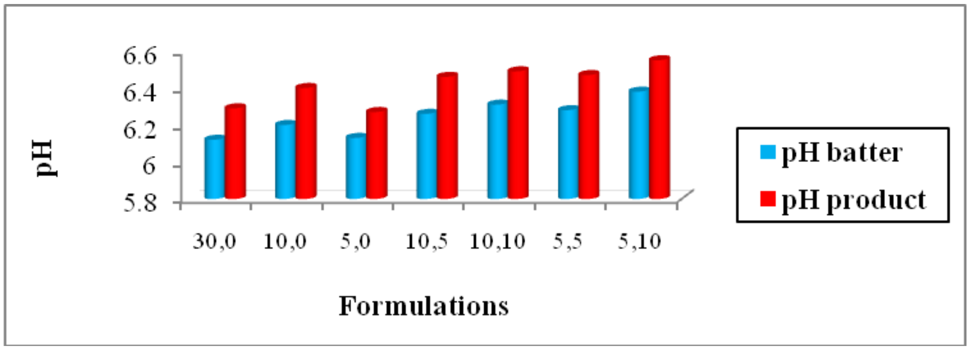


Fig. 3. pH of uncooked and cooked different formulations of LFF

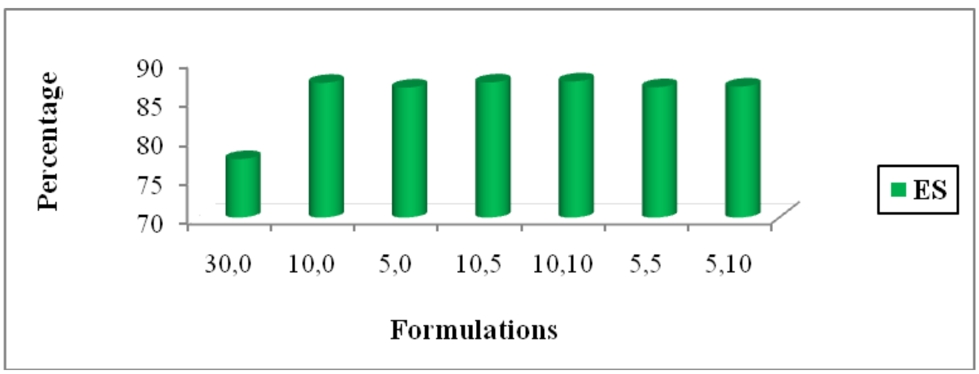


Fig. 4. Emulsion stability of different formulations of LFF

30,0 - 30% full fat; **10,0** - 10% low fat; **5,0** - 5% low fat;
10,5 - 10% fat with 5% PSC; **10,10** - 10% fat with 10% PSC;
5,5 - 5% fat with 5% PSC; **5,10** - 5% fat with 10% PSC
PSC- Pork Skin Collagen; **ES**- Emulsion stability; **LFF**-Low fat frankfurter

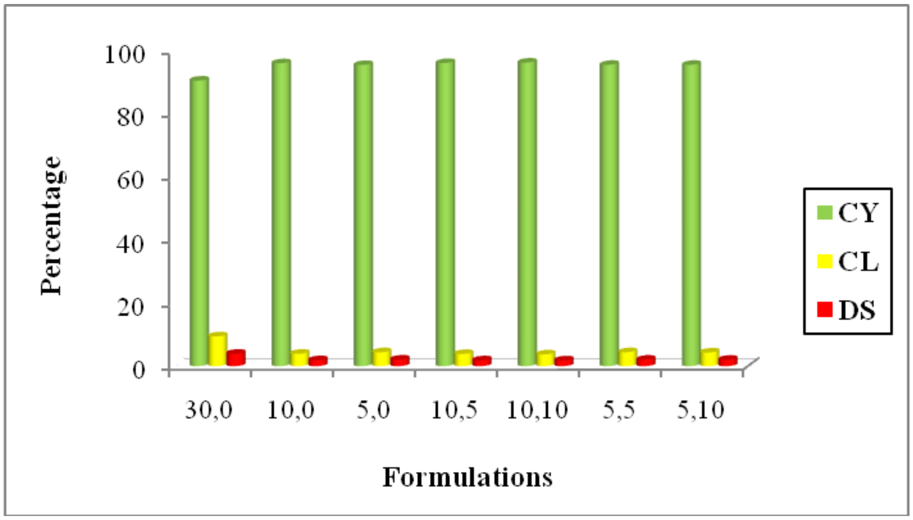


Fig. 5. Cooking characteristics of different formulations of LFF

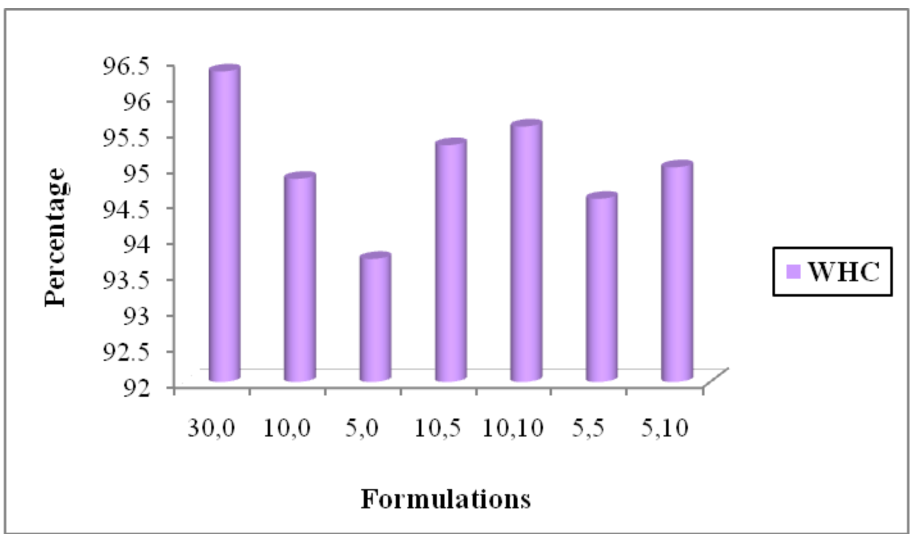


Fig. 6. Water Holding Capacity of different formulations of LFF

30,0-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10, 10** -10% fat with 10% PSC; **5,5** 5% fat with 5% PSC; **5, 10** -5% fat with 10% PSC; **PSC**- Pork Skin Collagen; **CY-** Cook Yield; CL- Cooks Loss; **DS**-Dimensional Shrinkage; **WHC**-Water Holding Capacity; **LFF-**Low fat frankfurter

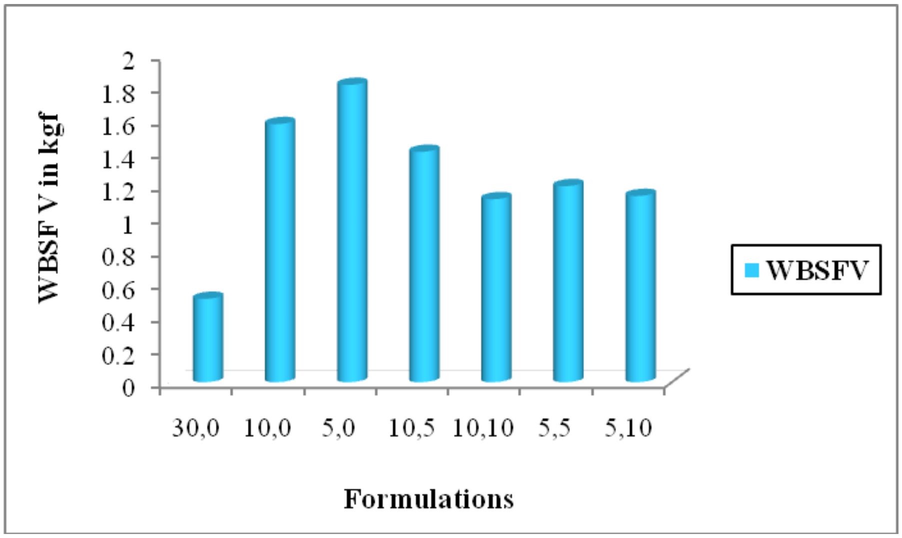


Fig. 7. WBSFV of different formulations of LFF

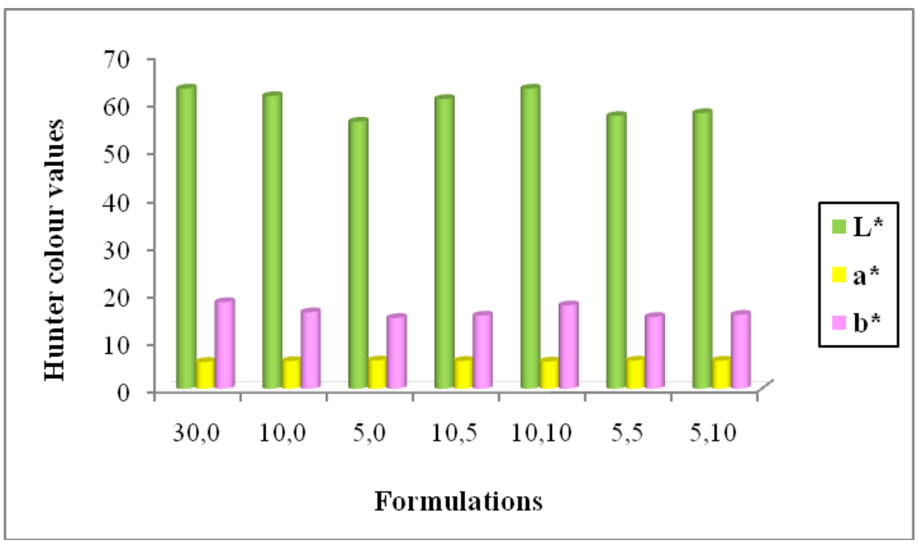


Fig. 8. Hunter L*a*b* colour values of different formulations of LFF

30,0-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10, 10** -10% fat with 10% PSC; **5, 5** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC**- Pork Skin Collagen; **WBSFV**-Warner-Bratzler Shear Force Value; **L***- Lightness; **a***- redness; **b***- yellowness; **LFF**-Low fat frankfurter

The pH of the batter 5/10 and 10/10 were significantly higher ($P < 0.05$) than that of 30/0, 10/0, and 5/0. There was no significant difference ($P > 0.05$) in the pH value of the batter of 30/0, 10/0, and 5/0. Similarly, no significant difference ($P > 0.05$) was noted in the pH values between 10/5 and 5/5.

The pH of all frankfurter formulations was significantly higher ($P < 0.05$) than their corresponding uncooked batter. Among the products there was no significant difference ($P > 0.05$) between 30/0 and 5/0 and between 10/5, 10/10, 5/5 and 5/10. Formulations 10/10 and 5/10 showed significantly higher ($P < 0.05$) pH values than the full fat control 30/0, 10/0 and 5/0. pH of the formulations with PSC were significantly higher than those without PSC.

4.1.2. Emulsion Stability

The Emulsion Stability (ES) of different formulations are shown in Table 3 and the trend is illustrated in Fig.4.

The percentage ES of 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 were 77.48 ± 0.28, 87.22 ± 0.18, 86.61 ± 0.36, 87.27 ± 0.2, 87.40 ± 0.19, 86.66 ± 0.39, 86.74 ± 0.42, respectively.

The formulations 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 had significantly higher ($P < 0.05$) emulsion stability than the full fat control. Among treatments no significant difference ($P > 0.05$) was noticed. All formulations of LFF with or without PSC showed significantly higher ES than the full fat frankfurter.

4.1.3. Cooking Characteristics

The percentages of cook yield (CY), cook loss (CL), and dimensional shrinkage (DS) are given in Table 3 and depicted in Fig.5.

4.1.3.1. Cook Yield and Cook Loss

The percentage cook yield was 90.56 ± 0.56, 96.05 ± 0.10, 95.51 ± 0.12, 96.13 ± 0.09, 96.28 ± 0.08, 95.56 ± 0.12, 95.56 ± 0.16 for the formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, 5/10, respectively. The cook loss percentages of these formulations were 9.44 ± 0.56, 3.95 ± 0.10, 4.42 ± 0.11, 3.87 ± 0.09, 3.73 ± 0.08, 4.44 ± 0.12, 4.32 ± 0.15, respectively. The cook yield of 10/10 was significantly the highest ($P < 0.05$) of all formulations compared to full fat control though, there was no significant difference ($P > 0.05$) between treatments. The cook loss percentage was maximum in full fat control compared to all treatments.

4.1.3.2. Dimensional Shrinkage

The percentage DS of formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 were 3.95 ± 0.03, 1.88 ± 0.02, 2.09 ± 0.13, 1.86 ± 0.02, 1.83 ± 0.02, 2.06 ± 0.13, 2.04 ± 0.12, respectively.

All the treated formulations, viz., 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 showed significantly lower ($P < 0.05$) DS compared to full fat control, 30/0. Formulation 10/10 had the lowest DS. However, there was no significant difference ($P > 0.05$) between treated formulations.

4.1.4. Water Holding Capacity

The Water Holding Capacity (WHC), expressed as percentage is given in Table 3 and the trend is shown in Fig.6.

The WHC of 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 were 96.34 ± 0.18, 94.84 ± 0.03, 93.72 ± 0.01, 95.31 ± 0.05, 95.57 ± 0.09, 94.56 ± 0.20, 95.00 ± 0.24, respectively.

Formulations 10/5 and 10/10 had significantly higher ($P < 0.05$) WHC values than all other frankfurter formulations. Full fat frankfurter had significantly the highest ($P < 0.05$) WHC of all formulations and gradually reduced with reduction in fat content. Frankfurter 5/0 had significantly the lowest ($P < 0.05$) WHC of all formulations but the capacity significantly increased on adding PSC.

4.1.5. Warner-Bratzler Shear Force Value

The WBSFV in kgf of different formulations of LFF are given in the Table 3 and the variations in the shear force are presented in Fig. 7.

The WBSFV of different formulations of LFF such as 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 were 0.51 ± 0.02, 1.58 ± 0.01, 1.82 ± 0.01, 1.41 ± 0.02, 1.12 ± 0.01, 1.20 ± 0.01, 1.14 ± 0.02, respectively.

The shear force of 5/0 formulation showed significantly the highest ($P < 0.05$) value of all formulations. On the contrary, significantly the lowest ($P < 0.05$) shear force was observed for full fat control among all formulations. Among treatments, 10/10 formulation had significantly the lowest ($P < 0.05$) shear force.

4.1.6. Colour

The Hunter L* a* b^* values are given in the Table 4 and the trend of each is illustrated in Fig. 8.

The L* values were 62.99 ± 0.01, 61.43 ± 0.03, 56.04 ± 0.19, 60.80 ± 0.05, 62.99 ± 0.23, 57.24 ± 0.02, 57.85 ± 0.16, respectively for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10.

Among treatment formulations 10/10 had significantly the highest ($P < 0.05$)

L* value and it was not significantly different ($P > 0.05$) from full fat frankfurter. Formulation 5/0 had significantly the lowest ($P < 0.05$) L* value among all.

The a* values for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 were 5.62 ± 0.02, 5.80 ± 0.01, 5.93 ± 0.01, 5.83 ±0.02, 5.70 ± 0.03, 5.90 ± 0.22, 5.88 ± 0.02, respectively.

Formulation 5/0 had significantly the highest ($P < 0.05$) a* value among all formulations. The full fat control 30/0 had significantly the lowest ($P < 0.05$) a* value of all formulations. Among treated formulations 10/10 had significantly lower ($P < 0.05$) a* value than 10/0, 5/0, 10/5, 5/5. There was no significant difference ($P > 0.05$) noticed between 10/0, 10/5 and between 5/0, 5/5.

The b* values were 18.18 ± 0.04, 16.05 ± 0.05, 14.85 ± 0.11, 15.34 ±0.04, 17.51 ± 0.11, 15.06 ± 0.01, 15.52 ± 0.05, respectively for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10.

Among the treated formulations 10/10 had significantly the highest ($P < 0.05$) b* value and 5/0 had significantly the lowest ($P < 0.05$) b* value.

4.2. PROXIMATE COMPOSITION

4.2.1. Lean Beef and Pork Trimmings, Their Combinations and Pork Skin Collagen

The percentage proximate composition of Lean Beef Trimmings (LBT), Lean Pork Trimmings (LPT), their 1:1 mix and PSC are presented in Table 5 and the comparative profile is depicted in Fig. 9.

The percentage of moisture in LBT, LPT, their 1:1 mix and PSC were 77.30 ±

0.30, 75.90 ± 0.27, 76.67 ± 0.57, 72.67 ± 0.13, respectively. The protein content of the same were 18.99 ± 0.43, 19.37 ± 0.18, 19.13 ± 0.63, 25.77 ± 0.22, respectively. The fat percentages were 1.69 ± 0.18, 2.27 ± 0.32, 1.98 ± 0.23, 0.60 ± 0.11, respectively. The carbohydrate content were 0.98 ± 0.07, 1.07 ± 0.09, 1.01 ± 0.08, 0.38 ± 0.02, respectively. Similarly, the ash content were 1.04 ± 0.04, 1.39 ± 0.04, 1.21 ± 0.08, 0.58 ± 0.02, respectively.

Table 5. Proximate composition of different meat ingredients

Parameters %	Lean Beef trimmings	Lean Pork trimmings	Beef : Pork (1:1)	Pork skin collagen
Moisture	77.30^{a} ± 0.30	75.90^{b} ± 0.27	76.67^{ab} ± 0.57	72.67^{c} ± 0.13
Protein	18.99^{a} ± 0.43	19.37^{a} ± 0.18	19.13^{a} ± 0.63	25.77^{b} ± 0.22
Fat	1.69^{a} ± 0.18	2.27^{a} ± 0.32	1.98^{a} ± 0.23	0.60^{b} ± 0.11
Carbohydrate	0.98^{a} ± 0.07	1.07^{a} ± 0.09	1.01^{a} ± 0.08	0.38^{b} ± 0.02
Ash	1.04^{a} ±0.04	1.39^{b} ± 0.04	1.21^{c} ± 0.08	0.58^{d} ± 0.02

Means bearing same alphabets in the same row do not indicate significant difference (P< 0.05).

The LBT contained significantly higher (P< 0.05) percentage of moisture than LPT and PSC. The moisture content of meat mix did not differ significantly from that of either LBT or LPT. The moisture content of PSC was significantly the lowest (P< 0.05) among all. The protein and fat content of LBT, LPT and their mix were not significantly different (P> 0.05). The protein content of PSC was significantly the highest (P< 0.05) and fat content was significantly the lowest (P< 0.05) than that of LBT, LPT and their 1:1 mix. The carbohydrate content of PSC was significantly the lowest (P< 0.05). There was significant difference in the ash content of all of them with significantly higher (P< 0.05) content in LPT and significantly lower (P< 0.05) content in PSC.

4.2.2. Low Fat Frankfurters

4.2.2.1. Uncooked Frankfurter Batter

The proximate composition in 100g of frankfurter batter of all seven formulations before cooking is given in Table 6 and illustrated in Fig. 10.

The percentage of moisture in the uncooked batter of 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, 5/10 were 57.65 ± 0.25, 75.08 ± 0.35, 79.88 ± 0.33, 74.32 ± 0.33, 72.90 ± 0.35, 79.66 ± 0.37, 77.03 ± 0.40, respectively. The protein content was 8.90 ± 0.29, 10.40 ± 0.28, 11.30 ± 0.25, 11.87 ± 0.20, 12.31 ± 0.17, 11.53 ± 0.22, 11.90 ± 0.14, respectively. The fat percentages were, 30.01 ± 0.23, 10.05 ± 0.20, 5.10 ± 0.16, 10.08 ± 0.10, 10.10 ± 0.15, 5.05 ± 0.08, 5.10 ± 0.11, respectively. The carbohydrate contents were 2.43 ± 0.32, 3.49 ± 0.44, 2.84 ± 0.16, 2.82 ± 0.29, 3.73 ± 0.53, 2.76 ± 0.52, 4.95 ± 0.51, respectively. The ash contents were 1.01 ± 0.04, 0.98 ± 0.02, 0.88 ± 0.04, 0.91 ± 0.07, 0.96 ± 0.03, 1.00 ± 0.03, 1.02 ± 0.05, respectively.

The proximate composition revealed significantly the highest (P< 0.05) fat content and significantly lowest moisture content in full fat control. The formulation 10/0 and 5/0 had significantly higher (P< 0.05) moisture content than 10/10 and 5/10 respectively. The protein content in full fat frankfurter had the significantly lowest (P< 0.05) level than all other treatments. The frankfurter formulation 10/10 showed significantly the highest (P< 0.05) protein content of all treatments. There was no significant difference (P> 0.05) noticed in the fat content of 10/0, 10/5 and 10/10 but they were significantly higher (P< 0.05) in their fat content than 5/0, 5/5 and 5/10 which were showing no significant difference (P> 0.05) among them. The amount of carbohydrate in the 5/10 was significantly the highest (P< 0.05) among all.

Table 6. Proximate composition of different formulations of uncooked LFF

Formulations (Fat/PSC)	Parameters, %				
	Moisture	Protein	Fat	Carbohydrate	Ash
30/0	57.65[a] ± 0.25	8.90[a] ± 0.29	30.01[c] ± 0.23	2.43[a] ± 0.32	1.01 ± 0.04
10/0	75.08[c] ± 0.35	10.40[b] ± 0.28	10.05[b] ± 0.20	3.49[a] ± 0.44	0.98 ± 0.02
5/0	79.88[e] ± 0.33	11.30[c] ± 0.25	5.10[a] ± 0.16	2.84[a] ± 0.16	0.88 ± 0.04
10/5	74.32[c] ± 0.33	11.87[cd] ±0.20	10.08[b] ± 0.10	2.82[a] ± 0.29	0.91 ± 0.07
10/10	72.90[b] ± 0.35	12.31[d] ± 0.17	10.10[b] ± 0.15	3.73[a] ± 0.53	0.96 ± 0.03
5/5	79.66[e] ± 0.37	11.53[c] ± 0.22	5.05[a] ± 0.08	2.76[a] ± 0.52	1.00 ± 0.03
5/10	77.03[d] ± 0.40	11.90[cd] ± 0.14	5.10[a] ± 0.11	4.95[b] ± 0.51	1.02 ± 0.05

Means bearing same alphabets in the same column do not indicate significant difference ($P < 0.05$)

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** - 10% fat with 10% PSC; **5/5** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low Fat Frankfurter

4.2.2.2. Cooked Frankfurters

The proximate composition of cooked frankfurters of seven different formulations is presented in the Table 7 and Fig. 10.

The percentage moisture in 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, 5/10 were 56.80 ± 0.10, 72.43 ± 0.44, 76.54 ± 0.20, 72.32 ± 0.22, 71.30 ± 0.36, 76.66 ± 0.21, 74.53 ± 0.17, respectively. The protein content were 12.15 ± 0.35, 12.46 ± 0.28, 12.52 ± 0.28, 13.01 ± 0.13, 13.09 ± 0.10, 12.77 ± 0.32, 12.85 ± 0.14, respectively. The fat contents were 25.79 ± 0.24, 9.85 ± 0.33, 5.30 ± 0.13, 9.57 ± 0.22, 9.82 ± 0.25, 5.38 ± 0.14, 5.79 ± 0.14, respectively. The percentage of carbohydrate were 4.18 ± 0.22, 4.23 ±

0.21, 4.52 ± 0.22, 4.17 ± 0.14, 4.81 ± 0.10, 4.12 ± 0.28, 5.70 ± 0.19, respectively. The ash content were 1.08 ± 0.03, 1.03 ± 0.05, 1.12 ± 0.06, 0.93 ± 0.02, 0.98 ± 0.03, 1.07 ± 0.04, 1.13 ± 0.05, respectively.

Table 7. Proximate composition of different formulations of cooked LFF

Formulations (Fat/PSC)	**Parameters %**				
	Moisture	Protein	Fat	Carbohydrate	Ash
30/0	56.80^{a} ± 0.10	12.15^{a} ± 0.35	25.79^{c} ± 0.24	4.18^{ab} ± 0.22	1.08^{bc} ± 0.03
10/0	72.43^{c} ± 0.44	12.46^{ab} ± 0.28	9.85^{b} ± 0.33	4.23^{ab} ± 0.21	1.03^{abc} ± 0.05
5/0	76.54^{e} ± 0.20	12.52^{ab} ± 0.28	5.30^{a} ± 0.13	4.52^{ab} ± 0.22	1.12^{c} ± 0.06
10/5	72.32^{c} ± 0.22	13.01^{b} ± 0.13	9.57^{b} ± 0.22	4.17^{a} ± 0.14	0.93^{a} ± 0.02
10/10	71.30^{b} ± 0.36	13.09^{b} ± 0.10	9.82^{b} ± 0.25	4.81^{b} ± 0.10	0.98^{ab} ± 0.03
5/5	76.66^{e} ± 0.21	12.77^{ab} ± 0.32	5.38^{a} ± 0.14	4.12^{a} ± 0.28	1.07^{bc} ± 0.04
5/10	74.53^{d} ± 0.17	12.85^{ab} ± 0.14	5.79^{a} ± 0.14	5.70^{c} ± 0.19	1.13^{c} ± 0.05

Means bearing same alphabets in the same column do not indicate significant difference (P< 0.05).

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** - 10% fat with 10% PSC; **5/5** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

The mean moisture content was the lowest and highest (P< 0.05) in full fat control and 5/5 formulations, respectively. The moisture content of 10/0 and 10/5 formulations did not differ significantly (P> 0.05), but they had significantly lower (P< 0.05) moisture content than 5/0, 5/5 and 5/10 formulations. Similarly, moisture content of 5/0 and 5/5 formulations also not differ significantly (P> 0.05) among them, but they had significantly higher (P< 0.05) moisture level than other formulations. The protein content of formulations 30/0, 10/0, 5/0, 5/5, 5/10 did not

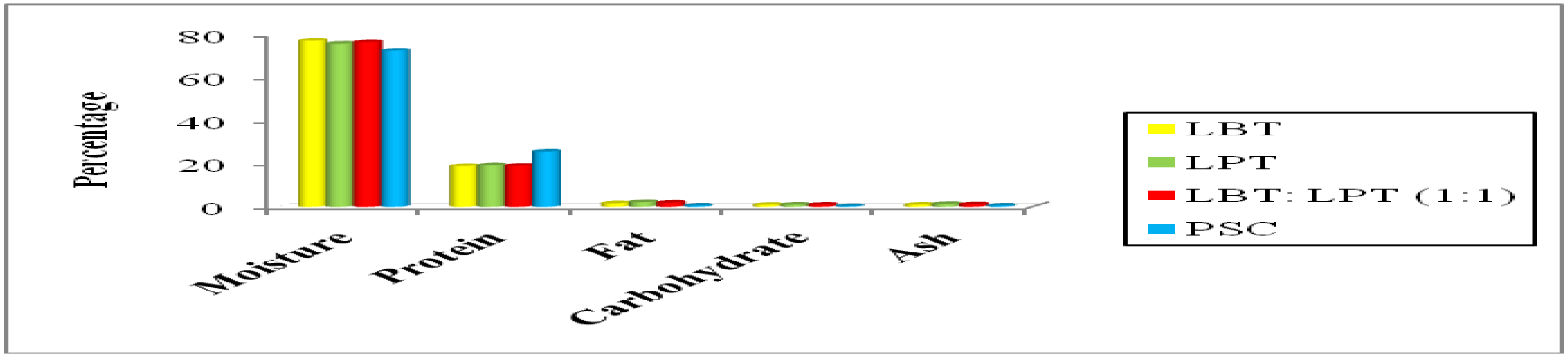


Fig. 9. Proximate composition of of meat ingredients

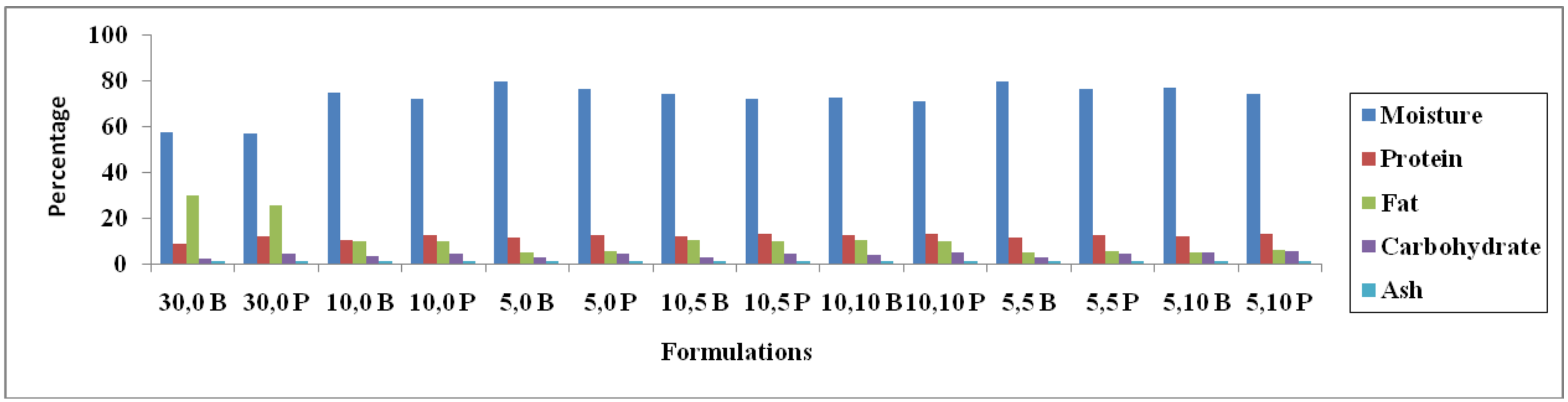


Fig. 10. Proximate composition of of differnt formulations of uncooked batter and cooked LFF

LBT-Lean beef trimmings; **LPT**-Lean pork trimmings; **PSC**- Pork Skin Collagen; **30,0**-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **B**-Batter; **P**-Product; **LFF** –Low fat frankfurter differ significantly (P> 0.05). The formulation 10/10 had significantly the highest (P< 0.05) protein content among all. The fat content of full fat frankfurter was significantly the highest (P< 0.05) than all other formulations. The formulations 10/0, 10/5 and 10/10 showed significantly higher (P< 0.05) fat content than 5/0, 5/5, and 5/10 which showed no significant difference (P> 0.05) among them.

4.2.2.3. Effect of Cooking on the Proximate Composition of the Frankfurters

On comparison of the proximate composition of the uncooked frankfurter batter and cooked frankfurters, an apparent reduction (P< 0.05) in the moisture content and apparent increase (P< 0.05) in the protein, fat, carbohydrate and ash content of all formulations except a reduction in fat content of full fat frankfurter was noticed.

4.3. NUTRITIONAL VALUE

4.3.1. Calorific value of nutrients and their per cent contribution to the Recomm ended Daily Allowance (RDA)

Calorific value obtained from carbohydrate, protein and fat present in 100g of frankfurter of different formulations and their percentage contribution to the RDA is given in the Table 8 and the trend is illustrated in Fig 11 and 12.

The calories from carbohydrate and its percentage contribution to RDA were, 16.72 ± 1.50 and 0.76 ± 0.07, 16.92 ±0.55 and 0.77 ± 0.09, 18.08 ± 0.85 and 0.82 ± 0.11, 16.68 ± 0.75 and 0.76 ± 0.08, 19.24 ± 0.81 and 0.87 ± 0.06, 16.48 ± 0.97 and 0.75 ± 0.08, 22.80 ± 1.01, 1.04 ± 0.13, respectively for the formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10. Similarly, for protein these were 48.60 ± 0.75 and 2.21 ± 0.07, 49.84 ± 0.77 and 2.27 ± 0.04, 50.08 ± 0.69 and 2.28 ± 0.11, 52.04 ± 0.81 and 2.37 ± 0.13, 52.36 ± 0.92 and 2.38 ± 0.08, 51.08 ± 0.75 and 2.32 ± 0.09, 51.40 ± 0.84, 2.34 ± 0.10, respectively in these formulations. The calories from fat and its percentage contribution to RDA from the formulations 30/0, 10/0, 5/0,

Table 8. Calorific value of nutrients and their per cent contribution to the RDA in different formulations of LFF

Formulations		Carbohydrate	Protein	Fat	Total
30/0	kcal/100g	$16.72^{ab} \pm 0.62$	$48.60^{a} \pm 0.75$	$232.11^{c} \pm 0.92$	$297.43^{c} \pm 0.85$
	% of RDA	$0.76^{ab} \pm 0.07$	$2.21^{a} \pm 0.07$	$10.55^{c} \pm 0.12$	$13.52^{c} \pm 0.02$
10/0	kcal/100g	$16.92^{ab} \pm 0.55$	$49.84^{ab} \pm 0.77$	$88.65^{b} \pm 0.71$	$155.41^{b} \pm 0.62$
	% of RDA	$0.77^{ab} \pm 0.09$	$2.27^{ab} \pm 0.04$	$4.03^{b} \pm 0.08$	$7.06^{b} \pm 0.08$
5/0	kcal/100g	$18.08^{ab} \pm 0.85$	$50.08^{ab} \pm 0.69$	$47.70^{a} \pm 0.40$	$115.86^{a} \pm 0.54$
	% of RDA	$0.82^{ab} \pm 0.11$	$2.28^{ab} \pm 0.11$	$2.17^{a} \pm 0.07$	$5.27^{a} \pm 0.07$
10/5	kcal/100g	$16.68^{a} \pm 0.75$	$52.04^{b} \pm 0.81$	$86.13^{b} \pm 0.71$	$154.85^{b} \pm 0.44$
	% of RDA	$0.76^{a} \pm 0.08$	$2.37^{b} \pm 0.13$	$3.92^{b} \pm 0.11$	$7.05^{b} \pm 0.07$
10/10	kcal/100g	$19.24^{b} \pm 0.81$	$52.36^{b} \pm 0.92$	$88.38^{b} \pm 0.65$	$159.98^{b} \pm 0.45$
	% of RDA	$0.87^{b} \pm 0.06$	$2.38^{b} \pm 0.08$	$4.02^{b} \pm 0.12$	$7.27^{b} \pm 0.06$
5/5	kcal/100g	$16.48^{a} \pm 0.97$	$51.08^{ab} \pm 0.75$	$48.42^{a} \pm 0.71$	$115.98^{a} \pm 0.74$
	% of RDA	$0.75^{a} \pm 0.08$	$2.32^{ab} \pm 0.09$	$2.20^{a} \pm 0.15$	$5.27^{a} \pm 0.04$
5/10	kcal/100g	$22.80^{c} \pm 1.01$	$51.40^{ab} \pm 0.84$	$52.11^{a} \pm 0.45$	$126.31^{a} \pm 0.63$
	% of RDA	$1.04^{c} \pm 0.13$	$2.34^{ab} \pm 0.10$	$2.37^{a} \pm 0.11$	$5.74^{a} \pm 0.08$

Means bearing same alphabets in the same column do not indicate significant difference ($P < 0.05$).

10/5, 10/10, 5/5 and 5/10 were 232.11 ± 0.92 and 10.55 ± 0.12, 88.65 ± 0.71 and 4.03 ± 0.08, 47.70 ± 0.40 and 2.17 ± 0.07, 86.13 ± 0.71 and 3.92 ± 0.11, 88.38 ± 0.65 and 4.02 ± 0.12, 48.42 ± 0.71 and 2.20 ± 0.15, 52.11 ± 0.45, 2.37 ± 0.11, respectively.

The total energy value in each frankfurter formulation and their per cent contribution to RDA were 297.43 ± 0.85 and 13.52 ± 0.02, 155.41 ± 0.62 and 7.06 ± 0.08, 115.86 ± 0.54 and 5.27± 0.07, 154.85 ± 0.44 and 7.05 ± 0.07, 159.98 ± 0.45 and 7.27 ± 0.06, 115.98 ± 0.74 and 5.27 ± 0.04, 126.31 ± 0.63 and 5.74 ± 0.08 in 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, 5/10, respectively.

In the full fat frankfurter, the calories from fat, total calories and their contribution to RDA were the highest (P< 0.05) compared to other treatments. But the corresponding values in 10/0, 10/5 and 10/10 did not differ significantly (P> 0.05). Formulation 5/0 was the one with the lowest total calories and its energy value from fat did not differ significantly (P> 0.05) from that of 5/5 and 5/10. Formulation 10/10 contained significantly more (P< 0.05) calories from protein and carbohydrate followed by 10/5 formulation. Similarly, the calories from protein and their contribution to RDA were not significantly different (P> 0.05) among 10/0, 5/0, 5/5 and 5/10 formulations. The total calories significantly decreased in the following order: 30/0, 10/10, 10/0, 10/5, 5/10, 5/5 and 5/0.

4.3.2. Per Cent Daily Value of Protein

The percentage contribution of protein present in 100g of different formulations of to the RDA is presented in the Table 9 and in Fig 13.

The per cent daily values of protein from formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 were 20.25 ± 0.35, 20.77 ± 0.51, 20.87 ± 0.64, 21.68 ± 0.47, 21.82 ± 0.56, 21.28 ± 0.65, 21.42 ± 0.47, respectively.

Table 9. Per cent of RDA of protein in different formulations of LFF

Nutrient	**Formulations (Fat/PSC)**						
% of RDA	**30/0**	**10/0**	**5/0**	**10/5**	**10/10**	**5/5**	**5/10**
Protein	20.25^{a} ± 0.35	20.77^{ab} ± 0.51	20.87^{ab} ± 0.64	21.68^{b} ± 0.47	21.82^{b} ± 0.56	21.28^{ab} ± 0.65	21.42^{ab} ± 0.47

Means bearing same superscripts in the same row do not indicate significant difference (P< 0.05).

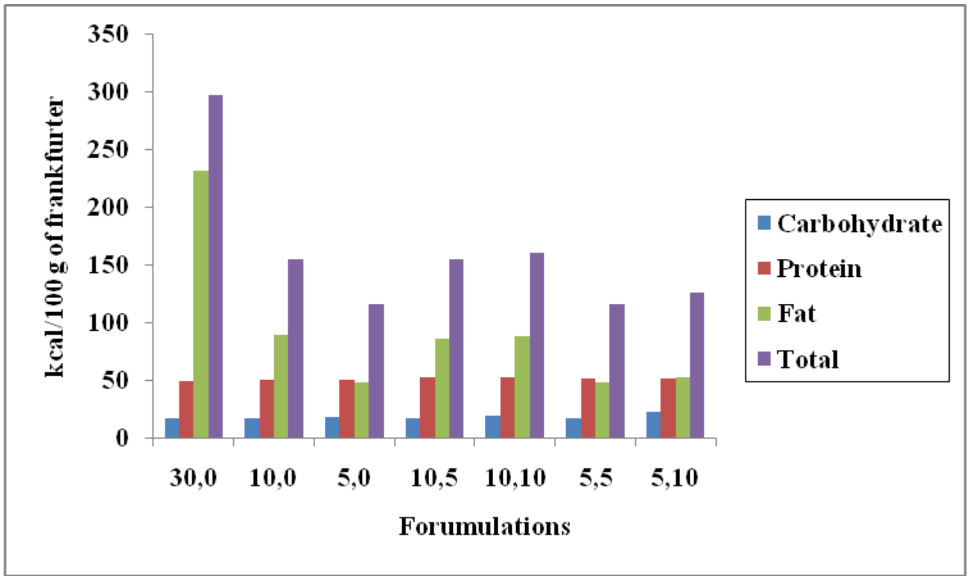


Fig.11. Calorific value of nutrients in different formulations of LFF

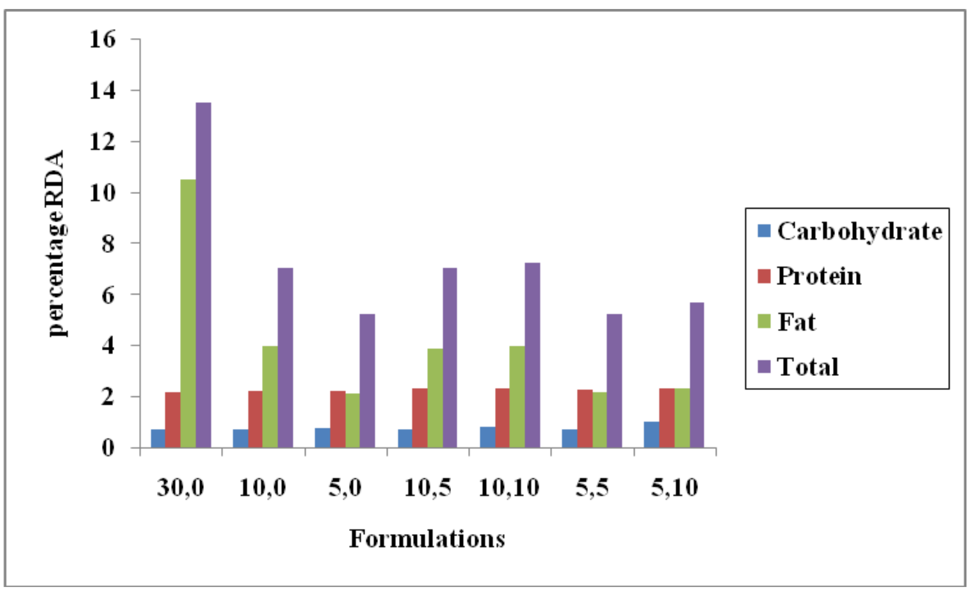


Fig.12. Contribution of nutrients to RDA

30,0-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10, 10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC**- Pork Skin Collagen; **LFF**-Low fat frankfurter

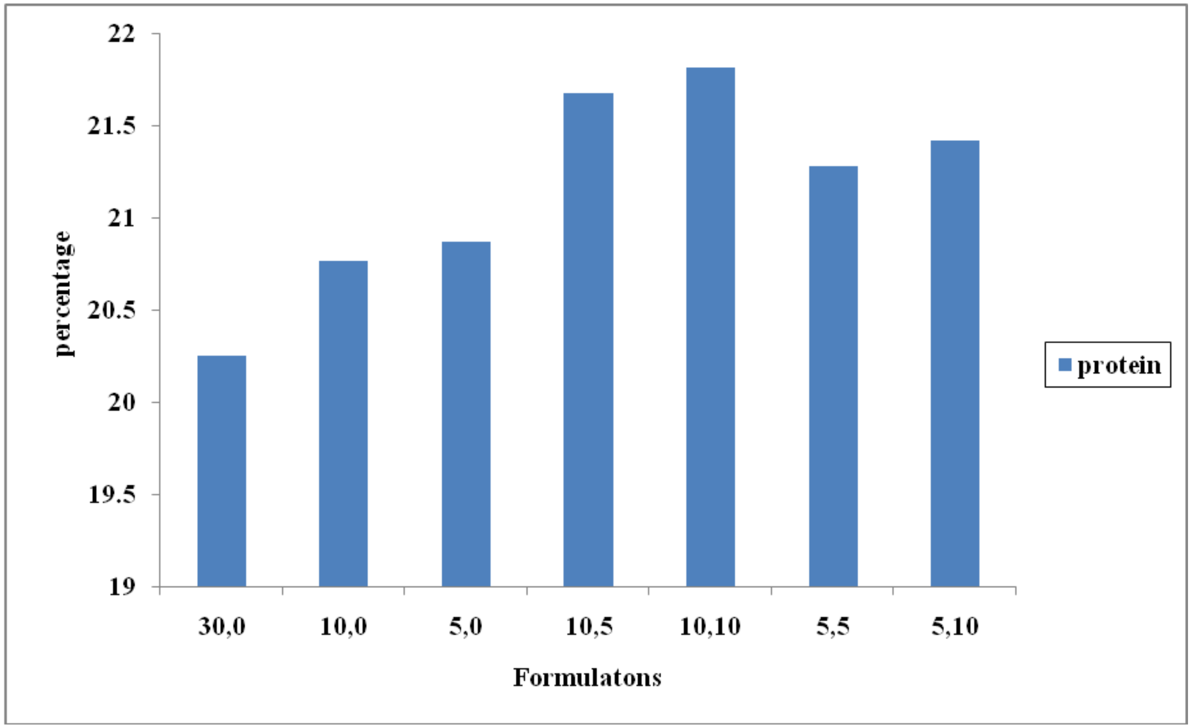


Fig.13. percent daily value of protein in different formulations of LFF

30,0-30% full fat;**10,0-**10% low fat;**5,0-**5% low fat; **10,5-**10% fat with 5% PSC; **10, 10 -**10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC-** Pork Skin Collagen; **LFF-**Low fat frankfurter

The per cent daily value of protein from 10/5 and 10/10 were significantly higher ($P < 0.05$) than full fat control. There was no significant difference ($P > 0.05$) noted in the protein content of 10/0, 5/0, 5/5 and 5/10 formulations.

4.4. EFFECT OF PACKAGING AND STORAGE ON THE QUALITY OF LFF

4.4.1. Purge Loss (PL)

The changes in the PL of different formulations of frankfurters packaged aerobically and under vacuum from d zero to d 75 on storage at -20 ^{o}C are presented in Table 10 and the trend is illustrated in Fig. 14.

The purge loss of different formulations, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 under AP on d 15 were 0.11 ± 0.01, 0.40 ± 0.01, 0.56 ± 0.02, 0.38 ± 0.01, 0.36 ± 0.01, 0.53 ± 0.01, 0.50 ± 0.01, respectively. Under VP they were 0.11 ± 0.01, 0.38 ± 0.01, 0.55 ± 0.02, 0.37 ± 0.01, 0.35 ± 0.01, 0.51 ± 0.02, 0.47 ± 0.01, respectively.

Under AP full fat frankfurter showed significantly the lowest ($P < 0.05$) purge among all formulations. Among treatments 10/10 showed significantly lower ($P < 0.05$) purge and 5/0 showed significantly higher ($P < 0.05$) purge. Formulations 5/5 and 5/10 showed significantly higher ($P < 0.05$) purge than 10/5 and 10/10 formulations. The formulation 5/0 showed significantly the highest ($P < 0.05$) purge among all formulations. The same trend was noticed between formulations on d 30, 45, 60 and 75 in both packaging systems. The corresponding values gradually and significantly increased ($P < 0.05$) to 0.76 ± 0.09, 1.26 ± 0.03, 1.39 ± 0.03, 1.16 ± 0.04, 1.06 ± 0.04, 1.29 ± 0.04, 1.17 ± 0.04, respectively for formulations, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 under AP on d 75 while under VP they were 0.75 ± 0.10, 1.24 ± 0.03, 1.37 ± 0.04, 1.08 ± 0.02, 0.99 ± 0.02, 1.21 ± 0.03, 1.11 ± 0.04, respectively on d 75.

There was no significant difference (P> 0.05) in purge loss irrespective of the packaging systems on different days of storage.

4.4.2. 2-TBARS value

The changes in 2-TBARS values (mg malonaldehyde/kg of LFF) of different formulations of frankfurters packaged aerobically and under vacuum from day zero to d 75 on storage at -20^{o}C are presented in Table 11 and illustrated in Fig. 15.

The TBARS values of different formulations, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 on the day of preparation were 0.321 ± 0.02, 0.263 ± 0.00, 0.194 ± 0.01, 0.251 ± 0.01, 0.253 ± 0.01, 0.202 ± 0.01, 0.199 ± 0.02, respectively. The TBARS values were significantly the highest (P< 0.05) in 30/0 and significantly the lowest (P< 0.05) in 5/0 among all formulations. The low fat formulations 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 had significantly lower (P< 0.05) TBARS values than full fat control. There was no significant difference (P> 0.05) noticed between 10/0, 10/5 and 10/10. Similarly, no significant difference (P> 0.05) was noticed between 5/0, 5/5 and 5/10. The same trend was noticed on d 15, 30, 45 and 60 in both AP and VP systems. The corresponding values gradually and significantly increased (P< 0.05) from d 45 to 0.511 ± 0.03, 0.360 ± 0.02, 0.317 ± 0.01, 0.357 ± 0.03, 0.335 ± 0.03, 0.315 ± 0.03 and 0.306 ± 0.02 on d 75 in AP and 0.474 ± 0.03, 0.341 ± 0.01, 0.295 ± 0.01, 0.321 ± 0.03, 0.298 ± 0.00, 0.286 ± 0.00 and 0.274 ± 0.02 on d 75 in VP.

Under AP on d 75 full fat control showed significantly the highest (P< 0.05) TBARS value among all formulations. There was no significant difference (P> 0.05) noticed between treatments. The same trend was noticed under VP also on d 75 except that 10/0 formulation had significantly higher (P< 0.05) TBARS value than 5/10 formulation. There was no significant increase (P< 0.05) in the TBARS values

Table 10. Effect of packaging and period of storage on purge loss of LFF stored at -20ºC

Formulations (Fat/PSC)	Aerobic packaging					Vacuum packaging				
	Period of storage, d					Period of storage, d				
	15	**30**	**45**	**60**	**75**	**15**	**30**	**45**	**60**	**75**
30/0	0.11^{aA} ±0.01	0.24^{aAB} ±0.02	0.35^{aB} ±0.03	0.64^{aC} ±0.09	0.76^{aC} ±0.09	0.11^{aA} ±0.01	0.23^{aAB} ±0.01	0.35^{aB} ±0.02	0.63^{aC} ±0.09	0.75^{aC} ±0.10
10/0	0.40^{cA} ±0.01	0.70^{cB} ±0.02	0.97^{dC} ±0.02	1.12^{cdD} ±0.03	1.26^{cdE} ±0.03	0.38^{bA} ±0.01	0.68^{cdB} ±0.02	0.94^{d3C} ±0.02	1.10^{e4D} ±0.03	1.24^{d5E} ±0.03
5/0	0.56^{eA} ±0.02	0.81^{dB} ±0.02	1.05^{dC} ±0.03	1.18^{eD} ±0.02	1.39^{dE} ±0.03	0.55^{dA} ±0.02	0.79^{eB} ±0.02	1.03^{eC} ±0.03	1.16^{eD} ±0.03	1.37^{eE} ±0.04
10/5	0.38^{bcA} ±0.01	0.60^{bB} ±0.04	0.79^{bcC} ±0.03	0.96^{bcD} ±0.05	1.16^{bcE} ±0.04	0.37^{bA} ±0.01	0.57^{bB} ±0.04	0.73^{cC} ±0.03	0.89^{bcD} ±0.04	1.08^{bcE} ±0.02
10/10	0.36^{bA} ±0.01	0.55^{bB} ±0.03	0.70^{bC} ±0.03	0.82^{bD} ±0.04	1.06^{bE} ±0.04	0.35^{bA} ±0.01	0.52^{bB} ±0.03	0.63^{bC} ±0.02	0.78^{bD} ±0.03	0.99^{bE} ±0.02
5/5	0.53^{dA} ±0.01	0.71^{cB} ±0.04	0.95^{dC} ±0.05	1.08^{cdeD} ±0.05	1.29^{cdE} ±0.04	0.51^{cdA} ±0.02	0.69^{dB} ±0.05	0.91^{dC} ±0.04	1.05^{deD} ±0.04	1.21^{cdE} ±0.03
5/10	0.50^{dA} ±0.01	0.63^{bcB} ±0.04	0.82^{cC} ±0.05	1.01^{cdD} ±0.05	1.17^{bcE} ±0.04	0.47^{cA} ±0.01	0.60^{bcB} ±0.03	0.77^{cC} ±0.03	0.96^{cdD} ±0.04	1.11^{bcdE} ±0.04

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference ($P< 0.05$).

PSC- Pork Skin Collagen: **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter.

Table 11. Effect of packaging and period of storage on 2-TBARS value of LFF stored at -20^{o}C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	0.321^{cA} ±0.02	0.334^{Ca} ±0.01	0.351^{dAB} ±0.01	0.381^{bB} ±0.03	0.442^{cC} ±0.01	0.511^{bD} ±0.03	0.321^{cA} ±0.02	0.347^{dA} ±0.02	0.345^{dA} ±0.01	0.371^{cAB} ±0.03	0.421^{cBC} ±0.01	0.474^{cC} ±0.03
10/0	0.263^{bA} ±0.00	0.272^{bAB} ±0.01	0.295^{cABC} ±0.02	0.305^{aBC} ±0.02	0.324^{bC} ±0.00	0.360^{aD} ±0.02	0.263^{bA} ±0.01	0.272^{cA} ±0.02	0.289^{cAB} ±0.01	0.298^{bABC} ±0.02	0.319^{bBC} ±0.02	0.341^{bC} ±0.01
5/0	0.194^{aA} ±0.01	0.215^{aA} ±0.01	0.248^{abB} ±0.02	0.278^{aBC} ±0.02	0.305^{abCD} ±0.01	0.317^{aD} ±0.01	0.194^{aA} ±0.01	0.211^{aAB} ±0.00	0.236^{abBC} ±0.02	0.269^{abCD} ±0.02	0.284^{abD} ±0.02	0.295^{abD} ±0.01
10/5	0.251^{bA} ±0.01	0.269^{bAB} ±0.00	0.291^{bcAB} ±0.02	0.307^{aB} ±0.01	0.312^{abB} ±0.02	0.357^{aC} ±0.03	0.251^{bA} ±0.01	0.257^{cA} ±0.01	0.267^{bcA} ±0.02	0.279^{abAB} ±0.01	0.288^{abAB} ±0.01	0.321^{abB} ±0.03
10/10	0.253^{bA} ±0.01	0.261^{bAB} ±0.01	0.281^{bcAB} ±0.02	0.296^{aABC} ±0.02	0.305^{abBC} ±0.01	0.335^{aC} ±0.03	0.253^{bA} ±0.01	0.254^{bcA} ±0.02	0.265^{bcAB} ±0.02	0.271^{abAB} ±0.01	0.287^{abAB} ±0.02	0.298^{abB} ±0.00
5/5	0.202^{aA} ±0.01	0.216^{aA} ±0.01	0.235^{aAB} ±0.02	0.268^{aBC} ±0.007	0.295^{abC} ±0.02	0.315^{aC} ±0.03	0.202^{aA} ±0.01	0.214^{abAB} ±0.008	0.220^{aAB} ±0.02	0.250^{abBC} ±0.03	0.267^{aC} ±0.01	0.286^{abC} ±0.00
5/10	0.199^{aA} ±0.02	0.203^{aA} ±0.01	0.217^{aA} ±0.02	0.262^{aB} ±0.01	0.282^{aBC} ±0.01	0.306^{aC} ±0.02	0.199^{aA} ±0.02	0.200^{aA} ±0.01	0.204^{aA} ±0.01	0.226^{aAB} ±0.02	0.251^{aBC} ±0.01	0.274^{aC} ±0.02

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference ($P< 0.05$).

PSC- Pork Skin Collagen: **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10%; PSC; **5/5**-5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter.

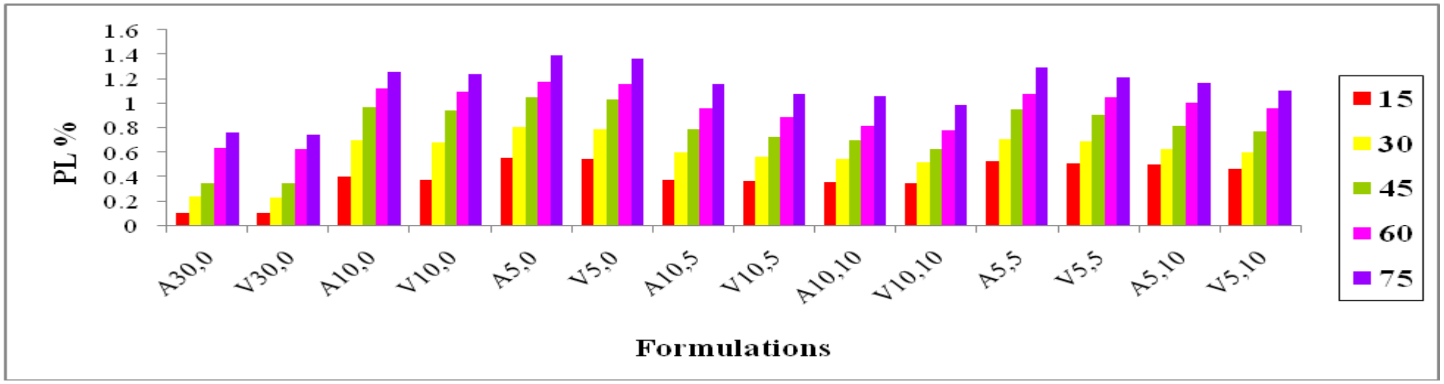


Fig. 14. Effect of packaging and period of storage on PL value of LFF stored at -20ºC

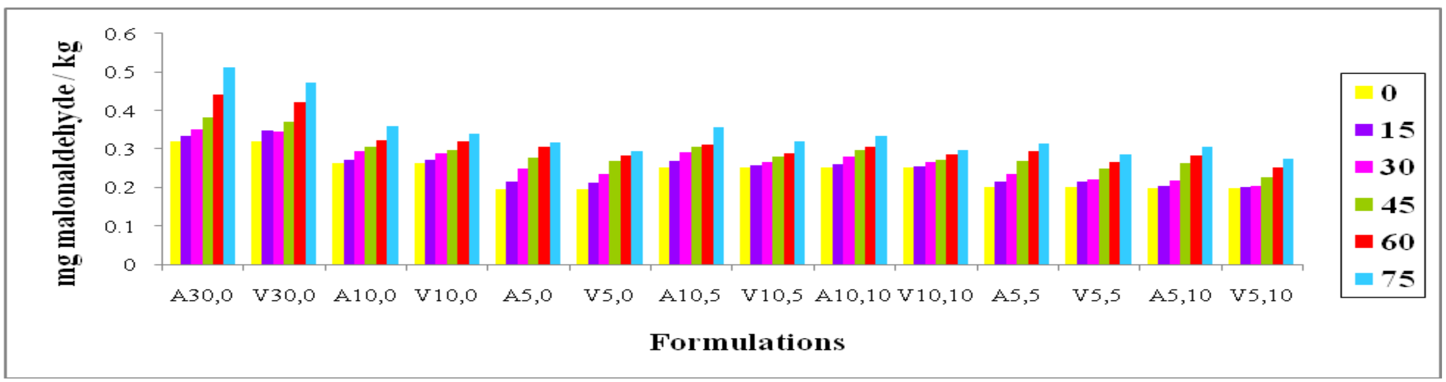


Fig. 15. Effect of packaging and period of storage on 2-TBARS value of LFF stored at -20ºC

30,0-30% full fat;**10,0-**10% low fat;**5,0-**5% low fat; **10,5-**10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC-** Pork Skin Collagen; **PL**-Purge loss; **2-TBARS-** 2-Thiobarbituric Acid Reactive Substances Value:.**LFF-**Low fat frankfurter; **A-**Atmospheric packaging; **V-**Vacuum packaging.

of LFF formulations treated with PSC during the initial 30 days of storage in both types of packaging at -20^{o}C.

There was no significant difference (P> 0.05) in TBARS values between packaging systems on different days of storage.

4.4.3. Sensory Evaluation on the Day of Preparation and on Storage

The panel scores of sensory qualities of different formulations of LFF on the day of preparation are given in the Table 12 and in Fig. 16.

The sensory panel scores for appearance and colour on an eight point Hedonic scale of control and treatments 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 were 7.00 ± 0.00, 7.00 ± 0.00, 7.17 ± 0.17, 7.00 ± 0.13, 7.25 ± 0.11, 7.08 ± 0.08, 7.00 ± 0.13, respectively. The scores for flavour were 6.52 ± 0.18, 6.67 ± 0.17, 6.42 ± 0.15, 6.50 ± 0.13, 6.72 ± 0.13, 6.47 ± 0.13, 6.50 ± 0.13, respectively. The scores for texture were 7.27 ± 0.20, 6.67 ± 0.17, 6.02 ± 0.29, 6.39 ± 0.20, 6.88 ± 0.20, 5.81 ± 0.21, 6.06 ± 0.19, respectively. The scores for saltiness were 6.97 ± 0.30, 6.60 ± 0.40, 6.43 ± 0.24, 6.52 ± 0.26, 7.00 ± 0.34, 6.83 ± 0.21, 6.77 ± 0.21, respectively. The scores for juiciness were 6.68 ± 0.60, 6.63 ± 0.25, 6.96 ± 0.28, 6.85 ± 0.35, 7.00 ± 0.34, 7.22 ± 0.28, 6.47 ± 0.27, respectively. The scores for mouth coating were 6.85 ± 0.33, 6.47 ± 0.26, 6.52 ± 0.18, 5.93 ± 0.31, 6.52 ± 0.26, 6.50 ± 0.26, 6.35 ± 0.28, respectively. The overall acceptability scores were 7.01 ± 0.07, 6.68 ± 0.26, 6.05 ± 0.36, 6.68 ± 0.21, 6.85 ± 0.34, 6.12 ± 0.24, 6.39 ± 0.14, respectively.

The panel scores for appearance and colour, flavour, saltiness, juiciness, mouth coating and overall acceptability did not show any significant difference (P> 0.05) among seven frankfurter formulations which indicated that neither reduction of fat nor addition of PSC did not affect these sensory attributes of LFF. But significant

Table 12. Sensory evaluation score of different formulations LFF on the day of preparation

Attributes	Formulations (Fat/PSC)						
	30/0	**10/0**	**5/0**	**10/5**	**10/10**	**5/5**	**5/10**
Appearance and colour	7.00±0.00	7.00±0.00	7.17± 0.17	7.00±0.13	7.25±0.11	7.08±0.08	7.00±0.13
Flavour	6.52±0.18	6.67±0.17	6.42± 0.15	6.50±0.13	6.72±0.13	6.47±0.13	6.50±0.13
Texture	7.27^{d} ±0.20	6.67^{bcd}±0.17	6.02^{ab}±0.29	6.39^{abc}±0.20	6.88^{cd}±0.20	5.81^{a}±0.21	6.06^{ab}±0.19
Saltiness	6.97±0.30	6.60±0.40	6.43± 0.24	6.52±0.26	7.00± 0.34	6.83±0.21	6.77±0.21
Juiciness	6.68±0.6	6.63±0.25	6.96± 0.28	6.85±0.35	7.00±0.34	7.22±0.28	6.47±0.27
Mouth coating	6.85±0.33	6.47± 0.26	6.52± 0.18	5.93±0.31	6.52± 0.26	6.50±0.26	6.35±0.28
Overall acceptability	7.01±0.07	6.68± 0.26	6.05± 0.36	6.68±0.21	6.85±0.34	6.12±0.24	6.39±0.14

Means bearing same alphabets in the same row do not indicate significant difference (P< 0.05).

(P> 0.05) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter.

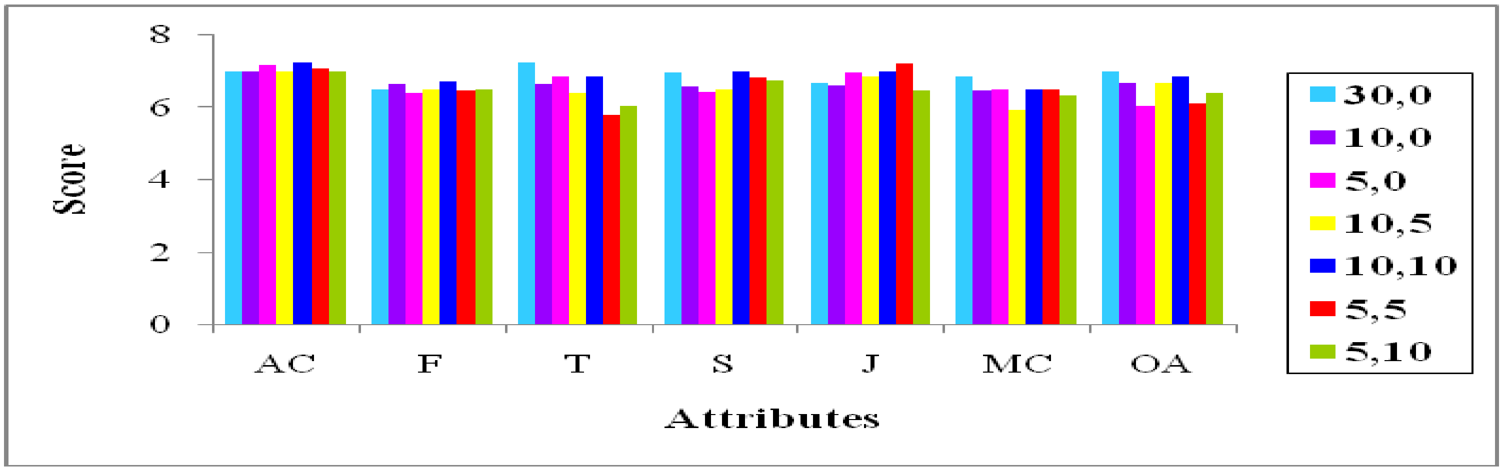


Fig. 16. Sensory evaluation score of different formulations of LFF on the day of preparation

30,0-30% full fat;**10,0-**10% low fat;**5,0-**5% low fat; **10,5-**10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC-** Pork Skin Collagen; **LFF-**Low fat frankfurter; AC- Appearance and Colour; F- Flavour;T- Texture; S- Saltiness; J- Juiciness; MC- Mouth coating; OA- Overall acceptability.

difference among formulations on the textural quality of the products was noticed. The full fat frankfurter had significantly the highest ($P < 0.05$) score for texture among all formulations followed by 10/10. Significant differences in texture were noticed among the treatments 5/0, 10/5, 5/5 and 5/10.

For overall acceptability, the panelists rated 10/10 LFF formulation only next to full fat control. In general, none of the panelists registered negative comments on any of the attributes of all seven formulations.

4.4.3.1. Appearance and Colour

The effect of packaging and period of storage on the appearance and colour scores of LFF stored at -20^{o}C is presented in Table 13 and trend illustrated in Fig. 17.

The appearance and colour scores of the different formulations, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 packaged aerobically, on different days of storage up to d 75 ranged from 7.00 ± 0.00 to 7.00 ± 0.00, 7.00 ± 0.00 to 7.00 ± 0.00, 7.17 ± 0.17 to 6.83 ± 0.17, 7.00 ± 0.13 to 7.00 ± 0.00, 7.25 ± 0.11 to 7.00 ± 0.00, 7.08 ± 0.08 to 7.00 ± 0.22, 7.00 ± 0.13 to 7.00 ± 0.00 , respectively. Panel scores for the same, packaged under vacuum ranged from 7.00 ± 0.00 to 7.00 ± 0.00, 7.00 ± 0.00 to 6.67 ± 0.17, 7.17 ± 0.17 to 6.60 ± 0.17, 7.00 ± 0.00 to 6.83 ± 0.17, 7.25 ± 0.11 to 7.00 ± 0.00, 7.08 ± 0.08 to 6.67 ± 0.21, 7.00 ± 0.13 to 6.83 ± 0.17, respectively.

No significant difference ($P > 0.05$) in the appearance and colour among formulations noticed during the entire period of storage under AP. Formulation 10/10 had significantly lower ($p < 0.5$) score on d 15, 45 and 60 than rest of the days.

Under VP on d 45, the formulation 5/5 had significantly higher ($P < 0.05$) score than 10/0 and 5/0 formulations. There was no significant difference ($P > 0.05$)

noticed between the rest of the formulations on appearance and colour. Formulation 5/0 had significantly the highest ($p < 0.5$) score than other days of storage.

There was no significant difference ($P > 0.05$) between the appearance and colour score of two different packaging systems.

4.4.3.2. Flavour

The effect of packaging and period of storage on the flavour scores of LFF stored at -20^{o}C are presented in Table 14 and the trend is depicted in Fig. 18.

The flavour score for the formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 under AP were 6.52 ± 0.18, 6.67 ± 0.17, 6.42 ± 0.15, 6.50 ± 0.13, 6.72 ± 0.13, 6.47 ± 0.13 and 6.50 ± 0.13 on day zero to 6.17 ± 0.48, 6.00 ± 0.00, 5.67 ± 0.17, 6.00 ± 0.22, 6.00 ± 0.22, 6.00 ± 0.00 and 5.92 ± 0.08 on d 75.

The flavour score of only 5/0 formulation under aerobic packaging significantly ($p < 0.5$) reduced on d 60. The flavour scores for all other formulations were not affected on storage up to 75 days under AP.

The flavour score for the formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 under VP at -20^{o}C were 6.52 ± 0.18, 6.67 ± 0.17, 6.42 ± 0.15, 6.50 ± 0.13, 6.72 ± 0.13, 6.47 ± 0.13 and 6.50 ± 0.13 on d zero and 6.83 ± 0.31, 5.83 ± 0.17, 5.67 ± 0.17, 6.33 ± 0.21, 6.33 ± 0.21, 5.83 ± 0.31, 6.33 ± 0.21 on d 75, respectively.

Under VP on d 75 full fat frankfurter had significantly the highest ($P<0.05$) score for flavour than 10/0, 5/0 and 5/5 formulations.

There was no significant difference ($P > 0.05$) between the flavour score of

Table 13. Effect of packaging and period of storage on appearance and colour of LFF stored at -20ºC

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	7.00± 0.00	7.08± 0.20	6.93± 0.07	6.75± 0.17	6.67± 0.21	7.00± 0.00	7.00± 0.00	7.08± 0.20	7.00± 0.00	6.92^{bc} ±0.08	6.75± 0.17	7.00± 0.00
10/0	7.00± 0.00	7.22± 0.16	6.92± 0.08	7.00± 0.00	6.67± 0.21	7.00± 0.00	7.00± 0.00	7.00± 0.00	6.50± 0.22	6.50^{ab} ±0.22	6.17± 0.17	6.67± 0.17
5/0	7.17± 0.17	6.83± 0.17	6.85± 0.10	6.75± 0.17	6.17± 0.31	6.83± 0.17	7.17^{A}± 0.17^{1}	6.33^{B}± 0.21^{2}	6.25^{B}± 0.25^{2}	6.25^{aB} ±0.17	6.17^{B} ±0.17	6.60^{B}± 0.17
10/5	7.00± 0.13	6.92± 0.08	6.92± 0.08	7.00± 0.00	6.50± 0.22	7.00± 0.00	7.00± 0.00	6.89± 0.19	6.83± 0.17	6.75^{bc} ±0.17	6.67± 0.17	6.83± 0.17
10/10	7.25^{A} ±0.11	6.75^{BC} ±0.17	6.88^{ABC} ±0.12	6.75^{BC} ±0.17	6.50^{C} ±0.22	7.00^{AB} ±0.00	7.25± 0.11	7.06± 0.05	6.93± 0.07	6.92^{bc} ±0.08	6.88± 0.08	7.00± 0.00
5/5	7.08± 0.08	6.92± 0.08	6.85± 0.10	7.00± 0.00	6.50± 0.22	7.00± 0.22	7.08± 0.08	6.83± 0.28	6.77± 0.17	7.00^{c} ±0.00	6.71± 0.16	6.67± 0.21
5/10	7.00± 0.13	6.92± 0.08	6.75± 0.17	6.67± 0.17	6.67± 0.21	7.00± 0.00	7.00± 0.13	6.67± 0.17	6.88± 0.18	6.83^{bc} ±0.17	6.63± 0.24	6.83± 0.17

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference (P< 0.05).

(P> 0.05) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; 30/**0** - 30% full fat; 10/**0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

Table 14. Effect of packaging and period of storage on flavour of LFF stored at -20°C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	15	30	45	60	75	0	15	30	45	60	75
30/0	6.52± 0.18	6.89± 0.27	6.50± 0.18	6.92± 0.27	6.17± 0.40	6.17± 0.48	6.52± 0.18	6.92± 0.27	6.67± 0.21	6.75± 0.17	6.50± 0.22	6.83^{b} ±0.31
10/0	6.67± 0.17	6.67± 0.33	6.33± 0.21	6.75± 0.31	6.00± 0.37	6.00± 0.00	6.67± 0.17	6.50± 0.22	6.00± 0.26	6.08± 0.27	5.83± 0.31	5.83^{a} ±0.17
5/0	6.42^{A} ± 0.15	6.05^{AB} ±0.05	6.00^{AB} ±0.22	6.08^{AB} ±0.45	5.50^{B} ±0.34	5.67^{AB} ±0.17	6.42± 0.15	6.08± 0.08	5.58± 0.33	5.58± 0.33	5.33± 0.33	5.67^{a} ±0.17
10/5	6.50± 0.13	6.42± 0.20	6.05± 0.42	6.42± 0.20	5.83± 0.31	6.00± 0.22	6.50± 0.13	6.22± 0.16	6.00± 0.26	5.96± 0.48	6.50± 0.22	6.33^{ab} ±0.21
10/10	6.72± 0.13	6.43± 0.20	5.93± 0.33	6.42± 0.20	6.00± 0.37	6.00± 0.22	6.72± 0.13	6.28± 0.18	6.10± 0.28	6.33± 0.42	6.61± 0.18	6.33^{ab} ± 0.21
5/5	6.47± 0.13	6.51± 0.17	5.93± 0.17	6.08± 0.33	5.83± 0.31	6.00± 0.00	6.47± 0.13	6.53± 0.18	6.10± 0.19	5.78± 0.49	6.55± 0.39	5.83^{a} ±0.31
5/10	6.50± 0.13	6.08± 0.20	5.93± 0.17	6.29± 0.31	5.75± 0.31	5.92± 0.08	6.50± 0.13	6.25± 0.17	6.22± 0.31	5.71± 0.36	6.55± 0.39	6.33^{ab} ±0.21

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference (P< 0.05).

(P> 0.05) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfur

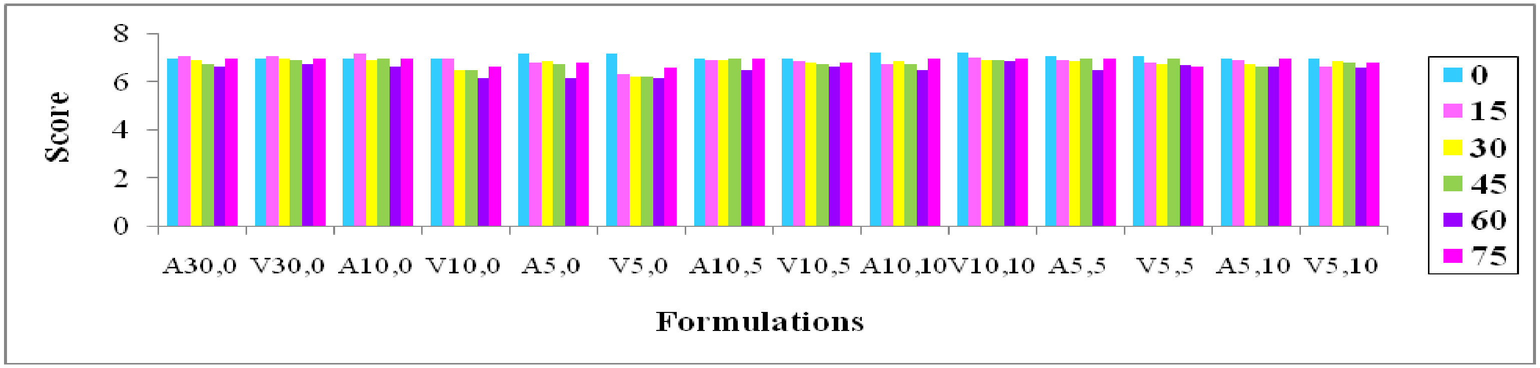


Fig. 17. Effect of packaging and period of storage on appearance and colour of LFF stored at -20ºC

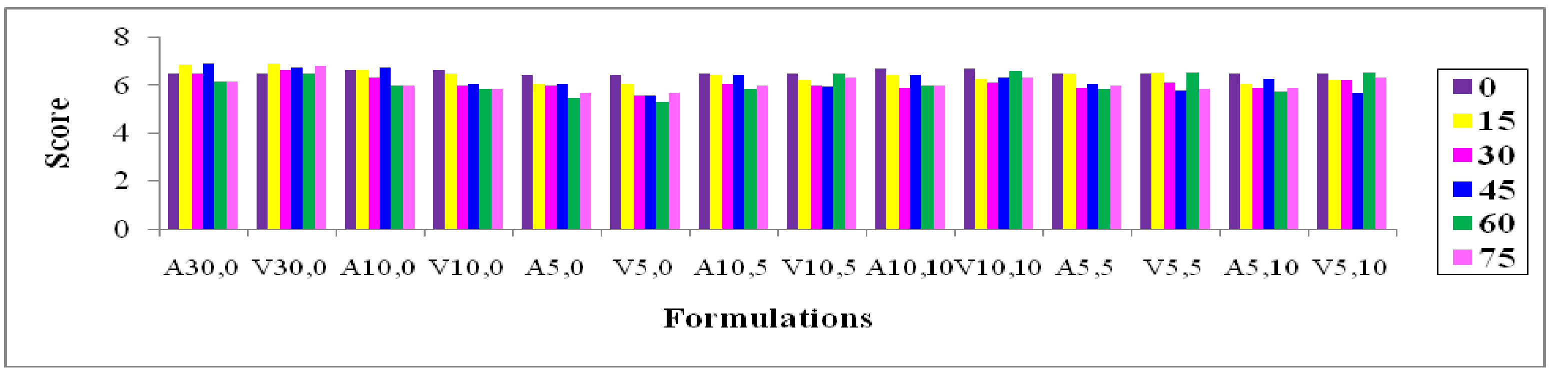


Fig. 18. Effect of packaging and period of storage on flavour of LFF stored at -20ºC

30,0-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC**- Pork Skin Collagen; **LFF**-Low fat frankfurter; **A**-Atmospheric packaging; **V**-Vacuum packaging.

two different packaging systems. During the course of study and sensory evaluation no off flavour or spoilage was detected at any stage by the sensory panelists.

4.4.3.3. Texture

The effect of packaging and period of storage on the texture scores of LFF stored at -20^{o}C are presented in Table 15 and illustrated in Fig. 19.

The mean panel scores for samples packaged aerobically were 7.27 ± 0.20, 6.67 ± 0.17, 6.02 ± 0.29, 6.39 ± 0.20, 6.88 ± 0.20, 5.81 ± 0.22 and 6.06 ± 0.19 on day zero and 6.50 ± 0.22, 6.50 ± 0.18, 6.00 ± 0.34, 6.80 ± 0.13, 7.00 ± 0.37, 6.67 ± 0.42, and 6.33 ± 0.31 on d 75, respectively for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10. The respective scores for vacuum packaged samples for the same formulations ranged from 7.27 ± 0.20, 6.67 ± 0.17, 6.02 ± 0.29, 6.39 ± 0.20, 6.88 ± 0.20, 5.81 ± 0.21 and 6.06 ± 0.19 on day zero to 7.00 ± 0.26, 6.17 ± 0.31, 5.58 ± 0.27, 6.83 ± 0.31, 6.67 ± 0.33, 6.50 ± 0.22 and 6.50 ± 0.22 on d 75.

Under AP the full fat control 30/0 had significantly the highest ($P< 0.05$) score than all other formulations on d zero. Formulation 10/10 had significantly higher ($P< 0.05$) textural score than other formulations except full fat control.

Under VP on d zero full fat frankfurters had significantly the highest ($P< 0.05$) textural score than all other formulations. Formulation 10/10 had significantly higher ($P< 0.05$) textural score than other formulations except full fat control. On d 75 there was no significant difference ($P> 0.05$) noticed among formulations on their textural score.

There was no significant difference ($P> 0.05$) noticed during the different periods of storage in both types of packaging and also between packaging systems.

4.4.3.4. Saltiness

The effect of packaging and period of storage on the saltiness scores of LFF stored at -20^{o}C are presented in Table 16 and illustrated in Fig. 20.

The mean panel scores for saltiness of samples packaged under aerobically ranged from 6.97 ± 0.30, 6.60 ± 0.24, 6.43 ± 0.24, 6.52 ± 0.26, 7.00 ± 0.34, 6.83 ± 0.21 and 6.77 ± 0.21 on d zero to 7.33 ± 0.31, 6.83 ± 0.38, 6.50 ± 0.41, 7.17 ± 0.28, 7.33 ± 0.31, 6.75 ± 0.36, 6.83 ± 0.46, respectively for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 on d 75.

The respective scores for vacuum packaged samples for the same formulations ranged from 6.97 ± 0.30, 6.60 ± 0.24, 6.43 ± 0.24, 6.52 ± 0.26, 7.00 ± 0.34, 6.83 ± 0.21 and 6.77 ± 0.21 on d zero to 7.33 ± 0.17, 6.42 ± 0.27, 5.92 ± 0.33, 6.83 ± 0.31, 7.00 ± 0.26, 6.83 ± 0.31, 6.75 ± 0.36 respectively on d 75.

Under AP there was no significant difference (P> 0.05) among treatments during the different periods of storage.

Under VP on d 30 formulation 5/0 showed significantly the lowest (P< 0.05) saltiness score than all other formulations.

There was no significant difference (P> 0.05) during different periods of storage noticed in AP and VP.

Vacuum packaged 10/0 showed a significantly lower (P< 0.05) score on d 30 and d 45 than its counter part packaged aerobically. Similarly, vacuum packaged 5/0, 10/5 formulations on d 45 and 5/5 formulation on d 15 showed significant decrease (P< 0.05) in the score than the same formulations packaged aerobically.

Table 15. Effect of packaging and period of storage on texture of LFF stored at -20ºC

Formulations (Fat/PSC)	Aerobic packaging Period of storage, d						Vacuum packaging Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	7.27^{d} ± 0.20	6.93± 0.30	6.50± 0.22	6.83± 0.17	6.50^{ab} ±0.50	6.50± 0.22	7.27^{d} ±0.20	6.92± 0.30	6.50± 0.22	7.17^{c} ±0.17	6.67± 0.33	7.00± 0.26
10/0	6.67^{bcd} ± 0.17	6.63± 0.18	6.63± 0.20	6.67± 0.25	6.33^{ab} ± 0.00	6.50± 0.18	6.67^{bcd} ± 0.17	6.50± 0.22	6.13± 0.13	6.08^{ab} ±0.20	6.17± 0.17	6.17± 0.31
5/0	6.02^{ab} ± 0.29	6.38± 0.15	5.80± 0.19	5.75± 0.36	6.08^{ab} ±0.37	6.00± 0.34	6.02^{ab} ± 0.29	5.92± 0.20	5.58± 0.20	5.50^{a} ±0.22	5.83± 0.31	5.58± 0.27
10/5	6.39^{abc} ±0.20	6.67± 0.36	6.38± 0.32	6.42± 0.20	6.33^{ab} ±0.21	6.80± 0.13	6.39^{abc} ±0.20	6.44± 0.20	6.43± 0.33	6.46^{bc} ± 0.29	6.67± 0.17	6.83± 0.31
10/10	6.88^{cd} ±0.20	6.43± 0.29	6.58± 0.20	6.83± 0.21	7.08^{b} ± 0.08	7.00± 0.37	6.88^{cd} ±0.20	6.56± 0.20	6.62± 0.21	6.50^{bc} ±0.22	6.61 ±0.18	6.67± 0.33
5/5	5.81^{a} ±0.21	6.08± 0.20	6.18± 0.24	6.25± 0.28	5.75^{a} ±0.36	6.67± 0.42	5.81^{a} ±0.21	6.33± 0.21	6.37± 0.24	6.33^{abc} ±0.31	6.33± 0.31	6.50± 0.22
5/10	6.06^{ab} ±0.19	6.09± 0.21	5.76± 0.16	6.00± 0.26	5.92^{a} ±0.27	6.33± 0.31	6.06^{ab} ±0.19	6.27± 0.28	6.00± 0.26	5.92^{ab} ±0.42	6.25± 0.17	6.50± 0.22

Means bearing same alphabets in the same column do not indicate significant difference ($P< 0.05$).

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

Table 16. Effect of packaging and period of storage on saltiness of LFF stored at -20^{o}C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	6.97± 0.30	7.02± 0.22	6.63± 0.35	7.08± 0.27	6.67± 0.33	7.33± 0.31	6.97± 0.30	7.00± 0.22	6.67^{b} ±0.33	7.33^{b} ±0.17	6.83± 0.31	7.33± 0.17
10/0	6.60± 0.24	7.02± 0.22	6.72± 0.16	6.83± 0.21	6.33± 0.21	6.83± 0.38	6.60± 0.24	6.83± 0.11	6.08^{ab*} ±0.08	6.25^{a*} ±0.17	6.33± 0.21	6.42± 0.27
5/0	6.43± 0.24	6.77± 0.31	5.92± 0.27	6.92± 0.08	6.33± 0.21	6.50± 0.41	6.43± 0.24	6.42± 0.20	5.50^{a} ±0.22	6.08^{a*} ±0.20	5.83± 0.17	5.92± 0.33
10/5	6.52± 0.26	6.50± 0.22	6.63± 0.20	7.00± 0.00	6.50± 0.22	7.17± 0.28	6.52± 0.26	6.60± 0.33	6.43^{b} ±0.33	6.33^{a*} ± 0.21	6.67± 0.33	6.83± 0.31
10/10	7.00± 0.34	7.02± 0.22	6.67± 0.21	6.92± 0.08	6.67± 0.33	7.33± 0.31	7.00± 0.34	6.85± 0.28	6.80^{b} ±0.16	6.67^{a} ±0.21	6.58± 0.32	7.00± 0.26
5/5	6.83± 0.21	6.93± 0.27	6.22± 0.21	6.67± 0.21	6.25± 0.17	6.75± 0.36	6.83± 0.21	6.17^{*} ±0.17	6.60^{b} ±0.20	6.42^{a} ±0.20	6.71± 0.30	6.83± 0.31
5/10	6.77± 0.21	6.60± 0.20	6.30± 0.24	6.25± 0.25	6.42± 0.27	6.83± 0.46	6.77± 0.21	6.58± 0.33	6.80^{b} ±0.16	6.33^{a} ±0.25	6.54± 0.32	6.75± 0.36

Means bearing same alphabets in the same column do not indicate significant difference (P< 0.05).

* represents significant difference between two packaging systems in the same time period (P< 0.05).

(P> 0.05) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

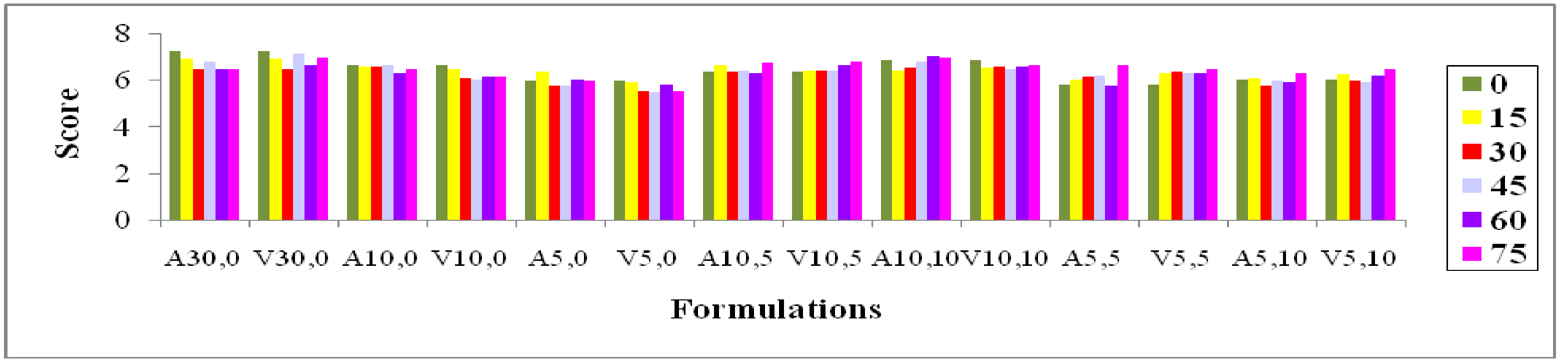


Fig. 19. Effect of packaging and period of storage on texture of LFF stored at -20ºC

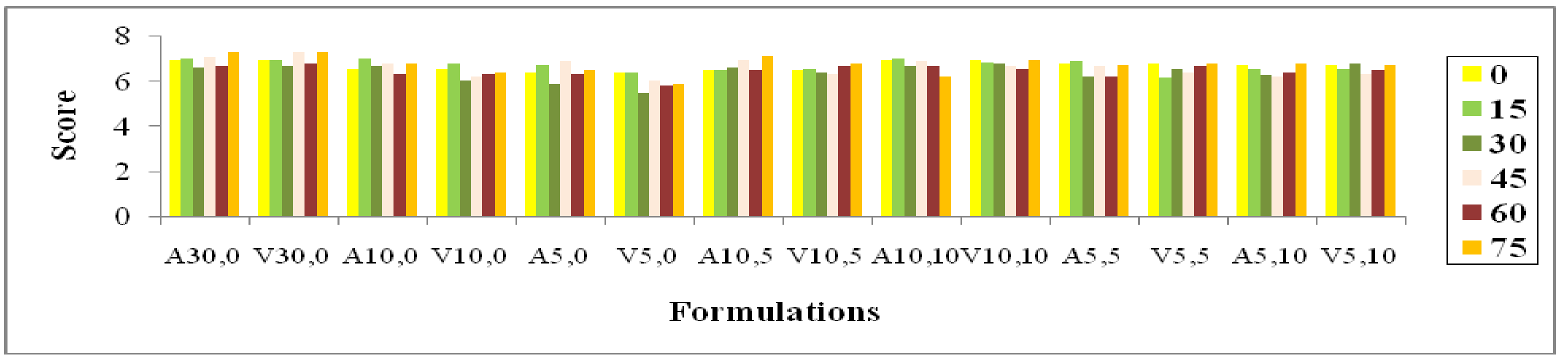


Fig. 20. Effect of packaging and period of storage on saltiness of LFF stored at -20ºC

30,0-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC-** Pork Skin Collagen; **LFF**-Low fat frankfurter; **A** -Atmospheric packaging; **V**-Vacuum packaging.

4.4.3.5. Juiciness

The effect of packaging and period of storage on the juiciness scores of LFF stored at -20^{o}C are presented in Table 17 and the trend in Fig. 21.

The mean panel scores for samples packaged aerobically were 6.68 ± 0.16, 6.63 ± 0.25, 6.96 ± 0.28, 6.85 ± 0.35, 7.00 ± 0.34, 7.22 ± 0.28 and 6.47 ± 0.27 on day zero and 6.33 ± 0.21, 6.50 ± 0.18, 6.25 ± 0.31, 6.50 ± 0.18, 6.67 ± 0.21, 6.33 ± 0.17 and 6.17 ± 0.28 on day 75 for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10. The scores for vacuum packaged samples of same formulations ranged 6.68 ± 0.16, 6.63 ± 0.25, 6.96 ± 0.28, 6.85 ± 0.35, 7.00 ± 0.34, 7.22 ± 0.28 and 6.47 ± 0.27 on day zero to 6.33 ± 0.21, 6.00 ± 0.26, 5.98 ± 0.22, 6.33 ± 0.21, 6.33 ± 0.21, 6.00 ± 0.37 and 5.83 ± 0.31 on d 75.

There was no significant difference (P> 0.05) among treatments during different periods of storage in both types of packaging. Similarly, no significant difference (P> 0.05) was noticed during different periods of storage of formulations in both systems of packaging.

There was no significant difference (P> 0.05) noticed in the juiciness of frankfurters packed under AP and VP systems.

4.4.3.6. Mouth Coating

The effect of packaging and period of storage on the mouth coating scores of LFF stored at -20^{o}C are presented on Table 18 and the trend illustrated in Fig. 22.

The mean panel scores for mouth coating of samples packaged aerobically were 6.85 ± 0.33, 6.47 ± 0.26, 6.52 ± 0.18, 5.93 ± 0.31, 6.52 ± 0.26, 6.50 ± 0.26 and 6.35 ± 0.28 on d zero and 6.83 ± 0.31, 6.83 ± 0.31, 6.58 ± 0.37, 6.67 ± 0.33, 6.83 ±

Table 17. Effect of packaging and period of storage on juiciness of LFF stored at -20^{o}C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	15	30	45	60	75	0	15	30	45	60	75
30/0	6.68± 0.16	6.88± 0.28	6.38± 0.24	6.67± 0.33	5.50± 0.50	6.33± 0.21	6.68± 0.16	7.00± 0.26	6.50± 0.18	6.58± 0.20	6.00± 0.45	6.33± 0.21
10/0	6.63± 0.25	6.80± 0.29	6.42± 0.20	6.17± 0.17	5.33± 0.33	6.50± 0.18	6.63± 0.25	6.25± 0.17	6.08± 0.08	6.00± 0.00	5.33± 0.33	6.00± 0.26
5/0	6.96± 0.28	5.88± 0.38	6.05± 0.26	6.03± 0.23	5.42± 0.27	6.25± 0.31	6.96± 0.28	5.83± 0.40	5.83± 0.17	5.67± 0.31	5.25± 0.17	5.98 ±0.22
10/5	6.85± 0.35	6.25± 0.36	5.97± 0.42	6.08± 0.27	5.75± 0.36	6.50± 0.18	6.85± 0.35	6.47± 0.26	6.54± 0.18	6.17± 0.40	5.83± 0.48	6.33± 0.21
10/10	7.00± 0.34	6.97± 0.35	6.57± 0.49	6.50± 0.41	5.83± 0.40	6.67± 0.21	7.00± 0.34	6.38± 0.28	6.48± 0.32	6.17± 0.48	6.00± 0.26	6.33± 0.21
5/5	7.22± 0.28	6.52± 0.25	6.30± 0.24	6.00± 0.29	5.00± 0.37	6.33± 0.17	7.22± 0.28	6.42± 0.27	6.67± 0.21	5.83± 0.40	6.00± 0.26	6.00± 0.37
5/10	6.47± 0.27	6.63± 0.31	6.42± 0.20	5.75± 0.36	6.00± 0.26	6.17± 0.28	6.47± 0.27	6.00± 0.26	6.15± 0.28	5.50± 0.43	5.88± 0.27	5.83± 0.31

(P> 0.05) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5**-5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

Table 18. Effect of packaging and period of storage on mouth coating of LFF stored at -20ºC

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	6.85± 0.33	7.02± 0.22	6.75± 0.25	6.75± 0.25	6.83± 0.17	6.83± 0.31	6.85± 0.33	7.08± 0.20	6.92^{b} ±0.20	6.67± 0.21	6.83^{abc} ± 0.17	7.00± 0.26
10/0	6.47± 0.26	7.00± 0.26	6.67± 0.25	6.33± 0.21	6.50± 0.34	6.83± 0.31	6.47± 0.26	6.50± 0.22	6.10^{a} ±0.16	6.33± 0.21	6.08^{a} ± 0.37	6.33± 0.21
5/0	6.52± 0.18	6.68± 0.35	6.43± 0.24	6.42± 0.27	6.67± 0.21	6.58± 0.37	6.52± 0.18	6.33± 0.25	6.10^{a} ±0.16	6.25± 0.17	6.17^{ab} ± 0.31	6.08± 0.20
10/5	5.93± 0.31	6.75± 0.31	6.67± 0.25	6.42± 0.27	6.67± 0.21	6.67± 0.33	5.93± 0.31	6.63± 0.31	6.77^{b} ±0.20	6.58± 0.27	7.25^{c} ±0.17	6.67± 0.33
10/10	6.52± 0.26	7.08± 0.33	6.78± 0.26	6.50± 0.32	6.83± 0.17	6.83± 0.31	6.52± 0.26	6.80± 0.29	6.98^{b} ±0.09	6.67± 0.21	7.11^{c} ± 0.19	6.67± 0.33
5/5	6.50± 0.26	6.85± 0.32	6.52± 0.18	6.33± 0.31	6.67± 0.21	6.67± 0.33	6.50± 0.26	6.58± 0.33	6.77^{b} ±0.20	6.42± 0.27	6.94^{bc} ±0.26	6.33± 0.33
5/10	6.35± 0.28	6.85± 0.28	6.52± 0.18	6.25± 0.36	6.50± 0.34	6.50± 0.43	6.35± 0.28	6.58± 0.33	6.77^{b} ±0.17	6.50± 0.22	6.82^{abc} ±0.28	6.33± 0.33

Means bearing same alphabets in the same column do not indicate significant difference (P< 0.05).

(P> 0.05) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5**-5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

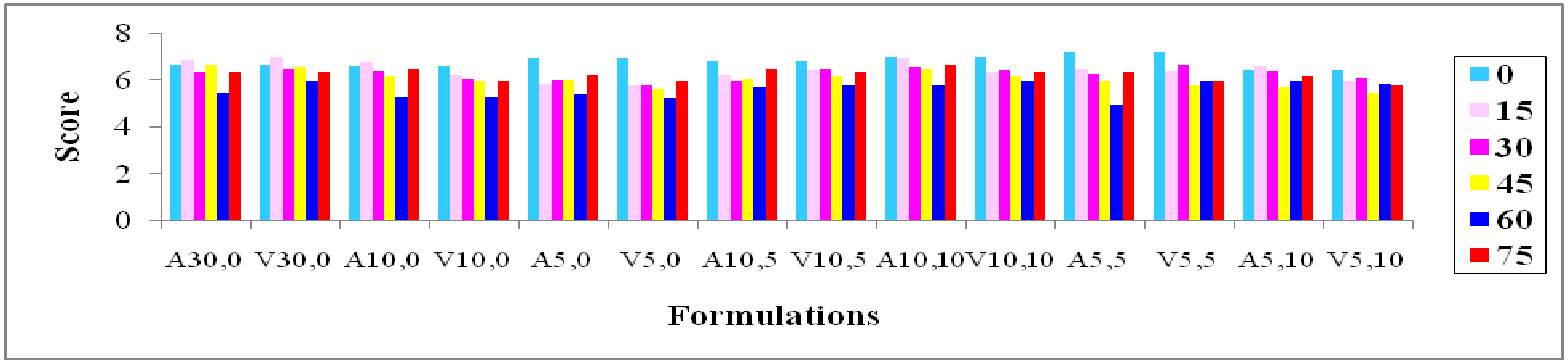


Fig. 21. Effect of packaging and period of storage on juiciness of LFF stored at -20ºC

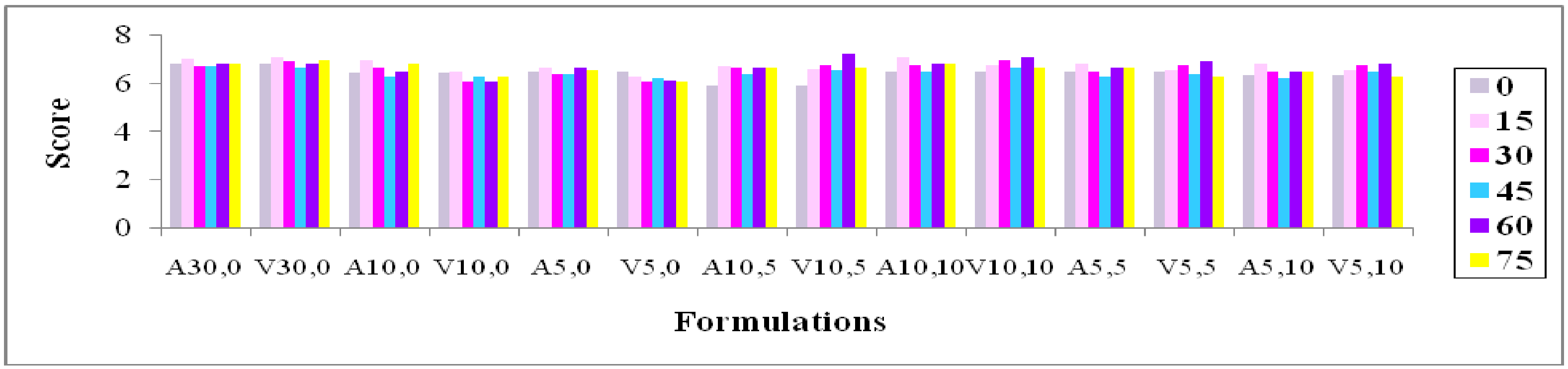


Fig. 22. Effect of packaging and period of storage on mouth coating of LFF stored at -20ºC

30,0-30% full fat;**10,0**-10% low fat;**5,0**-5% low fat; **10,5**-10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5, 10** -5% fat with 10% PSC; **PSC**- Pork Skin Collagen; **LFF**-Low fat frankfurter; **A**-Atmospheric packaging; **V**-Vacuum packaging.

0.31, 6.67 ± 0.33, 6.50 ± 0.43, respectively for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 on d 75.

The respective scores for vacuum packaged samples for the same formulations ranged from 6.85 ± 0.33, 6.47 ± 0.26, 6.52 ± 0.18, 5.93 ± 0.31, 6.52 ± 0.26, 6.50 ± 0.26 and 6.35 ± 0.28 on d zero to 7.00 ± 0.26, 6.33 ± 0.21, 6.08 ± 0.20, 6.67 ± 0.33, 6.67 ± 0.33, 6.33 ± 0.33, 6.33 ± 0.33, respectively on d 75.

There was no significant difference (P> 0.05) in mouth coating between treatments of aerobically packaged samples during different periods of storage. Under vacuum packaging on d 30, the formulation 10/0 and 5/0 had significantly the lowest (P< 0.05) score than rest of the formulations which did not differ significantly (P> 0.05) among them.

There was no significant difference (P> 0.05) during different periods of storage and systems of packaging on the mouth coating of frankfurters.

4.4.3.7. Overall Acceptability

The effect of packaging and period of storage on the overall acceptability scores of LFF stored at -20^{o}C are presented on Table 19 and trend depicted in Fig. 23.

The mean panel scores of samples packaged aerobically were 7.01 ± 0.07, 6.68 ± 0.26, 6.05 ± 0.36, 6.68 ± 0.21, 6.85 ± 0.34, 6.12 ± 0.24 and 6.39 ± 0.14 on d zero and 6.67 ± 0.33, 6.25 ± 0.28, 5.67 ± 0.21, 6.33 ± 0.21, 6.66 ± 0.42, 5.83 ± 0.54, 6.17 ± 0.28 respectively for formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 on d 75.

The respective scores for vacuum packaged samples for the same formulations ranged from 7.01 ± 0.07, 6.68 ± 0.26, 6.05 ± 0.36, 6.68 ± 0.21, 6.85 ± 0.34, 6.12 ± 0.24 and 6.39 ± 0.14 on d zero to 7.13 ± 0.29, 5.83 ± 0.17, 5.17 ± 0.17, 6.42 ± 0.27, 6.72 ± 0.35, 5.92 ± 0.20, and 6.17 ± 0.17 on d 75.

Under AP on d 15 formulations 5/0 and 5/5 had significantly lower ($P < 0.05$) score than 30/0 and 10/10 formulations. On d 30, 5/0 formulation had significantly the lowest ($P < 0.05$) score than that rest of the formulations. The same trend was noticed on d 45 and on d 60, 5/0 showed significantly lower ($P < 0.05$) score than 30/0, 10/5 and 10/10 formulations.

Under VP, the formulation 5/0 showed significantly lower ($P < 0.05$) score than 30/0, 10/5, and 10/10 on d 15, 30, 45 and 60, respectively and on d 75 it showed significantly lower ($P < 0.05$) score than 30/0, 10/5, 10/10, 5/5 and 5/10 formulations. Formulation 10/10 showed significantly higher ($P < 0.05$) overall acceptability than 10/0, 5/0 and 5/5 formulations on d 75 under VP.

With respect to the effect of packaging 10/0 formulation showed significantly lower ($P < 0.05$) score on d 15 and 30 compared to its aerobically packed counter part.

There was no significant difference ($P > 0.05$) in the overall acceptability of frankfurters during different periods of storage in both AP and VP.

4.4.4. Proximate Composition

4.4.4.1. Moisture

The effect of packaging and period of storage on the moisture content of LFF stored at -20^{o}C are presented in Table 20 and depicted the trend in Fig 24.

The moisture content of different formulations, viz., 30/0, 10/0, 5/0, 10/5,

Table 19. Effect of packaging and period of storage on overall acceptability of LFF stored at -20^{o}C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of sorage, d						Period of storage, d					
	0	15	30	45	60	75	0	15	30	45	60	75
30/0	7.01± 0.07	6.95^{b} ± 0.24	6.93^{b} ±0.10	6.83^{d} ±0.11	6.68^{b} ±0.34	6.67± 0.33	7.01± 0.07	6.96^{d} ± 0.25	6.66^{bc} ±0.18	6.92^{d} ±0.08	6.83^{b} ±0.17	7.13^{d} ± 0.29
10/0	6.68± 0.26	6.60^{ab} ±0.13	6.50^{b} ± 0.18	6.40^{bcd} ± 0.20	6.30^{ab} ±0.29	6.25± 0.28	6.68± 0.26	6.10^{ab*} ±0.06	6.00^{ab*} ±0.00	5.92^{ab} ±0.20	5.92^{ab} ±0.30	5.83^{ab} ± 0.17
5/0	6.05± 0.36	6.00^{a} ±0.29	5.98^{a} ±0.27	5.72^{a} ±0.25	5.67^{a} ±0.21	5.67± 0.21	6.05± 0.36	5.75^{a} ± 0.25	5.88^{a} ±0.24	5.58^{a} ±0.20	5.50^{a} ±0.22	5.17^{a} ±0.17
10/5	6.68± 0.21	6.60^{ab} ±0.26	6.48^{ab} ±0.51	6.45^{bcd} ±0.16	6.57^{b} ±0.15	6.33± 0.21	6.68± 0.21	6.50^{bcd} ±0.22	6.78^{c} ±0.16	6.55^{bcd} ± 0.23	6.50^{b} ±0.18	6.42^{bcd} ±0.27
10/10	6.85± 0.34	6.78^{b} ±0.15	6.75^{ab} ±0.13	6.71^{cd} ±0.10	6.58^{b} ± 0.20	6.66± 0.42	6.85± 0.34	6.82^{cd} ± 0.15	6.79^{c} ±0.26	6.77^{cd} ±0.26	6.75^{b} ±0.28	6.72^{cd} ± 0.35
5/5	6.12± 0.24	6.10^{a} ±0.06	6.07^{ab} ±0.29	5.98^{ab} ±0.18	5.90^{ab} ±0.31	5.83± 0.54	6.12± 0.24	6.08^{ab} ±0.08	6.07^{ab} ±0.27	6.04^{ab} ±0.25	5.98^{ab} ±0.26	5.92^{b} ±0.20
5/10	6.39± 0.14	6.35^{ab} ±0.16	6.30^{ab} ±0.25	6.25^{bc} ±0.13	6.21^{ab} ±0.14	6.17± 0.28	6.39± 0.14	6.25^{abc} ±0.31	6.20^{abc} ±0.29	6.18^{abc} ±0.22	6.17^{ab} ±0.35	6.17^{bc} ±0.17

Means bearing same alphabets in the same column do not indicate significant difference ($P< 0.05$).

* represents significant difference between two packaging systems in the same time period ($P< 0.05$).

($P> 0.05$) Means without superscripts do not differ significantly.

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter

Table 20. Effect of packaging and period of storage on moisture content of LFF stored at -20^{o}C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	56.80^{aA} ±0.10	56.67^{aA} ±0.10	56.53^{aA} ±0.10	56.04^{aA} ±0.09	55.51^{aB} ±0.08	55.06^{aC} ±0.10	56.80^{aA} ±0.10	56.65^{aAB} ±0.11	56.44^{aB} ±0.10	56.04^{cA} ±0.11	55.49^{aD}± 0.11	55.20^{aD} ±0.10
10/0	72.43^{cA} ±0.44	72.18^{cB} ±0.42	71.95^{cAB} ±0.40	71.47^{cABC} ±0.42	71.01^{cBC} ±0.42	70.54^{cC} ±0.44	72.43^{cA} ±0.44	72.25^{cA} ±0.45	71.99^{cAB} ±0.44	71.58^{cAB} ±0.45	71.04^{bcBC} ±0.44	70.72^{bcC} ±0.45
5/0	76.54^{eA} ±0.20	76.25^{eA} ±0.18	75.99^{eA} ±0.17	75.44^{eB} ±0.18	74.91^{eB} ±0.18	74.30^{eC} ±0.19	76.54^{eA} ±0.20	76.29^{eA} ±0.19	76.08^{eAB} ±0.20	75.60^{eBC} ±0.19	75.13^{eC} ±0.19	74.47^{eD} ±0.20
10/5	72.32^{cA} ±0.22	72.11^{cA} ±0.21	71.89^{cAB} ±0.19	71.45^{cBC} ±0.22	71.04^{cCD} ±0.23	70.52^{cD} ±0.22	72.32^{cA} ±0.22	72.19^{cA} ±0.22	72.05^{cA} ±0.22	71.70^{cAB} ±0.23	71.34^{cBC} ±0.25	70.97^{cC} ±0.23
10/10	71.30^{bA} ±0.36	71.10^{bAB} ±0.34	70.91^{bAB} ±0.33	70.50^{bABC} ±0.36	70.11^{bBC} ±0.39	69.61^{bC} ±0.36	71.30^{bA} ±0.36	71.18^{bA} ±0.35	71.07^{bA} ±0.36	70.75^{bAB} ±0.37	70.43^{bBC} ±0.38	70.08^{bC} ±0.36
5/5	76.66^{eA} ±0.21	76.41^{eA} ±0.20	76.15^{eAB} ±0.19	75.67^{eBC} ±0.19	75.18^{eC} ±0.21	74.55^{eD} ±0.20	76.66^{eA} ±0.21	76.49^{eAB} ±0.21	76.30^{eAB} ±0.21	75.88^{eBC} ±0.22	75.44^{eCD} ±0.24	74.92^{eD} ±0.21
5/10	74.53^{dA} ±0.17	74.30^{dA} ±0.17	74.06^{dA} ±0.17	73.59^{dA} ±0.15	73.12^{dB} ±0.16	72.55^{dC} ±0.15	74.53^{dA} ±0.17	74.37^{dA} ±0.16	74.21^{dAB} ±0.16	73.83^{dBC} ±0.18	73.43^{cCD} ±0.20	73.03^{dD} ±0.17

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference ($P < 0.05$).

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter.

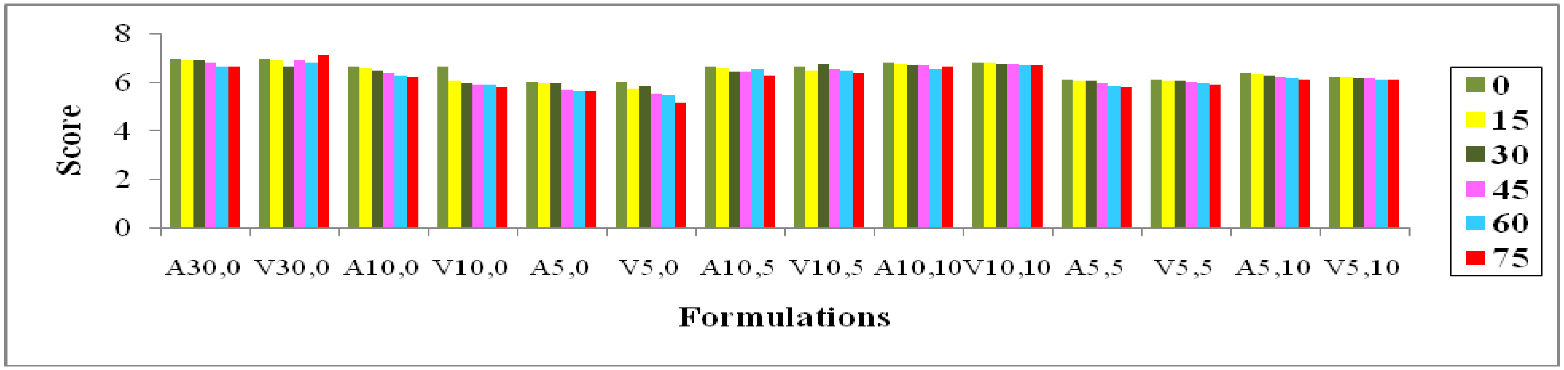


Fig. 23. Effect of packaging and period of storage on overall acceptability of LFF stored at -20ºC

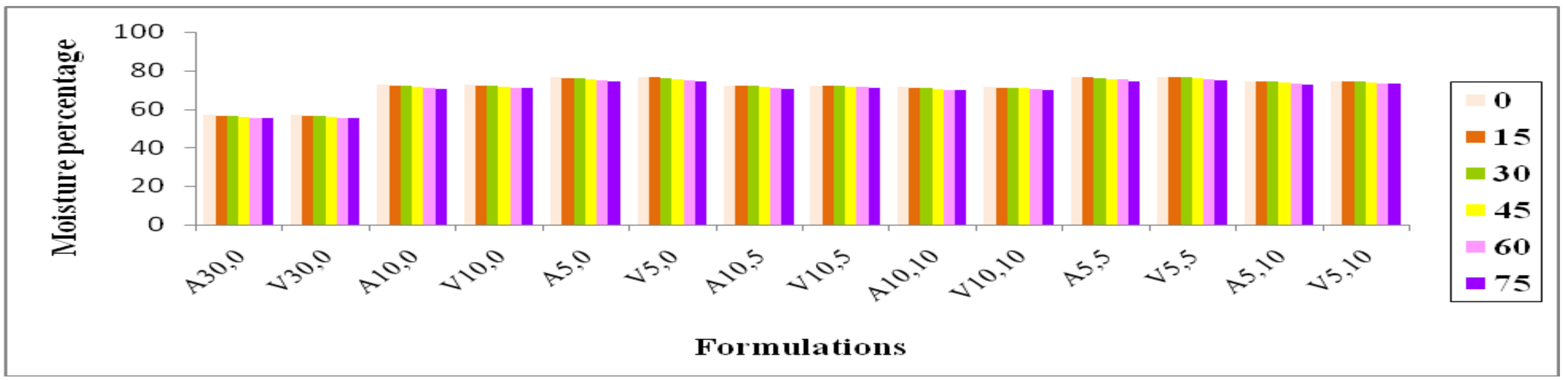


Fig. 24. Effect of packaging and period of storage on moisture content of LFF stored at -20ºC

30,0-30% full fat;**10,0-**10% low fat;**5,0-**5% low fat; **10,5-**10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5,10** -5% fat with 10% PSC; **PSC-** Pork Skin Collagen; **LFF-**Low fat frankfurter; **A-**Atmospheric packaging; **V-**Vacuum packaging.

10/10, 5/5 and 5/10 aerobically packaged and stored at -20ºC reduced to 55.06 ± 0.10, 70.54 ± 0.44, 74.30 ± 0.19, 70.52 ± 0.22, 69.61 ± 0.36, 74.55 ± 0.20, 72.55 ± 0.15, respectively on day 75 compared to those on the day of preparation. The respective values for same formulations packed under vacuum were 55.20 ± 0.10, 70.72 ± 0.45, 74.47 ± 0.20, 70.97 ± 0.23, 70.08 ± 0.36, 74.92 ± 0.21 and 73.03 ± 0.17 on day 75.

Under AP full fat control and 5/0 had significantly the lowest and the highest (P< 0.05) moisture content respectively among all formulations. The moisture content of 10/0, 10/5 and 5/0, 5/5 was significantly higher (P< 0.05) than 10/10 and 5/10 respectively. The same trend between treatments was noticed in all days of storage irrespective of the packaging systems.

Under AP full fat frankfurter and 10/0 had significantly lower (P< 0.05) moisture content from d 60 and d 15 onwards. Formulations 5/0, 10/5 and 10/10 showed significant decrease (P< 0.05) in their moisture content from d 45 and 60 onwards. Similarly formulation 5/5 and 5/10 showed significantly lower (P< 0.05) moisture content on d 45 d 60. On d 75 10/10 had significantly the lowest (P< 0.05) moisture content between treatments.

Under VP full fat control, 5/0 and both 10/5 and 10/10 had significantly lower (P< 0.05) moisture content from d 30, 45 and 60 onwards. Formulations 5/5, 5/10 and 10/10 showed significant decrease (P< 0.05) on moisture content from d 45 and d 60 onwards.

With respect to the effect of packaging there was no significant difference (P> 0.05) between packaging systems.

4.4.4.2. Protein

The effect of packaging and period of storage on the protein content of LFF stored at -20^{o}C are presented in Table 21 and the trend illustrated in Fig. 25.

The per cent protein content of aerobic and vacuum packaged formulations viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 stored at -20^{o}C was raised to 12.68 ± 0.35, 12.61 ± 0.35, 13.86 ± 0.26, 13.71 ± 0.26, 14.30 ± 0.28, 14.21 ± 0.28, 14.37 ± 0.13 and 13.77 ± 0.14, 14.34 ± 0.10, 13.80 ± 0.11, 14.24 ± 0.33, 13.99 ± 0.33, 14.27 ± 0.14, 13.95 ± 0.11, respectively on day 75 compared to those on the day of preparation.

The full fat frankfurter showed significantly lower (P< 0.05) protein content among all formulations from d 45 onwards in both AP and AP systems.

A significant increase (P< 0.05) in the protein content in all formulations except 30/0 in both systems of packaging was noticed.

With respect to the effect of packaging, vacuum packaged 10/5 and 10/10 formulations showed significantly lower (P< 0.05) protein content than their counter parts in the AP system on d 75.

4.4.4.3. Fat

The effect of packaging and period of storage on the fat content of LFF stored at -20^{o}C are presented on Table 22 and illustrated in Fig. 26.

The fat content of aerobically packaged samples of formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5 and 5/10 stored at -20^{o}C increased to 26.82 ± 0.25, 10.15 ± 0.34, 5.58 ± 0.14, 9.83 ± 0.22, 10.10 ± 0.25, 5.84 ± 0.13, 6.16 ± 0.14, respectively on

Table 21. Effect of packaging and period of storage on protein content of LFF stored at -20^{o}C

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	12.15± 0.35	12.19± 0.35	12.24± 0.35	12.44^{a} ±0.34	12.59^{a} ±0.35	12.68^{a} ±0.35	12.15± 0.35	12.20± 0.35	12.26± 0.36	12.42^{a} ±0.34	12.5^{a} ±0.34	12.61^{a} ±0.35
10/0	12.46^{A} ±0.28	12.62^{A} ±0.27	12.80^{AB} ±0.26	13.15^{abABC} ±0.25	13.48^{bBC} ±0.25	13.86^{bC} ±0.26	12.46^{A} ±0.28	12.59^{A} ±0.28	12.76^{AB} ±0.29	13.05^{abAB} ±0.27	13.40^{bB} ±0.26	13.71^{bB} ±0.26
5/0	12.52^{A} ±0.28	12.72^{A} ±0.28	12.90^{A} ±0.29	13.34^{bAB} ±0.28	13.79^{bBC} ±0.28	14.30^{bC} ±0.28	12.52^{A} ±0.28	12.69^{A} ±0.28	12.83^{AB} ±0.28	13.23^{bAB} ±0.29	13.63^{bBC} ±0.27	14.21^{bC} ±0.28
10/5	13.01^{A} ±0.13	13.13^{A} ±0.14	13.28^{AB} ±0.15	13.63^{bBC} ±0.17	13.95^{bCD} ±0.18	14.37^{bD} ±0.13	13.01^{A} ±0.13	13.10^{A} ±0.15	13.17^{AB} ±0.16	13.37^{bABC} ±0.15	13.58^{bBC} ±0.15	13.77^{bC*} ±0.14
10/10	13.09^{A} ±0.10	13.22^{A} ±0.10	13.35^{AB} ±0.11	13.67^{bBC} ±0.13	13.96^{bC} ±0.15	14.34^{bD} ±0.10	13.09^{A} ±0.10	13.16^{A} ±0.11	13.22^{A} ±0.12	13.41^{bAB} ±0.12	13.60^{bBC} ±0.11	13.80^{bC*} ±0.11
5/5	12.77^{A} ±0.32	12.89^{A} ±0.32	13.04^{A} ±0.32	13.38^{bAB} ±0.34	13.76^{bAB} ±0.36	14.24^{bB} ±0.33	12.77^{A} ±0.32	12.86^{AB} ±0.33	12.95^{AB} ±0.34	13.30^{bBC} ±0.34	13.62^{bCD} ±0.34	13.99^{bD} ±0.33
5/10	12.85^{A} ±0.14	12.99^{A} ±0.14	13.15^{AB} ±0.13	13.53^{bBC} ±0.16	13.86^{bCD} ±0.17	14.27^{bD} ±0.14	12.85^{A} ±0.14	12.93^{AB} ±0.14	13.01^{AB} ±0.14	13.32^{bBC} ±0.13	13.65^{bCD} ±0.14	13.95^{bD} ±0.11

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference ($P < 0.05$).

* represents significant difference between two packaging systems in the same time period ($P < 0.05$).

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5**-5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF**- Low fat frankfurter.

Table 22. Effect of packaging and period of storage on fat content of LFF stored at -20ºC

Formulations (Fat/PSC)	Aerobic packaging						Vacuum packaging					
	Period of storage, d						Period of storage, d					
	0	**15**	**30**	**45**	**60**	**75**	**0**	**15**	**30**	**45**	**60**	**75**
30/0	25.79^{cA} ±0.24	25.85^{cA} ±0.24	25.91^{cA} ±0.24	26.18^{cAB} ±0.23	26.49^{cAB} ±0.22	26.82^{cC} ±0.25	25.79^{cA} ±0.24	25.85^{cA} ±0.25	25.95^{cA} ±0.24	26.17^{cAB} ±0.24	26.52^{cAB} ±0.25	26.76^{cB} ±0.26
10/0	9.85^{b} ±0.33	9.90^{b} ±0.33	9.93^{b} ±0.34	10.02^{b} ±0.33	10.10^{b} ±0.33	10.15^{b} ±0.34	9.85^{b} ±0.33	9.90^{b} ±0.34	9.92^{b} ±0.35	10.01^{b} ±0.33	10.08^{b} ±0.34	10.14^{b} ±0.35
5/0	5.30^{a} ±0.13	5.36^{a} ±0.14	5.41^{a} ±0.14	5.47^{a} ±0.14	5.52^{a} ±0.14	5.58^{a} ±0.14	5.30^{a} ±0.13	5.36^{a} ±0.14	5.40^{a} ±0.15	5.44^{a} ±0.14	5.48^{a} ±0.14	5.53^{a} ±0.14
10/5	9.57^{b} ±0.22	9.63^{b} ±0.21	9.67^{b} ±0.21	9.73^{b} ±0.21	9.77^{b} ±0.22	9.83^{b} ±0.22	9.57^{b} ±0.22	9.61^{b} ±0.22	9.66^{b} ±0.22	9.78^{b} ±0.22	9.89^{b} ±0.21	10.04^{b} ±0.22
10/10	9.82^{b} ±0.25	9.86^{b} ±0.25	9.90^{b} ±0.25	9.95^{b} ±0.25	10.02^{b} ±0.25	10.10^{b} ±0.25	9.82^{b} ±0.25	9.85^{b} ±0.25	9.88^{b} ±0.25	9.98^{b} ±0.25	10.08^{b} ±0.23	10.18^{b} ±0.21
5/5	5.38^{a} ±0.14	5.48^{a} ±0.14	5.55^{a} ±0.13	5.65^{a} ±0.13	5.73^{a} ±0.13	5.84^{a} ±0.13	5.38^{a} ±0.14	5.43^{a} ±0.14	5.50^{a} ±0.14	5.56^{a} ±0.13	5.63^{a} ±0.13	5.74^{a} ±0.14
5/10	5.79^{a} ±0.14	5.86^{a} ±0.15	5.91^{a} ±0.15	5.98^{a} ±0.15	6.07^{a} ±0.14	6.16^{a} ±0.14	5.79^{a} ±0.14	5.85^{a} ±0.15	5.89^{a} ±0.15	5.92^{a} ±0.16	5.97^{a} ±0.15	6.03^{a} ±0.17

Means bearing same alphabets in the same column (a, b, c...) and same row (A, B, C...) do not indicate significant difference ($P < 0.05$).

PSC- Pork Skin Collagen; **30/0** - 30% full fat; **10/0** -10% low fat; **5/0** - 5% low fat; **10/5** - 10% fat with 5% PSC; **10/10** -10% fat with 10% PSC; **5/5-** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC; **LFF-** Low fat frankfurter.

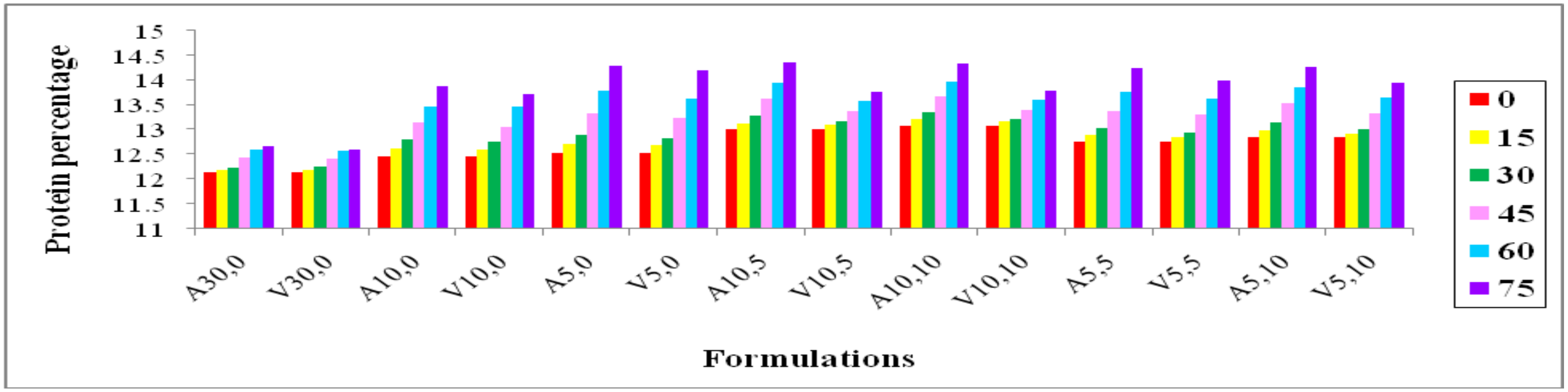


Fig. 25. Effect of packaging and period of storage on protein content of LFF stored at -20ºC

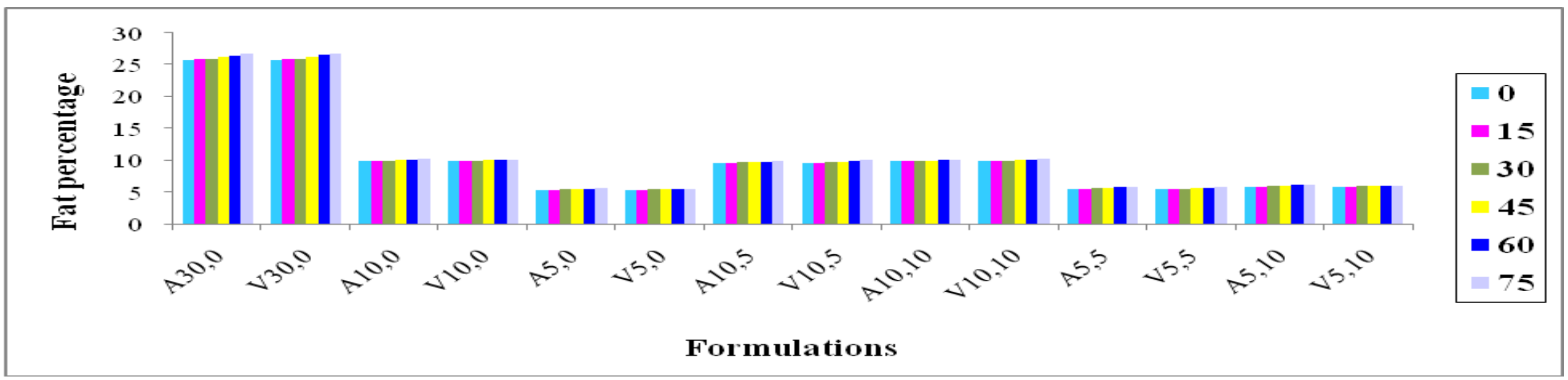


Fig. 26. Effect of packaging and period of storage on fat content of LFF stored at -20ºC

30,0-30% full fat;**10,0-**10% low fat;**5,0-**5% low fat; **10,5-**10% fat with 5% PSC; **10,10** -10%fat with 10% PSC; **5,5-** 5% fat with 5% PSC; **5, 10** -5% fat with 10% PSC; **PSC-** Pork Skin Collagen; **LFF**-Low fat frankfurter; **A-**Atmospheric packaging; **V**-Vacuum packaging.

day 75 compared to those on the day of preparation. The respective values for same formulations packed under vacuum were 26.76 ± 0.26, 10.14 ± 0.35, 5.53 ± 0.14, 10.04 ± 0.22, 10.18 ± 0.21, 5.74 ± 0.14, 6.03 ± 0.17 respectively on day 75.

Under AP system on all days of storage full fat control showed significantly higher ($P < 0.05$) fat content than rest of the formulations. There was no significant difference ($P > 0.05$) in the fat content of formulations 10/0, 10/5 and 10/10 were significantly higher ($P < 0.05$) than 5/0, 5/5 and 5/10 in all days of storage. The same was the case with VP also.

Between periods, on d 75 full fat frankfurters showed significantly higher ($P < 0.05$) fat content in both types of packaging systems. The packaging systems did not show any significant difference ($P > 0.05$) on the fat content through out the entire storage period of 75 days.

4.5. COST OF PRODUCTION

The cost of production of one kg Low Fat Frankfurter (LFF) of different formulations is presented in Table 23.

Table 23. Cost of production of different formulations of frankfurters

Formulations (Fat/PSC)	**Cost of Production/kg (Rs)**
30/0	92
10/0	81
5/0	81
10/5	84
10/10	86
5/5	88
5/10	86

PSC- Pork Skin Collagen; **30/0 -** 30% full fat; **10/0** -10% low fat; **5/0 -** 5% low fat; **10/5 -** 10% fat with 5% PSC; **10/10 -** 10% fat with 10% PSC; **5/5 -** 5% fat with 5% PSC; **5/10** -5% fat with 10% PSC.

Discussion

DISCUSSION

The study on the quality and shelf life of low fat frankfurters (LFF) were taken up with the objectives of developing a new formulary for low fat frankfurter using pork skin collagen (PSC) gel as a fat replacer by assessing its physico-chemical, cooking characteristics, Warner-Bratzler Shear Force Value (WBSFV), colour by Hunter L^*, a^*, b^* values, proximate composition, nutritive value, organoleptic qualities and the shelf life studies by assessing the purge loss, 2-Thiobarbituric Acid Reactive Substances (TBARS) Value, organoleptic qualities and moisture, protein and fat contents of frankfurters under aerobic packaging (AP) and vacuum packaging (VP) on storage at -20^{o}C. Seven different formulations of frankfurter were prepared, viz., one full fat control with out PSC (30/0), six treatments, 10 per cent low fat with out PSC (10/0), 5 per cent low fat with out PSC (5/0), 10 per cent fat with 5 per cent PSC (10/5), 10 per cent fat with 10 per cent PSC (10/10), 5 per cent fat with 5 per cent PSC (5/5), 5 per cent fat with 10 per cent PSC (5/10). The results are discussed in this chapter.

5.1. PHYSICAL CHARACTERISTICS OF LFF

5.1.1. pH

The pH of uncooked batter of different formulations of frankfurter, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 before cooking ranged from 6.12 to 6.38. The pH of the batter of LFF 10/10 and 5/10 were significantly higher ($P < 0.05$) than those with out PSC.

The addition of pork skin collagen increased the pH values. Similar results were obtained by Osburn *et al*. (1997) in reduced fat bologna. They opined that increased pH of collagen resulted in higher pH of the batter. Osburn and Mandigo (1998) in reduced fat bologna manufactured with poultry skin connective tissue gel and Eilert *et al*. (1993) in meat batters manufactured with modified beef connective tissue also reported similar observations. They reported an increase in pH from 5.82 to 5.99 and attributed it to the higher pH of the connective tissue.

The uncooked batter of full fat control and low fat treatments with out the addition of fat replacer did not differ significantly in pH values. This indicated that reduction of fat without incorporating any fat replacer did not affect the pH values. This was in agreement with Nazeera (2007) in low fat restructured turkey loaves.

The pH of cooked frankfurters of different formulations, viz., 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 ranged from 6.27 to 6.55. Compared to the uncooked batter, the pH of cooked frankfurters of all formulations increased significantly ($P < 0.05$) upon thermal processing. This was in agreement with the findings of Osburn *et al*. (1997) in pork skin connective tissue gel utilization in reduced fat bologna where the pH ranged from 6.87 in uncooked batter to 7.50 in cooked bologna. Beggs *et al.* (1997) in reduced fat turkey frankfurter observed a pH value of 5.97 to 6.24 for batter formulated with corn starch and water and 6.29 to 6.39 for the cooked frankfurter. Similar results were obtained by Schnell (1999) while using raw and pre heated pork skin collagen in low fat bologna.

An increase in pH after thermal processing was also observed by Claus and Hunt (1991) in low fat, high added water bologna formulated with texture modifying agents and Naveen *et al*. (2006) in partially cooked duck meat sausages. They suggested the reason for higher pH as inactivation of enzymes during thermal processing.

5.1.2. Emulsion Stability

The ES, in percentage, of 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and 5/10 ranged from 77.48 to 87.40. The formulations 10/5, 10/10, 5/5 and 5/10 had significantly higher ($P < 0.05$) emulsion stability values than full fat control. Similar results were obtained by Eilert *et al*. (1993) wherein increasing the level of fat caused less emulsion stability and addition of connective tissue helped to create a collagen myofibrillar protein matrix that bound water and fat better thus increasing the emulsion stability. Satterlee *et al.* (1973) also agreed with this concept of addition of collagen causing increased emulsion stability by increasing water binding and fat emulsification. High pH value of 10/10 formulation caused high emulsion stability. There was a positive correlation between pH and emulsion stability and expressible moisture. This was in agreement with the findings of Schnell (1999) in low fat bologna with pre gelatinized pork skin collagen.

The full fat control had significantly lower ($P < 0.05$) emulsion stability than the treated formulations. This was in order with the findings of Martinez *et al*. (2004) in pork frankfurters where 20 per cent fat frankfurter showed significantly lower emulsion stability values when compared with 9 per cent fat frankfurter. Eilert *et al.* (1993) reported decreased emulsion stability in 24 per cent fat than 8 per cent fat in meat batters manufactured with modified beef connective tissue.

5.1.3. Cooking Characteristics

5.1.3.1. Cook Yield and Cook Loss

Among different treatment combinations, the significantly highest ($P < 0.05$) cook yield of 96.28 ± 0.08 per cent was noticed for formulation 10/10 followed by formulation 10/5. There was significant reduction ($P < 0.05$) in the cook yield of full fat control. All the low fat formulations with/without addition of fat replacer had significantly higher ($P < 0.05$) cook yield than the full fat control. The cook loss showed a negative correlation with that of cook yield.

This was in agreement with the findings of Shackelford *et al.* (1991) who reported that 10 per cent fat formulation of pork frankfurters had a higher yield than 20 and 25 per cent fat formulations. Similarly, El-Magoli *et al*. (1996) observed an improved yield of low fat ground beef patties on reduction in the fat level from 22 to 11 per cent. Serdaroglu and Degirmencoglu (2004) also reported that the cook yield was lowest for 20 per cent fat than 10 and 5 per cent fat in turkish meat balls and attributed it to the excess fat separation and water release during cooking. Sheard *et al*. (1998) also agrees with these findings. They observed that sausages and burgers with high fat content lost more fat regardless of the cooking method followed.

Significant improvement ($P < 0.05$) in cook yield due to the addition of increasing levels of pork collagen in low fat frankfurters in the present study was in agreement with the findings of Arganosa *et al*. (1987) with 10 per cent collagen in pork sausages. Prabhu *et al*. (2004) reported increased cook yield when pork collagen content was increased from 0 to 3 per cent in emulsified and whole muscle meat products. Similar results were obtained in a study conducted by Abiola and Adegbaju (2001) in pork sausages.

The reduction in cook loss with subsequent increase in the cook yield could be attributed to the higher protein content of low fat treated formulations and the protein functionality of the collagen molecule by its ability to trap moisture and reduce moisture loss. On heating, gelation of protein entraps water and fat and stabilises the sausages. Also collagen work synergistically with myofibrillar structures in meat proteins to bind water. Moreover the positive correlation between pH and processing yield caused higher cook yield in 10/10 frankfurter formulation.

5.1.3.2. Dimensional Shrinkage

The DS, in percentage, of formulations 30/0, 10/0, 5/0, 10/5, 10/10, 5/5, and

5/10 were ranged from 1.83 to 3.95. The full fat control and formulation 10/10 showed the significantly highest value of 3.95 and the lowest value of 1.83 per cent among all formulations.

This was in total agreement with the findings of El-Magoli *et al*. (1996) who reported that reduction in fat level from 22 to 11 per cent caused reduced shrinkage in low fat ground beef patties and Troy *et al*. (1999) reported that full fat beef burger shrunk the most due to high fat loss and moisture during cooking. Similar results were reported by Serdaroglu and Degirmencoglu (2004) in turkish meat balls formulated with 20 per cent fat, which had the highest reduction in diameter.

The reduction in the DS value of 10/10 formulation could be attributed to its lower fat content and the collagen functionality of water binding and fat emulsification due to its higher collagen content.

5.1.4. Water Holding Capacity

Formulation 10/10 had significantly the highest ($P< 0.05$) WHC of all treatments. Cooking at 73 - 75^{o}C enhanced the water binding capacity. Heating at 70^{o}C for 30 min was sufficient to enhance the water binding ability of pork skin connective tissue and more water was bound at 70^{o}C (Osburn *et al.,* 1997). Schnell (1999) also reported that the expressible moisture per cent reduced from 31.9 per cent with no pork skin gel preparation to 15.3 per cent with 10 per cent pork skin gel preparation in low fat bologna. Similar results were reported by Osburn *et al*. (1997) in reduced fat bologna with pork skin collagen and in reduced fat bologna with pre-heated chicken connective tissue (Osburn and Mandigo, 1998). These results were also in agreement with the findings of Webster *et al.* (1982) who reported the ability of collagen to increase the water holding capacity in processed products.

Similarly, the high pH of 10/10 formulation increased the batter viscosity and increased water holding capacity of the product. There existed a positive correlation between pH and water holding capacity due to high emulsion stability and less expressible moisture.

The full fat control had higher WHC than low fat treatments with out fat replacers. This was in consistent with the findings of Colmenero *et al*. (1996) in bologna sausages, Hughes *et al*. (1997) in frankfurters and Yoo *et al*. (2007) in comminuted sausages.

5.1.5. Warner-Bratzler Shear Force Value (WBSFV)

The shear force of 10/10 was 1.12 kgf followed by 1.14 in 5/10 which were significantly lower ($P < 0.05$) than the low fat treatments with out PSC but higher than full fat control. Although the fat levels had been reduced to 10 per cent and 5 per cent in 10/10 and 5/10 formulations, respectively, the toughness did not increase. This indicated that incorporation of pork skin collagen at 10 per cent level significantly reduced the shear force of low fat frankfurters by improving the textural characteristics and increasing tenderness of the product.

Meullenet *et al*. (1994) reported that increasing the level of collagen fibres to 5 per cent lowered shear stress values in low fat high added water chicken frankfurters. Similar observations were made by Arganosa *et al*. (1987) who replaced 20 per cent fat with collagen in pork sausages. In reduced fat bologna, Osburn *et al*. (1997) and Osburn and Mandigo (1998), observed improved water binding and gelling ability of collagen, which might have improved the texture by diluting the stronger binding myofibrillar protein in low fat formulations.

5.1.6. Colour

The Hunter L^* and b^* values for lightness and yellowness were 62.99 ± 0.23 and 17.51 ± 0.11 for formulation 10/10, which were significantly higher ($P< 0.05$) and a^* value for redness was 5.70 ± 0.03 which was significantly lower ($P< 0.05$) among treatments. Similar results were obtained by Prabhu *et al*. (2004) who reported that use of collagen increased L^* and b^* values in frankfurters and hams. Similarly Eilert *et al*. (1996) and Osburn *et al*. (1997) reported that pork skin gel incorporation into bologna increased L^* and decreased a^* values. This was in agreement with the findings of Schilling *et al*. (2003) who reported addition of light coloured collagen diluted the dark colour of low fat products.

The L^* value of 10/10 formulation did not differ significantly from full fat control. This was in consistent with the results obtained by Arganosa *et al*. (1987) who reported the same trend with 10 per cent replacement of fat with collagen in pork sausage.

Low fat frankfurters without addition of pork skin collagen had darker colour. This was in agreement with the findings of Abiola and Adegbaju (2001) who reported that low fat pork sausages were darker than high fat pork sausages.

5.2. PROXIMATE COMPOSITION

5.2.1. . Lean Beef and Pork Trimmings, Their Combinations and Pork Skin Collagen

Proximate analysis of LBT and LPT revealed that the former contained 77.30 ± 0.30, a significantly higher content of moisture compared to latter. The ash content 1.39 ± 0.04, of LPT was significantly higher than that of LBT. The protein content of the PSC 25.77 ± 0.22 was significantly the highest ($P< 0.05$) of all.

5.2.2. Low Fat Frankfurters

5.2.2.1. Uncooked frankfurter batter

The proximate analysis revealed that the moisture and protein content were significantly higher for all the low fat treatments than the full fat control. Addition of PSC at 5 per cent level to 10 per cent low fat formulation significantly increased the protein content from 10.40 ± 0.28 to 11.87 ± 0.20. Addition of PSC at 10 per cent level to 10 per cent low fat formulation significantly increased the protein content from 10.40 ± 0.28 to 12.31 ± 0.17 compared to 10/0 low fat formulation.

The data on moisture and fat depicted an inverse relationship between fat and moisture. Similar findings were also reported by various other research workers in a wide variety of low fat meat products like low fat pork frankfurters (Shackelford *et al.*, 1991), low fat emulsified pork meat balls (Hsu and Chung, 2001), turkish type meat balls (Serdaroglu and Degirmencioglu, 2004), low fat pork patties (Kumar and Sharma, 2004) and low fat meat balls made of beef (Serdaroglu, 2006).

5.2.2.2. Cooked frankfurters

Moisture content of cooked frankfurters of all formulations was significantly lower than that in uncooked batter. The moisture content in 10/10 formulation of LFF was 71.30 ± 0.36. The moisture content in all formulations is within the USDA regulations of 4 times the protein content plus 10 per cent for cooked sausages Henrickson (1978). The moisture loss due to cooking was significantly lower for 10/10 formulation. Osburn and Mandigo (1998) also reported the same trend in reduced fat bologna. The protein content of formulation 10/10 was 13.09 ± 0.10, the highest among all owing to the high protein content of PSC. Protein, fat, carbohydrates and ash contents of the formulations were higher than in uncooked batter except the fat content of full fat control. This was due to the loss of moisture and consequently higher dry matter content in cooked LFF. This was in agreement with that of Serdaroglu and Degirmencioglu (2004) in turkish type meat balls, Serdaroglu (2006) in low fat beef balls and Naveen *et al*. (2006) in duck sausages.

5.3. NUTRIONAL VALUE

5.3.1. Calorific Value of Nutrients and Their Per cent Contribution to RDA

The 5/0 formulation had the lowest total calories of 115.86 kcal/100g and full fat control had the highest total calories of 297.43/100g and its energy value of 232.11/100g from fat was significantly higher than all other formulations. The LFF formulation 10/10 had a total calorific value of 88.38 from fat.

The total calorific value in each LFF formulation with PSC ranged from 115.98 to 159.98 kcal/100g. The per cent contribution to RDA of the same ranged from 5.27 to 7.27. The contribution of calories from protein in 10/10 LFF was significantly more than in other formulations.

The contribution of calories from fat to the RDA was below the recommended 30 per cent (NRC, 1989; WHO, 2003) in all frankfurter formulations including the full fat control. The RDAs are used by nutritionists and dietitians as the basis for most public health programmes (Lupton and Cross, 1999).

5.3.2. Per cent Daily Value of Protein

The per cent contribution of protein present in 10/5 and10/10 formulations to the RDA were 21.68 and 21.82, respectively were significantly higher than full fat control. This was due to the presence of PSC gel in the respective formulations.

5.4. EFFECT OF PACKAGING AND STORAGE ON QUALITY OF LFF

5.4.1. Purge Loss

The LFF with out PSC had significantly higher ($P < 0.05$) PL. This was in agreement with the results of Cavestany *et al*. (1994) and Claus *et al*. (1989) in bologna sausages. As the period of storage progressed, irrespective of the formulations and packaging systems PL showed a gradual and significant increase. Similar trend was reported by in the storage stability of low fat chicken sausages (Andres *et al*., 2006) and in low fat frankfurters (Bloukas and Paneras, 1993).

It is clear from the data obtained that addition of PSC either at 5 per cent or at 10 per cent level showed significant decrease (P< 0.05) in PL of the low fat frankfurters, with 10/10 formulation showing the lowest purge followed by 10/5 formulation. This agrees with the findings of Prabhu *et al*. (2004) in both frankfurters and restructured ham, Osburn *et al*. (1997) in reduced fat bologna with pork collagen gel utilisation and Osburn and Mandigo (1998) in reduced fat bologna with poultry skin connective tissue gel utilisation.

The lowest PL in formulation 10/10 was not only due to the addition of collagen that rendered increased emulsion stability, water holding capacity and better binding property which caused less amount of fluid separation during storage but also due to its high protein content. Same inference was reported by Carballo *et al*. (1995) in studies with bologna. Similarly, high pH of 10/10 formulation was correlated with its low purge compared to low fat treatments with out PSC.

5.4.2. 2-TBARS Value

It is clear from the data obtained that the TBARS Value of full fat control was significantly higher (P< 0.05) than other low fat formulations. This could better be attributed to the high fat content of the full fat frankfurter. As the storage period progressed, from d 45 onwards TBARS values of frankfurters irrespective of formulations and packaging systems showed a gradual significant increase (P< 0.05). This finding was in consistent with the observations of Arganosa *et al*. (1987) in pork sausages. Kao and Lin (2006) observed increasing trend in the TBARS values of reduced fat pork frankfurters during storage.

TBARS values of AP samples did not show any significant difference from that of VP samples through out the storage period. This was in agreement with Jo *et al*. (1999) who observed that the TBARS values of cooked pork sausages increased with increase in fat content regardless of storage and packaging types. Nazeera (2007) also revealed that there was no significant difference between the aerobically and vacuum packaged pouches of low fat restructured turkey loaves during storage.

In the present study there was no significant increase ($P < 0.05$) in the TBARS values of low fat frankfurter formulations with PSC during the initial 30 days of storage at -20^{o}C. The increase was evident from d 45 onwards only. This could be attributed to the synergistic antioxidant effect of ingredients like collagen and nonmeat ingredients such as sodium nitrite, sodium ascorbate, spices, onion, ginger and garlic. The proline and hydroxyproline in the collagen had been reported to exert antioxidant property along with spices (Arganosa *et al.*, 1987). The essential oils in the spices have been reported to have antioxidant effect in addition to flavour giving properties (Coggins, 2001). Similar to these findings Pearson and Gillet (1997) reported that sodium nitrite could prevent lipid oxidation in cured meat products. Younathan, *et al*. (1980) found that hot water extracts of onion peel effectively controlled rancidity in cooked ground turkey. They opined that onion peel contains numerous flavone aglycones which act as antioxidants in meat products. Kilic and Richards (2003) reported that sodium ascorbate inhibited lipid oxidation in poultry doner kebab.

In the present study TBARS values determined were lower than the acceptable range of 1mg malonaldehyde/kg for oxidative rancidity as suggested by

Ockerman (1976). Products with a TBARS value less than 1.0 generally do not contain detectable off odour and flavour due to lipid auto oxidation.

5.4.3. Sensory Evaluation on the Day of Preparation and on Storage

The appearance and colour of low fat frankfurters on the day of preparation was significantly improved with the addition of PSC. All the formulations were showing a slightly dark colour except the full fat control which was less dark in colour. The lighter colour of the full fat control was attributed to its high fat content which diluted the myoglobin content in the lean. This finding was in order with Hughes *et al*. (1997) who reported that decreasing fat content caused darker and reddish colour in reduced fat bologna and Abiola and Adegbaju (2001) reported that the high fat pork sausages were less dark than low fat pork sausages.

Formulation 10/10 containing 10 per cent PSC exhibited a desirable light brownish colour than the other treatments. This was in agreement with the findings of Schilling *et al*. (2003), who reported addition of light coloured collagen diluted the dark colour of low fat products. Dickson *et al*. (1995) also reported that additives used to replace fat tend to dilute the colour of meat, poultry and fish items and water binding agents will reduce Mb concentration in low fat products.

The panel score for texture of 10/10 formulation was comparable with the full fat control. The improved sliceability of 10/10 formulation could be attributed to the water binding and texture modifying ability of the pork skin collagen (Osburn *et al*. 1997; Prabhu *et al*. 2004; Schilling *et al*. 2003). The formulation 10/10 was more flavourful. This was in agreement with findings of Sadler and Young (1993) who reported that sausages with pre-heated collagen had more flavour. This was in agreement with the observations of Osburn *et al*. (1997) in reduced fat bologna with pork skin collagen gel utilization.

Formulation 10/10 had high juiciness score. This was in agreement with the findings of Osburn *et al*. (1997) who reported increasing pork connective tissue gel and water addition caused increased juiciness in reduced fat bolognas.

The panel scores for saltiness did not differ among formulations. The saltiness score of 6.47 to 7.22 showed that addition of 1.5 per cent salt produced desirable flavour in the products. There was no mouth coating for any of the formulations including the full fat control.

Among treatments 10/10 formulation scored highest overall acceptability score by having desirable flavour, more juiciness and fine texture attributes which are more acceptable to the panelists.

5.4.3.1. Appearance and Colour

The appearance and colour scores showed no significant difference between formulations on any day of the storage under aerobic packaging. On d 75, 10/10 formulation and full fat control were adjudged equally among all formulations. Formulation 10/10 under AP ranked at the same level on d zero and on d 75.

The deterioration in the colour of meat products on storage generally is as an indication of microbial spoilage. The visual observations indicated that frankfurter of all formulations did not undergo any microbial spoilage to cause an appreciable change in the appearance and colour.

5.4.3.2. Flavour

The 10/10 formulation did not differ significantly during its storage period. On d 75 under both systems of packaging, no significant difference was observed between the PSC added formulations. There was no significant difference between days of storage of these formulations. This showed the role of PSC in retaining the flavour of the product on storage. Sadler and Young (1993) also reported the same findings in their study on fine emulsion sausages with preheated tendons.

The flavour scores of full fat control improved under vacuum through out the storage period of 75 days. Similarly, formulations 10/10 and 10/5 scored higher than its counterparts under AP on d 75. This revealed the benefit of VP in retaining the flavour volatiles. Harte (1987) reported that polyamide (nylon) polymers like PA/PE possess good oxygen and aroma barrier properties.

During the course of study and sensory evaluation, no off flavour due to oxidative rancidity, fermentation, spoilage or addition of fat replacers was detected in any of the formulations.

5.4.3.3. Texture

During the entire storage period formulation 10/10 was ranked top among treatments, indicated that this formulation was desired by the panelists throughout the storage period. The improvement of the texture score of 10/10 formulation under AP indicated that the gel formed was stabilised by cold storage. In general the texture of PSC treatments were ranked acceptable through out the storage period. There was no significant effect on the texture of frankfurter formulations between days of storage period as well as between AP and VP.

5.4.3.4. Saltiness

There was a slight increase in the saltiness from d zero during the storage period in aerobically packaged samples compared to the samples in vacuum pouches irrespective of the formulations. This could be attributed to comparatively higher moisture loss and the consequent higher salt concentration in aerobically packaged samples. But the saltiness score was moderate to very desirable throughout the study irrespective of the formulations both under AP and VP. Nazeera (2007) reported the same trend in low fat restructured turkey loaves stored at -20^{o}C.

5.4.3.5. Juiciness

The formulation 10/10 remained high on juiciness followed by 10/5 in both types of packaging systems. This was previously indicated by Sadler and Young (1993) in fine emulsion sausages in which he attributed it to the water binding capacity of collagen there by retaining the succulence. All other formulations rated next to these two formulations.

5.4.3.6. Mouth Coating

The different packaging systems and period of storage did not affect the scores of mouth coating. Under VP on d 30 and 60, formulations 10/5 and 10/10 showed higher score among all formulations.

5.4.3.7. Overall Acceptability

The low fat formulations with out addition of PSC were ranked moderately acceptable among treatments. Both under AP and VP systems 10/10 followed by 10/5 scored significantly higher overall acceptability score among treatments.

The very acceptable nature of 10/10 formulation might be due to its moderate to very desirable flavour, texture and moderate to very juiciness. This was previously reported by Osburn *et al*. (1997) who used poultry connective tissue in reduced fat bologna and attributed the water binding and texture modifying characteristic of collagen molecules as reason for the betterment of key sensory traits like flavour, texture and juiciness in low fat meat products. This indicated PSC can be used as a fat replacer in meat products which are intended for a prolonged storage.

5.4.4. Proximate Composition

5.4.4.1. Moisture

The moisture content of 10/5 and 10/10 formulations decreased significantly from 71.89 and 70.91 on d 30 to 70.52 and 69.61 respectively on d 75 in aerobic packaging. Similarly in vacuum packaging also it significantly reduced to 70.97 and 70.08, respectively though the rate of decrease was less in vacuum packaging.

Similar trend in moisture content was noticed by Naveen *et al.* (2006) who reported that partially cooked duck meat sausages on storage at -18 ± 1°C showed a decreasing trend in moisture percentage from 49.3 on zero day to 40.6 by the end of 8th week. Similarly Nazeera (2007) reported the same decreasing trend in the moisture content of low fat restructured turkey loaves during its storage.

5.4.4.2. Protein

Formulations 10/5 and 10/10 showed significant increases in protein content on d 60 to d 75 irrespective of the packaging systems.

This increase in the protein content was consequent to the decrease in moisture content of the samples and subsequent increase in dry matter content. The extent of increase was slightly more in aerobically packaged samples compared to VP samples because the moisture barrier property of PA/PE packages used for vacuum packing might be more than that of the HDPE used for aerobic packaging. Naveen *et al*. (2006) also reported that frozen storage of raw and partially cooked duck meat sausages showed an increasing trend in per cent protein up to 6th week due to the decrease in the moisture content of the samples. The findings of Nazeera (2007) in quality and shelf life study of low fat restructured turkey loaves also agreed with this.

Vacuum packaged formulations of 10/5 and 10/10 on d 75 showed significantly lesser protein content than its counter parts in aerobic packaging. Higher protein content in aerobic packaging might be due to increased moisture loss with corresponding increase in dry matter percentage.

5.4.4.3. Fat

The fat content of full fat control showed significant increase on d 75 irrespective of the packaging systems. Similar increase in fat percentage was reported by Naveen *et al.* (2006) in duck meat sausages and Nazeera (2007) in low fat restructured turkey loaves due to moisture loss.

The formularies for low fat frankfurters with 10 and 5 per cent PSC as fat replacers were developed economically with good overall acceptability, cooking yield, nutritional quality and shelf life up to 75 days at -20^{o}C under AP and VP systems. The cost of production was calculated on laboratory scale and further investigations with large quantity are required for commercial production at industrial level.

Summary

SUMMARY

High fat intake is associated with increased risk for obesity, colon cancer, high blood cholesterol and coronary heart disease. Therefore, health conscious meat consumers prefer low fat meat products. However, fat contributes to nutritional and sensory properties of meat products and reducing fat content results in decrease in these properties. Manufacturing meat products with fat replacers provide greatest opportunity to reduce fat and to alleviate the problems encountered with the reduction of fat. Therefore, the present study was undertaken with the objectives of developing a suitable formulary for low fat frankfurters with pork skin collagen (PSC) gel as a fat replacer and to assess its physico-chemical, cooking characteristics, proximate composition, nutritional value, organoleptic qualities and shelf life under aerobic (AP) and vacuum packaging (VP) at -20^{o}C and their cost of production.

Emulsion for the frankfurters was prepared by grinding, mixing and chopping the minced lean beef and pork trimmings, lard, salt, sugar, sodium nitrite, sodium ascorbate, sodium tripolyphosphate, ground spices, onion, ginger, garlic paste, refined wheat flour and crushed ice. These emulsions were used to prepare seven different formulations with/without the fat replacer PSC, viz., one full fat control 30% (30/0) and six treatments 10% low fat with out PSC (10/0), 5% low fat with out PSC (5/0), 10% fat with 5% PSC (10/5), 10% fat with 10% PSC (10/10), 5% fat with 5% PSC (5/5), 5% fat with 10% PSC (5/10), respectively.

All seven emulsions were then stuffed into sheep casings (18 mm to 20 mm diameter) using manually operated stuffer and twist linked manually. Sausages were steam cooked under atmospheric pressure to a core temperature of 73^{o}C-75^{o}C. All the formulations were packaged under AP and VP systems in high density poly ethylene (HDPE) and polyamide/polyethylene (PA/PE) pouches, respectively and

stored at –20^{o}C for 75 days for further studies.

Various quality parameters of the frankfurters, viz., pH, emulsion stability, cook yield, cook loss, dimensional shrinkage, water holding capacity (WHC), Warner-Bratzler Shear Force Value (WBSFV), colour by Hunter L^*, a^*, b^* values, proximate composition of trimmings of beef, pork, their 1:1 mix, pork skin collagen gel, uncooked batter and cooked frankfurter, nutritive value, 2-Thiobarbituric Acid Reactive Substances Value (TBARS) and organoleptic qualities using an eight point Hedonic scale were studied on the day of preparation of the low fat frankfurters. The purge loss, TBARS value, organoleptic qualities and moisture, protein and fat contents of frankfurters were studied again on d 15, 30, 45, 60 and 75 of storage at -20^{o}C. Six trials of the experiment were conducted and the cost of production was also calculated.

The pH of the cooked low fat frankfurters with PSC was significantly more (P< 0.05) than those with out PSC, their uncooked batter and in the range of 6.46-6.55 and could be considered a low acid food. Increased pH of collagen resulted in the higher pH of the batter and frankfurters. The LFF formulation 10/10 had the significantly the highest (P< 0.05) emulsion stability of 87.40, cook yield of 96.28 and significantly the lowest (P< 0.05) dimensional shrinkage of 1.83 percentage. The higher pH and protein content in this formulation was very effective in improving the emulsion stability, retaining moisture, fat and thus improving the cooking yield and reducing the shrinkage.

The WHC of 95.57 of formulation 10/10 was significantly higher (P< 0.05) than other treatments and comparable with the full fat control. There existed a positive correlation between pH and water holding capacity due to high emulsion stability and less expressible moisture. The WBSFV of 10/10 was 1.12 kgf which was significantly lower ($P< 0.05$) than other treated formulations. Improved water binding and gelling ability of collagen might have improved the texture by diluting the stronger binding myofibrillar protein in low fat formulations.

The formulation 10/10 had a Hunter L^* value of 62.99 which was comparable with the full fat frankfurter and a^* and b^* values of 5.70 and 17.51 which were significantly higher and lower ($P< 0.05$), respectively than full fat control. Addition of light coloured collagen diluted the dark colour of low fat products.

The moisture, protein, fat, carbohydrate and ash contents in the 1:1 mix of beef and pork trimmings were 76.67, 19.13, 1.98, 1.01, 1.21, respectively and in the PSC they were 72.67, 25.77, 0.60, 0.38, 0.58, respectively. The percentages of moisture, protein, fat, carbohydrate and ash in 10/5 and 10/10 were 72.32, 13.01, 9.57, 4.17, 0.93 and 71.30, 13.09, 9.82, 4.81, 0.98, respectively. Steam cooking under atmospheric pressure caused significant reduction in moisture with corresponding increase in other proximate principles in low fat formulations with or with out PSC. The moisture content in all formulations was within the stipulations of USDA for cooked sausages. The level of all nonmeat ingredients in all frankfurter formulations is within the USDA regulations.

The total calorific value in 10/10 formulation was 159.98 kcal/100g and contribution to this from fat was only 88.38 kcal. The same in 10/5 was 154.85 kcal/100g and the contribution from fat was 86.13 kcal. The per cent contribution to the Recommended Daily Allowance (RDA) of 2200 kcal from 10/10 was 7.27 and 4.02, respectively. The same in 10/5 was 7.05 and 3.92, respectively. The contribution of calories from fat to the RDA was below the recommended 30% in all frankfurter formulations. The per cent contribution of protein in 10/10 and 10/5 formulations to the RDA were 21.82 and 21.68, respectively. This was due to the presence of PSC gel in the respective formulations.

On assessment of the shelf life of the frankfurters, the purge loss of 10/10 and 10/5 formulations were found significantly lower ($P < 0.05$) irrespective of the packaging systems and period of storage. This may be due to the increased water holding capacity of the PSC added to these formulations. The TBARS value of LFF with varying levels of PSC gel were within the acceptable range of 1mg malonaldehyde/kg of frankfurter for oxidative rancidity, irrespective of the packaging systems and period of storage. There was no significant increase in the oxidative rancidity of PSC added formulations noticed during the initial 30 days of storage. This oxidative stability of the product is attributed to the synergistic antioxidant effect of proline and hydroxyproline in collagen, sodium nitrite, sodium ascorbate and essential oils in spices, onion, ginger and garlic. Pork skin along with other ingredients had significant influence on reducing lipid oxidation during the initial days of storage period. But there was a significant increase ($P < 0.05$) in the TBARS value of all formulations as time progressed in the frozen storage irrespective of the packaging systems.

On organoleptic evaluation, the appearance and colour of formulation 10/10 was rated as very acceptable and comparable with the full fat control. Neither the reduction of fat nor the addition of PSC adversely affected the appearance, colour, flavour, saltiness, and juiciness of LFF. The formulation of 10/10 had the same textural effect of full fat frankfurter. The addition of PSC comparatively increased the sliceability due to its water binding and gel forming ability. The appearance, colour, flavour, texture, juiciness, saltiness, mouth coating and overall acceptability of 10/10 were very desirable and comparable with that of full fat control.

In both AP and VP systems, low fat frankfurters with PSC as a fat replacer on storage at -20°C for 75 days did not significantly affect the appearance, colour, flavour, texture, juiciness, saltiness, mouth coating and overall acceptability. Formulation of 10/10 had very desirable overall acceptability score among treatments and comparable with full fat control through out the storage period. Formulation 10/10 under vacuum packaging had a more beneficial effect in retaining flavour, texture, and juiciness followed by formulation 10/5 during the 75 days of storage at -20°C.

The period of storage had not affected the proximate composition of any of the formulations except for a significant reduction ($P < 0.05$) in the moisture on d 60 onwards and a significant increase ($P < 0.05$) in the protein content of all formulations treated with PSC on d 45 onwards. Vacuum packaging system significantly reduced the protein content of 10/5 and 10/10 formulations on d 75 of storage at -20°C. The cost of production calculated per kg of 10/5 and 10/10 were Rs.84/= and Rs.86/=, respectively under laboratory scale.

The formularies for LFF 10/10 and 10/5 were developed with very acceptable organoleptic attributes, cook yield and shelf life up to 75 days at -20°C, respectively under aerobic and vacuum packaging systems economically. The contribution of calories from fat to the RDA was below the recommended 30% in all frankfurter formulations making the product acceptable to health conscious consumers. The cost of production was calculated on laboratory scale and further investigations with large quantity are required for commercial production at industrial level.

Reference

REFERENCES

Abiola, S.S. and Adegbaju, S.W. 2001. Effect of substituting pork backfat with rind on quality characteristics of pork sausage. *Meat Sci.* 58: 409-412

*AHA. 1996. Dietary guidelines for healthy Americans. American Heart Association. Circulation. 94: 1795-1800

Akoh, C.C. 1988. Fat replacers. *Food Technol.* 52 (3): 47-52

Allen, P., Dreeling, N., Desmond, E.M., Hughes, E., Mullen, A.M. and Troy, D. 1999. New Technologies in the manufacture of low fat meat products. Final report. Project Armis No.4038. The National Food Centre, Castleknock, Dublin

Almeida-Dominguez, N.G., Higuera-Ciapara, I., Goycoalea, F.M. and Valencla, M.E. 1992. Pack, temperature and TBHQ effects on oxidative deterioration of corn-based snacks. *J. Food Sci*. 57: 112-117

AMSA, 1983. Guidelines of sensory, physical and chemical measurements in ground beef. *Recip. Meat Conf. Proc.* 36: 221-228

Andres, S.C., Garcia, M.E., Zaritzky, N.E. and Califano, A.N. 2006. Storage stability of low fat chicken sausages. *J. Food Eng.* 72: 311-319

AOAC, 1990. Meat and meat products. *Official Methods of Analysis of Analytical Chemists*, Fifteenth edition. Association of Official Analytical Chemists. Washington D.C., p. 587

Arganosa, G.C., Henrickson, R.L. and Rao, B.R. 1987. Collagen as a lean or fat replacement in pork sausage. *J. Food Qual.* 10: 319-333

Baliga, B.R. and Madaiah, N. 1970. Quality of sausages emulsion prepared from mutton. *J. Food Sci.* 35 (4): 383-385

Beggs, K.L.H., Bowers, J.A. and Broven, D. 1997. Sensory and physical characteristics of reduced fat turkey frankfurters with modified corn starch and water. *J. Food Sci.* 62: 1240-1244

Bloukas, J.G. and Paneras, E.D. 1993. Substituting olive oil for pork back fat affects quality of low fat frankfurters. *J. Food Sci.* 58 (4): 705-709

Brewer, M.S., Mc Keith, F.K. and Britt, K. 1992. Fat, soy and carrageenan Effects on sensory and physical characteristics of ground beef patties. *J. Food Sci.* 57 (5): 1051-1052, 1055

Brown, S. and Ledward, D.A.1987. Effect of temperature of communition on the stability and eating quality of English sausage. *Meat Sci.* 20: 97-105

Buckley, D.J., Morrissey, P.A. and Gray, J.I. 1995. Influence of dietary vitamin E on oxidative stability and quality of pig meat. *J. Anim. Sci.* 73: 3122-3130

Bullock, K.B., Huffman, D.L., Egbert, W.R., Mikel, W.B., Bradford, D.D. and Jones, W.R. 1994. Storage stability of low fat ground beef made with lower value cuts of beef. *J. Food Sci.* 59 (1): 6-9

Carballo, J., Mota, M., Barreto, G. and Colmenero, F.J. 1995. Binding properties and color of bologna sausage made with varying fat levels, protein levels and cooking temperatures. *Meat Sci.* 41 (3): 301-313

Carballo, J., Fernandez, P., Barreto, G., Solas, T.M. and Colmenero, F.J. 1996. Characteristics of high and low fat bologna sausages as affected by final internal cooking temperature and chilling storage. *J. Sci. Food Agric.* 72: 40-48

Cavestany, M., Colmenero, F.J. and Solas, M.T. 1994. Incorporation of sardine surimi in bologna sausage containing different fat levels. *Meat Sci.* 38: 27-37

*CCC. 1996. *Fat reduction in foods*. Calorie Control Council. Atlanta, Geneve

Claus, J.R. and Hunt, M.C. 1991. Low fat, high added water bologna formulated with texture modifying ingredients. *J. Food Sci.* 56 (3): 643-647, 652

*Claus, J.R., Hunt, M.C. and Kastner, C.L. 1989. Effects of substituting added water for fat on the textural, sensory and processing characteristics of bologna. *J. Muscle Foods.* 1: 1. In: Keeton, J.T. 1994.

Code of Federal Regulations. 1995. Title 9, Section 317, Subpart B, U.S. Government Printing Office, Washington, D.C. Cited by Quick, J. 1997. Labeling of low and reduced fat/salt products. In. *Production and Processing of Healthy Meat, Poultry and Fish products.* (Eds. Pearson, A.M. and Dutson, T.R.). Blackie Academic and Professional, New York. pp. 48-64

Cofrades, S., Carballo, J. and Colmnero, F.J. 1997. Heating rate effects on high fat and low fat frankfurters with a high content of added water. *Meat Sci.* 47 (1/2): 105-114

Coggins, P.C. 2001. Spices and flavourings for meat and meat products. In. *Meat Science and Application*. (Eds. Hui, Y.H., Nip, W.K., Rogers R.W. and Young, O.A.). Marcel Dekker Inc, New York. pp. 371-442

Colmenero, F.J. 2000. Relevant factors on strategies for fat reduction in meat products. *Trends in Food Sci. Technol.* 11: 56-66

Colmenero, F.J., Barreto, G., Fernandez, P. and Carballo, J. 1996. Frozen storage of bologna sausages as a function of fat content and of levels of added starch and egg white. *Meat Sci.* 42 (3): 325-332

Crehan, C.M., Hughes, E., Troy, D.J. and Buckley, D.J. 2000. Effect of fat level and maltodextrin in the functional properties of frankfurters formulated with 5, 12 and 30% fat. *Meat Sci.* 55: 463-469

Dawson, P.L. 2001. Packaging In: *Poultry meat processing* (Eds. Sams, A.R.) CRC Press, New York, pp. 73-95

*Dickson, R.L., Pearson, A.M., Anglemeir, A.F. and Holmes, Z. 1995. Konjac flour/carrageenan gel as a suitable fat replacer in a ground meat system. *Proc. Int. Cong. Meat Sci. Technol*. 41 (11): 433

Egbert, W.R., Huffman, D.L., Chen, C.M. and Jones, W.R. 1992. Mircrobial and oxidative changes in low fat ground beef during simulated retail distribution. *J. Food Sci.* 57 (6): 1269-1274, 1293

Eilert, S.J., Mandigo, R.W. 1993. Procedure of soluble collagen in thermally processed meat products. *J. Food Sci.* 58 (5): 948-949

Eilert, S.J. and Mandigo, R.W. 1997. Use of additives from plant and animal soruces in production of low fat meat and poultry products. In. *Production and Processing of Healthy Meat, Poultry and Fish Products.* (Eds Pearson, A.M. and Dutson, T.R.) Blackie Academic and Professional, London, pp. 210-225

Eilert, S.J., Mandigo, R.W. and Sumner, S.S. 1996. Phosphate and modified beef connective tissue effects on reduced fat high added water frankfurters. *J. Food Sci.* 61 (5): 1006-1011, 1029

Eilert, S.J., Blackmer, D.S., Mandigo, R.W. and Calkins, C.R. 1993. Meat batters manufactured with modified beef connective tissue. *J. Food Sci.* 58 (4): 691-696

El-Magoli, S.B., Laroia, S. and Hansen, P.M.T. 1996. Flavour and texture characteristics of low fat ground beef patties formulate with whey protein concentrate. *Meat Sci.* 42 (2): 179-193

FAO, 2002. Food energy methods of Analysis and conversion factors. Report of a technical workshop. FAO Food and Nutrition. Paper 77

Forrest, J.C., Aberle, E.D., Hedrick, H.B., Judge, M.D. and Merkel, R.A. 1975. *Principles of Meat Science*. W.H. Freeman & Co, Sanfrancisco, USA. p. 417

*Gedde, U.W. 1999. Pages 232-233 in Polymer Physics, Kluwer Academic Publishers, Dordrecht, Netherlands

Gregg, L.L., Claus, J.R., Hackney, C.R. and Marriot, N.G. 1993. Low fat, high added water bologna from massaged minced batter. *J. Food Sci.* 58 (2): 259-264

Hannan, R. S. 1984. The collagen content of meat products and its legislative implications. *J. Sci. Food Agric.* 35: 1265-1266

Harte, B.R. 1987. Packaging of restructured meats. In. *Restructured meat and Poultry Products*. (Eds. Pearson, A.M. and Dutson, T.R.). Van Nostrand Reinhold Company. New York. pp. 433-468

Henrickson, R.L. 1978. *Meat, poultry and sea food technology.* Prentical Hall Inc. N.J. P. 67

Ho, C.P., Huffman, D.L., Bradford, D.D., Egbert, W.R., Mikel, W.B. and Jones, W.R. 1995. Storage stability of vacuum packaged frozen pork sausage containing soy protein concentrate, carrageenan or antioxidants. *J. Food Sci.* 60 (2): 257-261

Hsu, S.Y. and Chung, H.Y. 2001. Effects of k-carrageenan, salt, phosphates and fat on qualities of low fat emulsified meatballs. *J. Food Engineering*. 47: 115-121

*Huffman, D.L. and Egbert, W.R. 1990. Advances in lean ground beef production. *Alabama Agric., Exp. Stat. Bull.* No.: 606

Hughes, E., Cofrades, S. and Troy, D.J. 1997. Effects of fat level, oat fibre and carrageenan on frankfurters formulated with 5, 12 and 30% fat. *Meat Sci.* 45 (3): 273-281

Hughes, E., Mullen, A.M., and Troy, D.J. 1998. Effects of fat level, tapioca starch and whey protein on frankfurters formulated with 5 and 12% fat. *Meat Sci.* 48 (1/2): 169-180

ICMR, 1990. Report of committee on dietary allowances, ICMR, New Delhi

Jo, C., Lee, J.I., Ahn, D.U. 1999. Lipid oxidation, color changes and volatile production in irradiated pork sausage with different fat content and packaging during storage. *Meat Sci.* 51: 355-361

Jobling, A. 1984. New applications for collagenous protein in meat products. *J. Sci. Food Agric.* 35: 1264-1265

Johns, A.M., Birkinshaw, L.H. and Ledward, D.A. 1989. Catalysts of lipid oxidation in meat products. *Meat Sci*. 25: 209-220

Kao, W.T. and Lin, K.W. 2006. Quality of reduced-fat frankfurter modified by konjac-starch mixed gels. *J. Food Sci.* 71 (4): 326-332

Keeton, J.T. 1994. Low fat Meat products – technological problems with processing. *Meat Sci.* 36: 261-276

*Kendall, R., and Mitolo, J.J. 1993. Use of starch in cured meats. Cured meat short course. Meat Laboratory, Iowa state University, Ames, IA. April 27-29

Kenney, P.B., Kastner, C.L. and Kropf, D.H. 1992. Raw and preheated epimysium and gelatin affect properties of low salt, low fat restructured beef. *J. Food Sci.* 57 (3): 551-554

Kilic, B. and Richards, M.P. 2003. Lipid oxidation in poultry doner kebab: Pro-oxidative and anti-oxidative factors. *J. Food Sci.* 68 (2): 686-689

Klettner, P.G., Ott,G. and Bohm,H. 2003. Firmness methods for pork, beef and turkey meat. *Fleischwirtschaft*. 83 (9): 132-135

Kumar, M. and Sharma, B.D. 2004. The storage stability and textural, physico-chemical and sensory quality of low fat ground pork patties with carrageenan as fat replacer. *Int. J. Food Sci. Technol.* 39: 31-34

*Liangi, M and Chen, N. 1991. Research in improving the WHC of meat in sausage products. In: *Proc. 37th Int. Cong. Meat Sci. Tech.* Copenhagen, Denmark, pp. 781-786

*Lin, K.W. and Chuang, C.H. 1999. Effects of dipping of raw meat materials on the physicochemical properties and shelf life of low fat Chinese style sausage. *Food Sci. Taiwan*. 26 (6): 583-595

Lupton, J.R. and Cross, H.R. 1999. Contributions of meat, poultry and fish to the health and well being of man. In. *Quality attributes and their measurement in meat, poultry and fish products.* (Eds. Pearson, A.M. and Dutson, T.R.). Aspen Publishers, Inc. Maryland USA. pp. 479-499

Martin, J.W. and Rogers, J. W. 1991. Cure levels, processing methods and meat source. Effects on low fat frankfurters. *J. Food Sci.* 56 (1): 59-61

Martinez, L.M.A., Quintana, V.A.M., Revilla, I. 2004. Effect of locust bean/xanthan gum addition and replacement of pork fat with olive oil on the quality characteristics of low fat frankfurters. Meat Sci. 68 (2004): 383-389

Matulis, J.R., Mc Keith, K.F., Sutherland, W.J. and Brewer, S.M. 1995. Sensory characteristics of frankfurters as affected by salt, fat, soy protein and carrageenan. *J. Food Sci.* 60 (1) 48-54

Meullenet, J.F., Chang, H.C., Carpenter, J.A. and Resurreccion, A.V.A. 1994. Textural properties of chicken frankfurters with added collagen fibers. *J. Food Sci.* 59 (4): 729-733

Monahan, F.J. and Troy, D.J. 1997. Overcoming sensory problems in low fat and low salt products. In. *Production and Processing of Healthy Meat, Poultry and Fish products.* (Eds. Pearson, A.M. and Dutson, T.R.). Blackie Academic and Professional. New York, pp. 257-281

Morrissey, P.A., Buckley, D.J. and Sheehy, P.J.A. 1994.Vitamin E and Meat quality. *Proc. of the Nutr. Soc.*. 53: 289-295

Murphy, E.W., Criner, P.E. and Gray, B.C. 1975. Comparison of methods for calculating retentions of nutrients in cooked foods. *J. Agric. Food Chem.* 23: 1153-1157

Naveen, Z., Reddy, P.M. and Reddy, K.P. 2006. Changes in the quality of duck meat sausages during frozen storage. *J.Food Sci.Technol*. 43 (3): 247-250

Nazeera, A.P. 2007. Quality and shelf life of low fat restructured turkey meat loaf. *MVSc thesis*, Kerala Agricultural University, Thrissur, Kerala

*NRC, 1989. *Recommended Dietary Allowances*, Tenth Edition, *National Academy of Sciences*, Washington, D.C

*Ockerman, H.W. 1976. Quality control of postmortem muscle and tissue. The Department of Animal Sciences, Ohio State University, Colombus, Ohio

Osburn, W.N. and Mandigo, R.W. 1998. Reduced fat bologna manufactured with poultry skin connective tissue gel. *Poultry Sci*. 77: 1574-1584

Osburn, W.N., Mandigo, R.W. and Eskridige, K.M. 1997. Pork skin connective tissue gel utilization in reduced fat bologna. *J. Food Sci.* 62 (6): 1176-1182

Page, J.K., Wulf, D.M. and Schwotzer, T.R. 2001. A survey of beef muscle colour and pH. *J. Anim. Sci.* 76: 678-687

Papadima, S.N. and Bloukas, J.G. 1999. Effect of fat level and storage conditions on quality characteristics of traditional Greek Sausages. *Meat Sci.* 51: 103-113

Pearson, A.M. 1997. Principles and applications in production of reduced and low fat products. In. *Production and Processing of Healthy Meat, Poultry and Fish* *products*. (Eds. Pearson, A.M. and Dutson, T.R.). Blackie Academic and Professional, New York, pp. 65-100

Pearson, A.M. and Gillet, T.A. 1997. *Processed Meats*. Third Edition. CBS Publishers and Distributors, New Delhi. p. 425

*Pearson, A.M., Asghar, A., Gray, J.I. and Booren, A.M. 1987. Impact of fat reduction on palatability and consumer acceptance of processed meats. *Proc. Recip. Meat Conf.* 40: 105

Prabhu, G.A., Doerscher, D.R. and Hull, D.H. 2004. Utilisation of pork skin collagen protein in emulsified and whole muscle meat products. *J. Food Sci.* 69 (5): 388-392

Puolanne, E. and Ruusunen, M. 1981. The properties of connective tissue membrane and pork skin as raw materials for cooked sausage. *Meat Sci.* 5 (5): 371-382

Ranken, M.D. 2000. *Handbook of Meat Product Technology*. Blackwell Science Ltd., Oxford. p.212

*Rogers, R.W., Healy, T., Armstron, T., Coggins, P., Hairi, F., Martin, M. and Williams, B. 1996. Effects of various starches on the characteristics of fat free bologna. *Proc. 49th A. Recip. Meat Conf*. Chicago, IL. p.196

Rust, R.E. 1987. Sausage Products. In. *The Science of Meat and Meat Products*. Third Edition. (Eds. Price, J.F. and Schweigert, B.S) Food and Nutrition Press, Inc., West Port, CT. pp. 457-485

Sadler, D.H.N. and Young, O.A. 1993. The effect of preheated tendon as a lean meat replacement on the properties of fine emulsion sausages. *Meat Sci.* 35: 259-268

Satterlee, L.D., Zachariah, R.A. and Levin, R.E. 1973. Utilisation of beef and pork skin hydrolyzates as binder or extender in sausage emulsions. *J. Food Sci.* 38: 268-270

Schilling, M.W., Mink, L.E., Gochenour, P.S., Marriott, N.G. and Alvarado, C.Z. 2003. Utilisation of pork collagen for functionality improvement of boneless cured ham manufactured from pale, soft and exudative pork. *Meat Sci.* 65: 547-553

Schnell, T.D. 1999. Effect of raw and pre heated pork skin collagen in low fat bologna. PhD dissertation submitted to the university of Nebraska, Lincoln, USA

Serdaroglu, M. 2006. Improving low fat meat ball characteristics by adding whey powder. *Meat Sci.* 72 (1): 155-163

Serdaroglu, M. and Degirmencioglu, O. 2004. Effects of fat level (5%, 10%, and 20%) and corn flour (0%, 2%, 4%) on some properties of Turkish type meat balls (koefte). *Meat Sci*. 68: 291-296

Shackelford, S.D., Reagan, J.O., Haydon, K.D., Lyon, C.E. and Miller, M.F. 1991. Acceptability of low fat frankfurters as influenced by the feeding of elevated levels of monounsaturated fats to growing – finishing swine. *Meat Sci.* 30: 59-73

Sheard, P.R., Nute, G.R. and Chappell, A.G. 1998. The effect of cooking on the chemical composition of meat products with special reference to fat loss. *Meat Sci.* 49 (2): 175-191

Shrimpton, D.H. 1984. Connective tissue. Diet and Nutrition. *J. Sci. Food Agric.* 35: 1266-1267

Smith, D.M. 1997. Low fat and low salt poultry products. In. *Production and processing of Healthy meat, Poultry and Fish Products.* (Eds. Pearson, A.M. and Dutson, T.R). Blackei Academic and Professional, London, pp. 298-320

Smith, D.M. and Alvarez, V.B. 1988. Stability of vacuum cook-in-bag turkey breast rolls during refrigerated storage. *J. Food Sci.* 53 (1): 46-48

Snedecor, G.W and Cochran, W. G. 1994 *Statistical Methods*. Eighth edition. The Iowa State University, Ames, Iowa. p. 313

St. John, L.C., Buyck, M.J., Keeton, J.T., Leu, R. and Smith, S.B. 1986. Sensory and physical attributes of frankfurters with reduced fat and elevated monounsaturated fats. *J. Food Sci.* 51(5): 1144-1146

Trindade, M.A., Contreras, C.C. and De Felicio, P.E. 2005. Mortadella sausage formulations with partial and total replacement of beef and pork back fat with mechanically separated meat from spent layer hens. *J. Food Sci.* 70(3): 236-241

Troutt, E.S., Hunt, M.C., Johnson, D.E., Claus, J.R., Kastner, C.L. and Kropf, D.H. 1992a. Characteristics of low fat ground beef containing texture modifying ingredients. *J. Food Sci.* 57(1): 19-24

Troutt, E.S., Hunt, M.C., Johnson, D.E., Claus, J.R., Kastner, C.L., Kropf, D.H. and Stroda, S. 1992b. Chemical, physical and sensory characterization of ground beef containing 5-30% fat. *J. Food Sci.* 57(1): 25-29

Troy, D. J, Desmond, E.M. and Buckley, D.J. 1999. Eating quality of low fat beef burgers containing fat replacing functional blends. *J. Sci.Food Agric*. 79: 507-516

*USDA – FSIS. 1990. Definitions and standards of identity of composition. Sub part G – cooked sausage. 'Code of Federal Regulations', Title 9, pt. 319-180. Office of Federal Register, National Archives and Records, GSA, Washington, D.C

*USDHHS, 1995. Nutrition and your health: Dietary guidelines for Americans. Fourth Edition. Home and Garden Bulletin, NO 232. Washington, D.C

Wang, F.S., Jiang, Y.N. and Lin, C.W. 1995. Lipid and cholesterol oxidation in chinese style sausage using vacuum and modified atmosphere packaging. *Meat Sci.* 40: 93-101

*Webster, J.D., Ledward, D.A. and Lawrie, R.A. 1982. Protein hydrolysates from meat industry by-products. Meat Sci. 7: 147-167

Whiting, R.C. 1988. Ingredients and processing factors that control muscle protein functionality. *Food Technol.* 42: 104, 110-114, 210

Witte, V.C., Krause, J.F. and Bailey, M.E. 1970. A new extraction method for determining 2-thiobarbituric acid values of pork and beef during storage. *J. Food Sci.* 35: 582-585

*WHO. 2003. Diet, Nutrition and the prevention of chronic diseases. WHO technical report series 916. Geneve

Wu, T.C. and Sheldon, B.W. 1988. Influence of phospholipids on the development of oxidized off flavors in cooked turkey rolls. *J. Food Sci.* 53(1): 55-61

Yoo, S.S., Kook, S.H., Park, S.Y., Shim, J.H. and Chin, K.B. 2007. Physico-chemical characteristics, textural properties and volatile compounds in comminuted sausages as affected by various fat levels and fat replacers. *Int. J. Food Sci. Technol.* 42: 1114-1122

Younathan, M.T., Marjan, Z.M. and Arshad, F.P. 1980. Oxidative rancidity in stored ground turkey and beef. *J. Food Sci*. 45: 274-275

Young, L.L., Garcia, J.M., Lillard, H.S., Llyon, C.E. and Papa, C.M. 1991. Fat content effects on yield, quality and microbiological characteristics of chicken patties. *J. Food Sci.* 56(6): 1527-1528, 1541

* Originals not consulted

EFFECT OF PORK SKIN COLLAGEN AS A FAT REPLACER IN LOW FAT FRANKFURTER

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ABSTRACT

The health conscious meat consumers prefer low fat meat products and therefore, the present study was designed with a view to developing a suitable formulary for low fat frankfurter (LFF) with pork skin collagen (PSC) gel as a fat replacer. Quality was assessed by measuring pH, emulsion stability, cook yield, cook loss, dimensional shrinkage, water holding capacity (WHC), Warner-Bratzler Shear Force Value (WBSFV) , colour by Hunter L^*, a^*, b^* values, proximate composition, nutritive value and organoleptic qualities on the day of production. The shelf life of frankfurters in aerobic (AP) and vacuum packaging (VP) systems at -20°C for 75 days were evaluated based on purge loss, 2-thiobarbituric acid reactive substances value (TBARS), organoleptic qualities, and moisture, protein and fat contents. Six trials of the experiment were conducted and the cost of production was also calculated.

Seven different formulations viz., one full fat control 30% with out PSC (30/0) and six treatments 10% low fat with out PSC (10/0), 5% low fat with out PSC (5/0), 10% fat with 5% PSC (10/5), 10% fat with 10% PSC (10/10), 5% fat with 5% PSC (5/5), 5% fat with 10% PSC (5/10) using beef and pork trimmings 1:1, pork skin collagen gel, lard, curing ingredients, spices and condiments were prepared. The steam cooked frankfurters were packaged under AP and VP systems in high density polyethylene and polyamide/polyethylene pouches, respectively and stored at -20°C for 75 days for shelf life studies

The pH of the cooked LFF with PSC was significantly more ($P < 0.05$) than those with out PSC, their uncooked batter and in the range of 6.46-6.55 and could be considered a low acid food. Formulation 10/10 had the highest ($P < 0.05$) emulsion stability of 87.40, cook yield of 96.28, water holding capacity (WHC) of 95.57 and

lowest ($P< 0.05$) dimensional shrinkage of 1.83 per cent. The WHC of 95.57 of formulation 10/10 was significantly higher ($P< 0.05$) than in other treatments. The WBSFV of 10/10 was 1.12 kgf, which was significantly lower ($P< 0.05$) than other treatments indicating its increased tenderness. The PSC significantly reduced the shear force of LFF. The 10/10 formulation had higher L^* and b^* values of 62.99 and 17.51 and a lower a^* value of 5.70. Formulation 10/5 was ranked second among treatments in all these traits. The moisture, protein, fat, carbohydrate and ash contents in 10/5 and 10/10 were 72.32, 13.01, 9.57, 4.17, 0.93 and 71.30, 13.09, 9.82, 4.81 and 0.98, respectively.

The per cent contribution of protein to the RDA from 10/10 and 10/5 were 21.82 and 21.68, respectively. The per cent RDA of calories from fat was 4.02 and 3.92, respectively and below the recommended 30 per cent. The purge of 10/10 and 10/5 formulations were significantly lower ($P< 0.05$) irrespective of the packaging systems and period of storage. The TBARS of low fat formulations with varying levels of PSC were within the acceptable range of 1mg malonaldehyde/kg of frankfurter for oxidative rancidity, irrespective of the packaging systems and storage period. There was no significant increase in the oxidative rancidity of PSC added formulations noticed during the initial 30 days of storage. The appearance, colour, flavour, texture, juiciness, saltiness, mouth coating and overall acceptability of 10/10 were very desirable and comparable with that of full fat control. Formulation 10/10 under VP had a more beneficial effect in retaining flavour, texture, and juiciness followed by formulation 10/5. The period of storage affected the moisture and protein content of 10/5 and 10/10 formulations on d 60 and on d 45 onwards, respectively and packaging systems affected the protein content on d 75 of storage at -20^{o}C. The cost of production calculated per kg of 10/5 and 10/10 were Rs.84/= and Rs.86/= respectively.

The formularies for LFF 10/10 and 10/5 were developed with very acceptable organoleptic attributes, cook yield and shelf life up to 75 days at -20ºC, respectively under AP and VP systems economically. The contribution of calories from fat to the Recommended Daily Allowance (RDA) was below the recommended 30% in all frankfurter formulations making the product acceptable to health conscious consumers. The cost of production was calculated on laboratory scale and further investigations with large quantity are required for commercial production at industrial level.