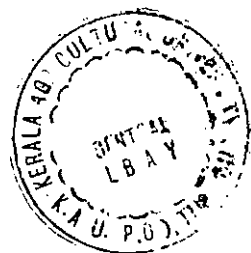


FORMULATION OF LOW FAT BEEF BURGER WITH FAT REPLACERS



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**Thesis submitted in partial fulfilment of the
requirement for the degree of**

Master of Veterinary Science

**Faculty of Veterinary and Animal Sciences
Kerala Agricultural University, Thrissur**

2010

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DECLARATION

I hereby declare that this thesis, entitled “**FORMULATION OF LOW FAT BEEF BURGER WITH FAT REPLACERS**” 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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CERTIFICATE

Certified that this thesis, entitled “**FORMULATION OF LOW FAT BEEF BURGER WITH FAT REPLACERS**” is a record of research work done independently by **Dr. Govande Premanand Laxmanrao** 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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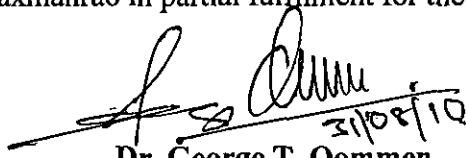


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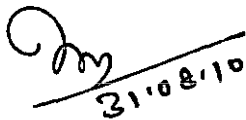
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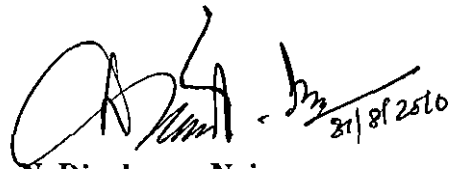
We, the undersigned members of the Advisory Committee of Dr. Govande Premanand Laxmanrao a candidate for the degree of Master of Veterinary Science in Livestock Products Technology, agree that the thesis entitled "**FORMULATION OF LOW FAT BEEF BURGER WITH FAT REPLACERS**" may be submitted by Dr. Govande Premanand Laxmanrao in partial fulfilment for the degree.



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Dedicated To

My Beloved Family

Introduction

INTRODUCTION

There are several factors boosting the demand for convenience in meal preparation. The key factors are higher standard of living and educational status, higher purchasing capacity of the employed community, increased awareness on requirement of nutritious food, increasing participation of women in the work force, lack of time due to job and recreational activities, growing number of single person family and small households, lack of skill, experience and facilities in preparing meals at home, migration of people to urban areas for job, students and other people at work taking packed meals and increasing catering establishments. The modern meat food industry and retailers in the developing countries have readily reacted to this growing demand by considerably expanding their variety of pre-prepared meat products which include fast food, ready-to-cook and ready-to-eat meat products available in market.

Among the coarsely ground meat products, beef burger is very popular in all classes of society and is evident from the continued increase in the sale from all catering establishments, fast food chains and super markets in urban areas.

Many researchers proved that high fat diet is associated with increased risk of obesity, colon cancer, cardiovascular disease, non-insulin dependent diabetes and saturated fat intake is associated with high cholestraemia, atherosclerosis and coronary heart disease (USDHHS, 1995; AHA, 1996; Alisons *et al.*, 2010).

By 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 comminuted meat products.

To maintain the desirable palatability in low fat meat products, several non meat ingredients that act as texture or water binding modifiers have been tested with same success (Troy *et al.*, 1999). Many of the researchers successfully used different fat mimetic and substitute in cooked meat products since no substitute has yet been found that imparts all the characteristics of the fat to the products without any undesirable effects such as unnatural taste (Allen *et al.*, 1999).

Hence there is great opportunity to develop low fat comminuted meat products like beef burger with appropriate fat replacers. The optimization of their concentration to produce low fat meat products is necessary for better consumer acceptability and market value.

Pregelatinised Pork Skin Collagen (PSC) is used to improve water retention and fat emulsification and thereby improving the texture, flavor and succulence of the product. PSC is an inexpensive adjunct to improve these traits. PSC gel, 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 (Prabhu *et al.*, 2004).

Carrageenan (CG) that derived from red seaweed consist of three basic types kappa, iota and lambda each having the different properties basically used to retain the texture, juiciness and excellent gelling properties in low fat meat products such as sausage and bologna (Pearson and Gillet, 1997).

Tapioca Starch (TS) widely used in meat industry due to its ability to bind and retain moisture during cooking also having high viscosity, clear appearance, and low production cost (Allen *et al.*, 1999).

Labeling of the manufactured products with its nutrition information and keeping quality has become a stipulation. Therefore, determination of the proximate composition, nutritional status and keeping quality of the product is

required for consumer acceptance and better marketing.

Therefore, the present study was undertaken with the following objectives.

1. To develop a palatable and economical formulation of Low Fat Beef Burger (LFBB) with pregelatinised pork skin collagen (PSC), carrageenan (CG), tapioca starch (TS) and their blends as fat replacers.
2. To assess the effect of different levels of fat reduction and incorporation of fat replacers and their blends on the physico-chemical characteristics, proximate composition, cooking, nutritional status, textural and organoleptic characteristics of LFBB.
3. To evaluate the keeping quality of LFBB under aerobic and vacuum packaging at 0-4°C and -20°C.

Review of Literature

REVIEW OF LITERATURE

The current study was conducted with the objectives of developing a low fat beef burger (LFBB) with pregelatinised pork skin collagen (PSC), carrageenan (CG), tapioca starch (TS) and their blends as a fat replacers and evaluating its physical and chemical characteristics, proximate composition, nutritional quality, cooking characteristics, texture, organoleptic qualities and shelf life under aerobic packaging (AP) and vacuum packaging (VP) at 0-4°C and -20°C. Search on available literature revealed that very little research has been carried out in the formulation of LFBB and a sizable volume of work has been carried out in other low fat meat products with different fat replacer.

Therefore, literature on the rationale for development of low fat meat products, effect of fat reduction on sensory, physical and cooking characteristics of low fat meat products with varying fat levels and PSC, CG and TS as fat replacers and effect of aerobic and vacuum packaging and storage on various quality attributes of low fat meat products are reviewed.

2.1. RATIONALE FOR DEVELOPMENT OF LOW FAT MEAT PRODUCTS

According to the nutritional guidelines and other health watch groups, dietary fat should provide between 15 and 30 per cent of total calories, and those saturated fats should be limited to between 0 and 10 per cent of calorie intake and cholesterol intake below 300 mg/day (Chizzolini *et al.*, 1999).

High fat intake is associated with increased risk for obesity and some types of cancer. However, saturated fat intake is associated with high blood cholesterol and coronary heart disease (Astrup *et al.*, 2000).

World Health Organization 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 GROUND MEAT PRODUCTS

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” (Keeton 1994; Pearson and Gillet, 1997).

The definitions for nutrient claims as per Code of Federal Regulations (1995) indicated that total fat in low fat meat products should be $\leq 3\text{g}$. 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.

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

For the design or formulation of low fat meat products several aspects, viz., sensory, nutritional, technological safety, legal, appreciation, cost, convenience, marketability and ethics are to be considered. 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 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). The palatability of ground beef is directly related to the fat content (Egbert *et al.*, 1991). Fat is the calorie dense nutrient that provides flavor, texture and juiciness in processed meat (Yang *et al.*, 1995).

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).

The primary role of the fat in low fat frankfurter is to trap the volatile and released compounds during mastication (Allen *et al.*, 1999).

Besides the biological and physiological functions, fat plays a major role the textural, functional and sensorial properties of comminuted meat products (Turhan *et al.*, 2005).

2.4. TECHNOLOGICAL PROBLEMS WITH LOW FAT MEAT PRODUCTS

Production of low fat products through simple fat reduction substantially reduces product juiciness, tenderness and flavor intensity (Pearson *et al.*, 1987).

Keeton (1994) in a review on the technological problems of low fat meat products has stated that, low fat (5 to 10 per cent) fresh or cooked/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.

Berry (1997) studied the improvement of the properties of low fat beef patties on addition of sodium alginate and modified tapioca starch, reported decline in palatability with fat reduction.

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

Technological aspects processing of low fat meat products include problems with texture, flavor and mouthfeel (Turhan *et al.*, 2005).

2.4.1. Color

Martin and Rogers (1991) observed that 50:50 beef-pork sausage 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.* (1992) revealed that low fat patties of 5 and 10 per cent had darker red color.

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 (Hughes *et al.*, 1997).

Reduced fat in comminuted meat products may be darker in color due to concentration of meat pigment (Osborn *et al.*, 1997). Hughes *et al.* (1998) opined that reducing fat level decreased the lightness and increased redness of the beef burgers although yellowness was not significantly affected.

Crehan *et al.* (2000) proved that 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 beef burgers.

2.4.2. Flavour

Egbert *et al.* (1991) reported that reducing the fat level in low fat ground beef are difficult and result in reduction in tenderness, juiciness, flavor intensity and overall products palatability.

Berry and Wergin (1993) showed that addition of modified pregelatinised potato starch resulted in lower incidence of beef flavor in low fat ground beef patties as fat reduced from 20 to 5 per cent.

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).

Chevance *et al.* (2000) reported that addition of carbohydrate fat replacers to low fat meat products could assist the flavour qualities of low fat meat products by slowing down the release of odour compounds. Reduction in fat content in beef burger, salami and frankfurter resulted in an increase in the quantities of volatiles released in the headspace.

Flavour is particularly a problem in low fat meat products owing to decreases when fat is reduced (Tokusoglu and Unal, 2003).

Role of fat is an important consideration in any formulation because it plays major role in the texture, juiciness and flavor of comminuted meat products (Luruena-Martinez *et al.*, 2004).

2.4.3. Tenderness and Texture

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

Reducing the fat content of high fat traditional products affects their flavour, texture, juiciness and mouth feel in addition to reducing satiety value (Allen *et al.*, 1999). Troy *et al.* (1999) studied the eating quality of low fat beef

burgers containing fat replacers and observed that low fat burgers were least tender and driest compared to full fat burgers.

As fat level decreases in beef patties, tenderness, juiciness and flavour ratings decreases and shear force increases (Gujral *et al.*, 2002).

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

2.4.4. Juiciness

Troutt *et al.* (1992) opined that low fat (5 and 10 per cent) ground beef patties had lower juiciness and moisture release.

Miller *et al.* (1993) studied the physical and sensory characteristics of low fat ground beef patties as fat per cent reduced from 20 per cent to 5-10 per cent resulted in reduction in juiciness.

As the fat content reduced from 25-30 to 5-10 per cent, the cooking loss, drip loss, juiciness, beef flavor, tenderness, oily mouth coating and consumer acceptance decreased (Keeton, 1994).

2.4.5. Saltiness

Matulis *et al.* (1995) have shown that as the salt levels rise, the increase in saltiness is more noticeable in more fatty products than lean ones.

Hughes *et al.* (1997) reported that decreasing fat content of pork beef burgers 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. LOW FAT BEEF BÜRGER FORMULATION WITH FAT REPLACERS

According to US Federal Regulations, hamburger consist of chopped fresh and/or frozen beef with or without the addition of beef fat as such and/or seasonings including salt, sweetening agents, flavouring, spices, MSG, HVP and the like in condimental proportions. It will not contain more than a total of 30 per cent fat (Henrickson, 1978).

Acceptable beef burger could be manufactured with a minimum of 1.3 per cent salt, as salt increased hardness, juiciness, saltiness and flavor (Matulis *et al.*, 1995).

One of the challenges 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).

The maximum fat level for beef burgers was set at 30 per cent and added water at 10 per cent (Pearson and Gillet, 1997). In UK, beef burger or ham burger may refer to a mixture of 80 -100 per cent meat and the rest cereal products. The fat content shall not be more than 40 per cent. The major function of salt in processed meat product is to reduce microbial growth, increase flavor, interact with lean meat protein to give increased water retention, yield and also increase the meat binding, cohesion, increase fat binding and texture (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).

2.5.1. Fat Replacers

Fat replacers are ingredients that contribute a minimum of calories to formulated meats and do not significantly alter juiciness, mouth feel, viscosity or other organoleptic and processing properties (Keeton, 1994).

Calorie Control Council (1996) recommended that as there is no single ideal fat replacer that can recreate all the functional and sensory attributes, a system approach using several ingredients individually or in combination is frequently used to achieve the characteristics of fat.

Fat replacers chemically resemble fat, protein, or carbohydrates and are categorized into fat substitutes and fat mimetics. The former is a macromolecule that physically and chemically resembles triglycerides and chemically synthesized or derived from conventional fats and oils by enzymatic modifications. Fat mimetics are substances that imitate organoleptic or physical properties of triglycerides. Fat mimetics are generally proteins or carbohydrate based fat replacers (Akoh, 1998).

Under the FDA regulations, fat replacers fall into one of two categories: food additives or Generally Recognized As Safe (GRAS) substances. Fat replacers made with a combination of existing ingredients such as starches, gums, fibers or proteins that are widely used in the food supply require no special approval (ADA, 2005).

2.5.1.1. Pork skin collagen

Preheating connective tissue to a gelatinous state, convert collagen to gelatin, a strong water binder (Satterlee and Zachariah, 1973). Pork skin collagen converted to gelatin by heating and addition of water could form a gel upon cooling and on incorporation in reduced fat meat products may improve the products yield, texture and palatability (Puolanne and Ruusunen, 1981).

Chevez *et al.* (1983) added wet collagen to ground beef at 0, 10 and 20 per cent as a lean meat replacement, stored the mixed products at -15°C up to two weeks and observed that the patties were superior in texture and juiciness compared to those with no added collagen.

Quint *et al.* (1987) found that pork skin pre emulsion could replace 7.5 per cent lean meat in pork loaf and increased the hardness and firmness. Pork collagen is an inexpensive adjunct that increases cooking yield and tensile strength in restructured beef (Kenny *et al.*, 1992).

Kenney *et al.* (1992) studied the effect of gelatin on low salt, low fat restructured beef and 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.

Eilert and Mandigo (1993) found that heating collagen for 10 to 15 min at 48°C to 52°C was adequate to separate soluble collagen in thermally processed products like beef burgers.

Addition of pork skin connective tissue gel in bologna decreased hardness and increased juiciness, which indicated the potential of pork skin connective tissue gel as water binder and texture modifying agents. Moist heating at 70°C for 30 min was sufficient to enhance the water binding ability of pork skin connective tissue and more water was bound at 70°C. Pork skin could hold 3g water/g of skin. The pH ranged from 7.42 to 7.69. 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).

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

Pork collagen has been refined so that it can be utilized in processed meat products to improve protein functionality through the immobilization of free water, increase the stability of the finished structure. Collagen from various sources has been used as the ingredient to improve the water and fat retention in

the meat products. Incorporation of pork collagen at 1 per cent and above significantly ($p < 0.05$) increased the cook yield and chilled yield in frankfurters (Prabhu *et al.*, 2004).

Selvakumar (2009) used 5 and 10 per cent pork skin collagen in low fat frankfurters and reported very acceptable organoleptic attributes, cook yield and shelf life up to 75 days at -20°C in both aerobic and vacuum packaging systems.

2.5.1.2. Carrageenan

Egbert *et al.* (1991) found the addition of 0.5 per cent iota-carrageenan to resulted in shear force reduction in cooked patties.

Berry *et al.* (1996) used iota-carrageenan in low fat ground beef products produced improvement in the tenderness and juiciness over low fat patties receiving no carrageenan or kappa-carrageenan. The melting point 49°C of carrageenan may cause premature loss of moisture or water soluble flavors (Keeton, 1994). Kappa-carrageenan improved gel strength and water retention of salt soluble gel in model systems by molecular interaction (De Frietas *et al.*, 1997).

Blouks *et al.* (1997) reported that iota-carrageenan at 0.5-1 per cent level appeared to be best for the production of low fat frankfurter as it reduced the hardness of skin and contributed to a softer product and higher overall acceptability. Hydrocolloids, especially carrageenan were often utilised as fat replacers in modifying both texture and sensory attributes of meat products (Keeton, 1994).

Carrageenan reacts synergistically with starches and mannan-containing carbohydrates, such as konjac flour. They have excellent functionality in low fat meat products and can be used in rather low concentrations of 0.1 to 0.5 per cent (Pearson and Gillet, 1997).

Carrageenan dissolves throughout meat during thermal processing and gels on cooling. It improves water retention, consistency and texture of comminuted meat products (Trius and Sebranek, 1996).

Troy *et al.*, (1999) reported that low fat beef burger containing blends of carrageenan was most effective in retaining moisture and in improving cooking characteristics such as cook yield and WHC. Allen *et al.*, (1999) observed increased flavour and overall quality in beef burger containing carrageenan, pectin, cellulose and oat fibre.

Hsu and Chung (2001) observed an increase in cooking yield, hardness and other textural profile analysis parameters by adding up to 2 per cent carrageenan to low-fat emulsified meatballs.

A combination of hydrocolloid fat substitutes like sodium alginate and carrageenan at 0.1 per cent and 0.75 per cent levels, respectively significantly increased most of the sensory attributes of low fat ground buffalo meat patties. Cooking yield was significantly higher at 0.25 and 0.5 per cent levels compared to 0.75 per cent level of carrageenan in low fat ground buffalo meat patties (Suman, 2001).

Carrageenan polysaccharides are used as additives to improve food texture, gelation, stability and viscosity and are generally regarded as safe (GRAS) by the Food and Drug Administration in the US (Yu *et al.*, 2002).

Hasret (2006) studied the effect of carrageenan and guar gum on the cooking and textural properties of low-fat meatballs found that kappa and iota-carrageenan improved the texture of this product and creates complexes with water and proteins.

Naseera (2007) reported that low fat restructured turkey meat loaf with carrageenan at 0.2 per cent had good overall acceptability, cook yield, nutritional value and shelf life up to 40 and 60 days at 0-4° C and -20° C, respectively under aerobic and vacuum packaging.

In low fat meat balls, addition of 0.5 -1 per cent carrageenan significantly increased product yield, water binding and texture (Ulu, 2004). One of the most promising fat substitutes is carrageenan. Carrageenan is a water soluble polysaccharide produced from red seaweeds (Rhodophyceae). It is a hydrocolloid consisting of potassium, sodium, magnesium and calcium sulphate esters of galactose and 3, 6-anhydrogalactose, copolymers (Cierach *et al.*, 2009).

2.5.1.3. Tapioca starch

Berry and Wergin (1993) found that the inclusion of tapioca starch in both low and high fat beef burgers could improve tenderness and cooking yields. This was due to the ability of the tapioca starch to bind water and hold it during cooking.

Products identified as meat loaves are restricted to have 3.5 per cent of extender material 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).

Berry (1997) studied the synergistic effects of hydrocolloids combinations in meat patties. The combinations of sodium alginate and modified tapioca starch improved juiciness of low-fat beef patties.

Texture Profile Analysis (TPA) indicated that addition of tapioca starch reduced cooking loss and increased emulsion stability. Tapioca starch increased overall flavour intensity. Response surface methodology used to study simultaneous effect of tapioca starch (ranging from 0 to 30 g/kg), oat fiber and

whey protein on the physical and organoleptic character of low fat beef burger revealed that the level of tapioca starch influence the cooking character and larger negative liner effect on the Warner Bratzler shear force. The following ingredients performed best as fat substitutes in low fat beef burgers: tapioca starch, carrageenan, oat fibre, pectin, whey protein and a commercial mixture of carrageenan and locust bean gum. Blends of these often performed better than single ingredients, with tapioca starch, oat fibre and whey protein performing best in terms of eating quality (Allen *et al.*, 1999).

Dry addition of tapioca starch has positive effect on physical, mechanical texture value and organoleptic parameter of low fat pork sausage. Increasing concentration of tapioca starch from 0 to 3 per cent in combination with preformed gel decreased the cook losses approximately 15 to 4 per cent. They also observed increased purge loss with increasing tapioca starch addition (Lyons *et al.*, 1999). Troy *et al.*, (1999) reported that tapioca starch with other fat replacers bind and retain water to produce a more tender product and reducing the shear force.

2.5.2. Processing

Trout *et al.* (1992) suggested that longer cooking time may be required with low fat beef patties to reach internal temperature equivalent to those of higher fat patties.

Frying causes a large increase in the fat content of poultry products. In order to reduce the fat content of pre-cooked products, cooking methods like roasting, broiling, grilling and baking are used. Longer cooking time may be necessary to maximize yield and to optimise texture (Smith, 1997).

Troy *et al.* (1999) prepared low fat beef burger with lean beef, fat trimming, water, salt, tapioca starch, carrageenan and their blends. Beef was coarsely ground through a 10mm mincer plate initially and during grinding

maintained 4° C. The meat mixture was finally minced through 5mm plate and made in to 113g beef burger.

Ranken (2000) reported that care must be taken during cooking to ensure proper pasteurisation. The core temperature must reach not less than 70°C for 2 min. The cooked products must be chilled hygienically to avoid recontamination.

Carbonell *et al.* (2005) manufactured beef burger with lean beef, beef back fat, water, salt and pepper in the simple traditional formulation. The trimmings were ground through a 5mm mincer plate. The burger mixture was shaped using a commercial burger maker 9 cm internal diameter, 1 cm thickness and 70 g weight.

Turhan *et al.* (2005) prepared low fat beef burger using ground beef, back fat, salt, onion, black pepper, red pepper and different level of hazelnut pellicle. Burger mix mixed in a blender for 10 min, weighed into 50g portions, and then compressed in a hand press.

Bochi *et al.* (2008) reported the preparation of fish burger with silver catfish found that all formulations were kneaded by hand until homogeneous dough was obtained. Portions of 60g were shaped in a circular mould of 9 cm diameter.

2.6. QUALITY ATTRIBUTES OF LOW FAT BEEF BURGER

2.6.1. pH

Claus and Hunt (1989) in bologna formulated with low fat and high added water and Selvakumar (2009) in low fat frankfurter reported higher pH value.

Ho *et al.* (1995) reported that the pH values were higher for reduced fat pork sausage patties added with carrageenan than for fat controls.

Osburn and Mandigo (1998) studied the effect of utilization of poultry skin connective tissue gel in reduced fat bologna and reported that the pH ranged from 5.92 to 5.98 in cooked bologna.

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. Meat batters manufactured with modified beef connective tissue had higher pH as reported by Eilert *et al.*, (1993).

2.6.2. Cooking Characteristics

The various cooking characteristics, viz., cook yield, cooking loss, fat and moisture retention and dimensional shrinkage of low fat beef burger (Desmond *et al.*, 1998; Troy *et al.*, 1999; Khalil, 2000; Carbonell *et al.*, 2005) and fish burger (Bochi *et al.*, 2008) were studied.

2.6.2.1. Cook yield and cook loss

Kregel *et al.* (1986) found that ground beef patties heated to 71° C had higher juiciness and tenderness scores than patties heated to 77° C.

Addition of tapioca starch reduced cook loss and increased overall flavour and emulsion stability (Hughes, 1994).

Incorporation of pork skin collagen at 1 per cent or above significantly increased ($P < 0.05$) cooked yield in low fat bologna (Schnell, 1999). Prabhu *et al.* (2004) reported increased cook yield when pork skin collagen content was increased from 0 to 3 per cent in emulsified and whole muscle meat products.

Pietrasik and Duda (2000) reported that cooking loss was decreased with increase in carrageenan. Kumar and Sharma (2004) observed significant improvement in cooking yield due to addition of carrageenan in low fat loaves.

Addition of carrageenan at 0.2 per cent in low fat restructured turkey meat loaf observed significant increase in cooking yield (Naseera, 2007).

Serdaroglu and Degirmencioglu (2004) reported that the cook yield was lower for 20 per cent fat than 10 or 5 per cent fat in turkish meat balls. Pinero *et al.* (2008) determined cook yield percentage of beef patties by measuring the weight differences of patties before and after cooking.

2.6.2.2. Fat and moisture retention

The moisture and fat retention values represent the amount of moisture and fat retained in the cooked product per 100g of raw sample expressed in percentage (El-Magoli *et al.*, 1996).

Desmond *et al.* (1998) observed that tapioca starch had the ability to retain moisture and had improved the physical traits of low fat beef burgers.

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.

Allen *et al.* (1999) showed that in low fat formulations with 8 per cent fat, water should not be added at above 8 per cent to avoid excessive weight loss during cooking.

Khali (2000) studied the quality characteristics of low fat beef patties modified with corn starch and water and observed significant increase in fat retention on cooking with increase in replacement of fat. Reduction of fat level from 25 to 10 per cent improved yield, fat retention and reduced shrinkage (Ulu, 2004).

Fat and moisture retention increased significantly in low fat ground pork patties incorporated with carrageenan compared to control product with 20 per cent fat (Kumar and Sharma, 2004).

2.6.2.3. Dimensional shrinkage

The percentage shrinkage was determined as the difference in the average diameter of individual cooked and uncooked beef burgers (Berry, 1992; Mansour and Khalil, 1997; Troy *et al.*, 1999 and Pinero *et al.*, 2008)

The addition of carrageenan, tapioca starch and their blends significantly reduced the diameter of low fat beef burger (Troy *et al.*, 1999).

Pinero *et al.* (2008) reported no reduction in diameter of beef patties due to binding and stabilizing properties of oat soluble fibre which held meat particles together.

2.6.3. Water Holding Capacity

Meat batter containing either 0.5 or 1 per cent iota-carrageenan or kappa-carrageenan increased water holding ability from 55-70°C (Foegeding and Ramsey, 1987).

Desmond *et al.* (1998) reported that low fat beef burger with carrageenan and tapioca starch as fat replacer had a WHC between 33-39 per cent.

Decreasing fat content from 30 to 5 per cent significantly increased the cook loss and decreased water holding capacity (Allen *et al.*, 1999).

Hart and Price (1993) and Troy *et al.* (1999) reported that low fat beef burger containing tapioca starch increased their water holding capacity. Ruusunen *et al.* (2003) who found that modified tapioca starch improved the water binding capacity of low fat frankfurter.

Bologna with pregelatinised pork skin and water at 5 and 25 per cent, respectively, reduced purge (Schnell, 1999). Collagen protein can be refined so that it can be used in the processed meat products to improve protein functionality through the immobilization of free water and it increased the stability of the finished products (Prabhu and Doerscher, 2000).

Ulu (2004) found that the addition of carrageenan at levels of 0.5–1 per cent in low-fat meatballs improved their water binding ability and hence yield and texture.

2.6.4. Texture Analysis

As fat content of ground beef patties decreases tenderness also decreases (Allen *et al.*, 1999). Troutt *et al.* (1992) reported that Warner Bratzler Shear and Lee Kramer Shear forces decreased as fat level increased in beef patties. 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. 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).

Chavez (1983) observed an increased collagen level in hamburger/patties which decreased the mean shear force. Meullenet *et al.* (1994) reported that increasing the level of collagen fibers to 5 per cent had lowered shear stress values in low fat high added water chicken sausage. Osburn *et al.* (1997) and Osburn and Mandigo (1998) observed increased water binding and gelling ability of collagen had improved the texture by diluting the stronger binding myofibrillar protein in low fat formulations.

Egbert *et al.* (1991) found that addition of 0.5 per cent iota-carrageenan resulted in decreased shear force value in cooked patties. Many researchers have

noted that kappa and iota carrageenans improved the texture of low-fat meat products, since these compounds create complexes with water and proteins (Cofrades *et al.*, 2000). Candogan and Kolsarici (2003) noted only slight improvements in the texture of low-fat frankfurters caused by carrageenan addition. In low fat meat balls it was observed that addition of carrageenan improved the water-binding capacity and texture (Ulu, 2004). Carrageenan as a fat substitute causes reduction in cooking and storage weight losses, increase hardness, gumminess and chewiness (Cierach *et al.*, 2009).

Hart and Price (1993) reported that addition of tapioca starch in low fat beef burger gave desirable tenderness. Bullock *et al.* (1994) used non meat ingredients in low fat ground beef patties and observed improvement in tenderness by addition of modified tapioca starch compared to pea flour, a blend of xanthan and locust bean gum or iota- or kappa- carrageenan. Burger containing tapioca starch, modified food starch derived from waxy maize, and oat fibre significantly reduced Instron shear force values (Desmond and Buckley, 1998). Khalil *et al.* (2000) found that incorporation of starch with water in low fat beef patties formulations improved the instrumental texture characteristics.

Troy *et al.* (1999) studied texture analyser using Instron Universal Testing Machine using two blade attachment: Warner- Bratzler shear and Multi Blade Kramer shearing device, attached to a 500N and 2 kN load cell, respectively. The 2.5cm wide section removed from burger for both type of blade and applied cross head speed 25cm per min and 20 cm per min, respectively.

Lopez *et al.* (2006) determined the texture of ostrich burgers by using Texture Profile Analyser performed on raw samples at chilled temperature. A cubic sample (1×1×1cm) were cut from patties and subjected to a two-cycle compression test. Samples were compressed to 70 per cent of their original height with a compression load of 25kg, and a cross head speed of 20cm/min.

2.6.5. Colour

Arganosa *et al.* (1987) studied the colour measurements for both cooked and uncooked sausage with a Hunter Lab D25-9 Optical Sensor. Patties were allowed to stand for 1 h at 23°C to allow the exposed surface to oxygenate. Colour data were reported as the 'L' (lightness- darkness), 'a' (redness-greenness) and 'b' (yellowness-blueness) values.

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 in chicken patties had higher L* and b* values though a* values were not affected.

2.6.6. Proximate Composition and Nutritional Quality

Bater *et al.* (1992) studied the quality characteristics of hydrocolloid added oven roasted turkey breast and they reported that the products added kappa carrageenan, starch, kappa-carrageenan plus starch had a higher moisture but lower protein, fat and ash values than the control. A dilution of fat content occurred because the products with added kappa carrageenan held more than 50 per cent brine.

Osburn *et al.* (1997) reported that proximate composition of raw pork connective tissue was 44.24 per cent moisture, 28.29 per cent fat, 26.47 per cent protein and 269.47 mg/g total collagen and pH 7.25. Selvakumar (2009) studied the effect of pork skin collagen as a fat replacer in low fat frankfurter and estimated proximate composition of raw pork skin collagen as moisture 72.67, protein 25.77, fat 0.60, carbohydrate 0.38 and ash 0.58.

The raw low fat beef burger had a mean moisture content 69.8 per cent, mean fat content 8.6 and protein content range from 16.9 – 20.3 per cent (Troy *et al.*, 1999). Turhan *et al.* (2005) reported that proximate composition of low fat beef burger was 59.43 per cent moisture, 15.24 per cent protein, 21.42 per cent fat, 2.16 per cent ash and carbohydrates was 1.74 per cent.

2.6.7. Sensory Analysis

Troy *et al.* (1999) reported that the tenderness, moisture/juiciness and meat flavour were evaluated by means of eight–point structured scales (8= extremely tender, juice and bland, respectively). They conducted sensory evaluation of low fat beef burgers containing fat replacing functional blends including carrageenan, observed that low fat beef burgers were lower in juiciness, texture and overall acceptability on reduction of fat from 20 per cent to 5 per cent.

Sensory analysis parameters of low fat meat products including colour, flavor, 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).

Khalil *et al.* (2000) reported that low fat beef patties formulated with starch/water had higher sensory rating for juiciness and tenderness than the control.

Kumar and Sharma (2004) used an eight point Hedonic scale for the sensory evaluation of appearance, colour, flavour, juiciness, texture and overall acceptability of low fat pork patties containing barley flour as fat substitutes.

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

2.7. PACKAGING AND STORAGE

2.7.1. Packaging Materials

Packaging material used for meat products are usually plastics in which polymers with good O₂ barrier properties (polyamide) are incorporated with polymers with good humidity barrier and sealing properties such as polyethylene 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 (Dawson, 2001).

Turhan *et al.* (2005) studied effect of hazelnut pellicle in low fat beef burgers and burgers were wrapped with polyethylene film and held in a refrigerator temperature.

The formulation of fish burger with silver catfish filleting residue was packaged in low density polyethylene bags and stored inside plastic boxes at -20°C (Bochi *et al.*, 2008).

2.7.2. Packaging Systems and Storage Stability on the Quality of LFBB

2.7.2.1. Purge Loss

Two randomly selected slices from Deli roll of each treatment weighed initially and packaged in the vacuum sealed by vacuum packaging machine, prior to 48 h storage (4°C). After storage, the residual moisture was eliminated with the paper towel and industrial slices reweighed. Purge loss was reported as [(initial weight- final weight)/ initial weight] × 100 (Prabhu *et al.*, 2004).

Bishop *et al.* (1993) found that accumulation purge in vacuum packages increase in water content of low fat meat products. Purge loss was expressed as percentage.

Shand *et al.* (1994) found that kappa carrageenan addition at 0.5 and 1.0 per cent levels to structured beef role reduce purge in vacuum packaged slices during refrigerated storage.

Colmenero *et al.* (2000) 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.

As carrageenan concentration increase, a decrease in purge loss was observed, which only significant at day 14 and 28 when carrageenan concentration increases 0.3 to 0.7 per cent (Candogan and Kolsarici, 2003).

Purge was measured in case of frankfurter initial weight was measured before packaging. After storage, the sample were removed from the bag, the sample and bag were dried with a paper towel, and the sample were weight again (final weight). Purge was calculated as percentage of initial treatment (Prabhu *et al.*, 2004).

Incorporation of pork skin collagen at 1 per cent or above significantly decreased ($P < 0.05$) purge loss in low fat bologna (Schnell, 1999). Pork collagen effectively controlled purge in both the frankfurter and restructured ham after 4 and 8 weeks of refrigerated storage (Prabhu *et al.*, 2004). Carrageenan in low-fat frankfurters led to a reduction in cooking loss ($P > 0.05$) and purge during storage (Cierach *et al.*, 2009).

2.7.2.2. Lipid Oxidation

Younathan *et al.* (1980) found out that rancidity in cooked ground turkey stored at 4°C was effectively controlled by hot water extracts of onion peel,

although initial values of TBA were high. As onion peel contains numerous flavones aglycones making them effective antioxidants they suggested that onion can be incorporated into many ready-to-eat meat products.

Ahn *et al.* (1992) found that elimination of O₂ contact by hot vacuum packaging turkey meat patties inhibited the development of lipid oxidation during storage at 4⁰C as compared to loosely packaged patties.

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).

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

The TBA values of beef burger samples increases gradually during frozen storage, this increase could mainly attributed to the oxidation of lipids and formation of same TBA reactive compounds during the storage period (Stahnke, 1995).

Degree of lipid oxidation depends on the composition of the phospholipids, amount of polyunsaturated fatty acids and concentration of metal ions, oxygen, salts and other pro-oxidants (Calkins *et al.*, 2007).

Lipid oxidation was assessed through determination of primary (conjugated dienes and hydroperoxides) and secondary (malondialdehyde) oxidation products formed during frozen storage (Georgantelis *et al.*, 2007).

2.7.2.3. *Sensory Qualities*

Bullock *et al.* (1994) reported that after 24 weeks storage at -20°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.

The fat reduction from 30 per cent to 12 per cent or 5 per cent brought about decreased adhesiveness but addition of 1 per cent of carrageenan increased the adhesiveness of cooked meat balls (Ulu, 2004).

Both aerobically and vacuum packaged low fat turkey loaves with fat replacers on storage at $0 - 4^{\circ}\text{C}$ and -20°C for 40 and 60 days, respectively did not affect the appearance, colour, texture and mouth coating. But flavour, juiciness and overall acceptability reduced (Naseera, 2007).

2.7.2.4. *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°C .

Wu and Sheldon (1988) reported that turkey breast tissue contained 2 per cent lipid which remained constant over 4 days storage at 4°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°C to 7°C and 65 to 75 per cent relative humidity for seven 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 beef burger (LFBB) with fat replacers (FR) , viz., carrageenan (CG), tapioca starch (TS) and pregelatinized pork skin collagen (PSC) gel as a fat replacer and to assess its physical, chemical, nutritional and sensory qualities. The keeping quality of the burger under aerobic and vacuum packaging at 0-4°C and -20°C were also studied.

3.1. FORMULATION OF LOW FAT BEEF BURGER

3.1.1. Meat Ingredients

3.1.1.1. *Beef and Tallow*

Fresh hot deboned lean beef trimmings were collected from Holstein Friesian crossbred bulls of 2-5 years age, humanely slaughtered in the Meat Technology Unit, Department of Livestock Products Technology, College of Veterinary & Animal Sciences, Mannuthy. External fat, blood clots, tendons and visible connective tissue, bones and cartilage, if any, were removed from trimmings. The beef samples were randomly analysed for fat content and those with a mean fat content of 1.76 per cent were collected, stored at 0-4°C for 48h for conditioning and then kept frozen at -20 °C till use for the preparation of beef burger (BB). Tallow was prepared from kidney fat of the same bulls and stored at -20 °C till further use.

3.1.1.2. *Preparation of Pregelatinised Pork Skin Collagen*

Pork skin of sound quality was collected from ham and belly regions of Large White Yorkshire pigs of 60 to 100 kg. Subcutaneous fat was completely removed from pork skin and cut into pieces of 2.5cm × 2.5 cm size and was frozen at -20°C till subsequent use for gel preparation. 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 80°C for 45 min instead of 70°C for 30 min. The gel was cooled to room temperature and homogenized in a

domestic food processor (Sumeet, Mumbai). The prepared gel was stored at -20°C till the preparation of burger.

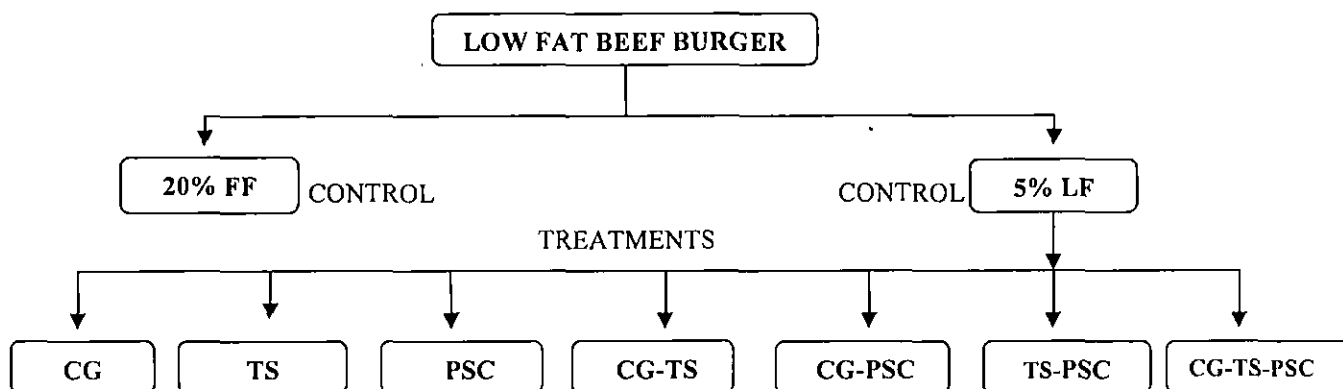
3.1.1.3. Nonmeat Ingredients

Carrageenan and tapioca starch were used as the fat replacers besides PSC. Carrageenan was collected from FMC Biopolymer, Rockland, USA and Tapioca starch from SPAC Tapioca Products Ltd. India. The other ingredients were good quality freshly ground onion, garlic, ginger, green chilli, salt, black pepper and rusk.

3.2. LOW FAT BEEF BURGER FORMULATION AND MANUFACTURE

Beef burgers were formulated in two different fat levels, viz., full fat 20 per cent and low fat 5 per cent. Seven formulations of LFBB with 5 per cent fat were prepared with CG, TS, PSC and their blends according to the scheme presented in Fig. 1.

Fig. 1. Schematic representation of the experiment



FF- Full Fat; LF- Low Fat

CG- Carrageenan, TS- Tapioca Starch, PSC- Pregelatinised Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS;

CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;

CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

The formulations for LFBB with seasonings and various blends of fat replacers are presented in Table 1.

Table 1. Formulations for low fat beef burgers with fat replacers

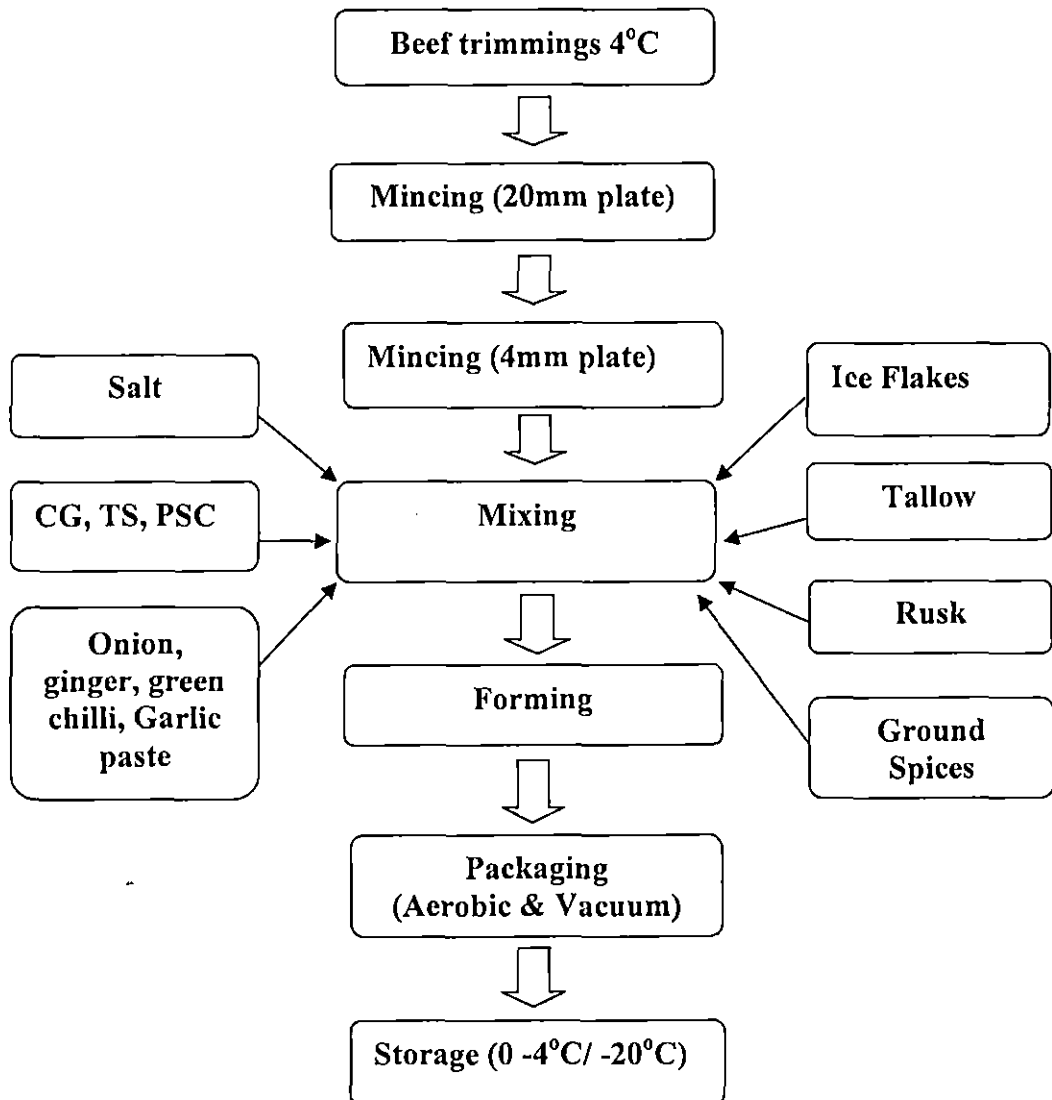
Ingredients	Formulations (g)								
	FF	LF	CG	TS	PSC	CG-TS	CG-PSC	TS-PSC	CG-TS-PSC
Lean Beef trimmings	738	881	875	862	864	857	858	846	840
Tallow	183	39	39	39	32	39	32	32	32
Water (8%)	80	80	80	80	80	80	80	80	80
Salt (1.2%)	14	14	14	14	14	14	14	14	14
Black Pepper (1.5%)	18	18	18	18	18	18	18	18	18
Onion Paste (4%)	48	48	48	48	48	48	48	48	48
Garlic Paste (3.3%)	40	40	40	40	40	40	40	40	40
Ginger Paste (1.2%)	14	14	14	14	14	14	14	14	14
Green chilli (2%)	24	24	24	24	24	24	24	24	24
Rusk (3.5%)	42	42	42	42	42	42	42	42	42
CG (0.5%)	-	-	6.0	-	-	6.0	6.0	-	6.0
TS (1.5%)	-	-	-	18	-	18	-	18	18
PSC (2%)	-	-	-	-	24	-	24	24	24

FF- 20% Full Fat; LF- 5% Low Fat

CG- Carrageenan, TS- Tapioca Starch, PSC- Pregelatinised Pork Skin Collagen

The steps in the manufacture of LFBB are illustrated in the flow diagram Fig.2.

Fig. 2. Flow diagram for the preparation of low fat beef burger



Beef trimmings stored at -20°C were thawed under refrigeration and coarsely ground through a 20mm grinder plate followed through a 4mm grinder plate in a meat mincer (MADO Primus Model MEW 613, Germany). For each formulation 1.2 kg burger mix was prepared as shown in Table 1. Appropriate amounts of lean beef trimmings and tallow were mixed with salt, black pepper, onion, garlic, ginger, green chilli, rusk and ice flakes. The fat replacers, *viz.*, CG,

TS, PSC and their blends were mixed thoroughly with the meat mix according to the formulary. The temperature of the burger mix was maintained between 2- 4°C and beef burgers of about 30g weight, 1.0cm thickness and 6cm diameter was manually formed using a mould.

All the burgers were packaged under two different packaging systems, *viz.*, Aerobic Packaging (AP) and Vacuum Packaging (VP). In the former, high density polyethylene (HDPE) pouches of 50 μ was used while in the latter, polyethylene-polyamide (PEPA) pouches (oxygen transmission rate: 208 cc/m²/24hr, water transmission rate: 5g/cc/24hr at 38°C and relative humidity 90 per cent using a single chamber vacuum packaging machine (Sevana, Kochi, India). The packaged LFBB were stored at 0 – 4°C and at -20°C for further studies.

Various physico-chemical parameters, *viz.*, pH, cook yield (CY), cook loss (CL), fat retention percentage (FRP), moisture retention percentage (MRP), dimensional shrinkage (DS), water holding capacity (WHC), Warner-Bratzler Shear Force (WBSF), colour (Hunter Lab), proximate and mineral composition and nutritional value, purge loss (PL), Thiobarbituric Acid Reactive Substances (TBARS) and sensory quality were assessed on day 0, 10, 20 and 30 of storage at 0-4°C and -20°C or till spoilage, whichever is earlier. Six trials of the experiment were conducted.

3.3. QUALITY EVALUATION OF LOW FAT BURGERS

3.3.1. Physico-chemical and Nutritional Characteristics

3.3.1.1. pH

The pH of the beef burgers from all the formulations, before and after cooking was determined using a combined electrode digital pH meter (μ pH system 362, Systronics, India).

3.3.1.2. *Cooking Characteristics*

The cook yield was calculated according to Berry (1992). Weights of LFBB before and after cooking from all formulations were recorded.

Cook yield (%) = (cooked weight/uncooked weight) x 100.

Cook loss (%) = 100 - cooking yield.

The dimensional shrinkage of LFBB was measured using Vernier Calipers in mm and the mean of five readings of the diameter of five burgers were taken before and after cooking. Dimensional shrinkage (%) = (Diameter before cooking - Diameter after cooking / Diameter before cooking) x 100

The fat and moisture retention were determined as describe as Alesson-Carbonell *et al.* (2005).

Fat retention percentage (FRP) = cooked weight × % fat in cooked burger x 100 / raw weight × % fat in raw burger

Moisture retention percentage (MRP) = cooked weight × % moisture in cooked burger x 100 / raw weight × % moisture in raw burger

3.3.1.3. *Water Holding Capacity*

Water Holding Capacity was calculated according to Liangi and Chen (1991). Ten gram samples of each uncooked BB were placed in glass jars and heated to 90°C for 10 min in a thermostatically controlled water bath. Samples were carefully removed from the jars and cooled to 4°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 RCF for 10 min, samples were weighed again.

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 burger.

3.3.1.4. Warner-Bratzler Shear Force

The WBSF values of the cooked burgers from all formulations were recorded as per the method of Troy *et al.* (1999). Burgers after cooling over night to 0-4°C were cut into two 2.5 cm wide sections. Each section was sheared in five separate locations using Warner-Bratzler shear having a cross head speed of 250 mm/min attached to a Universal Testing Machine (Shimadzu Texture Analyser Model EZ Shimadzu Corporation, Kyoto, Japan). The mean value of shear force in Newton (N) was recorded.

3.3.1.5. Colour

Colour of the cooked burger 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° 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 were measured thrice and mean values were taken.

3.3.1.6. Proximate Composition

Moisture, fat, protein and ash were evaluated according to AOAC (1990). The proximate composition of beef trimmings, PSC, uncooked and cooked BB of each formulation was determined in duplicate and values were expressed in g/100g of sample on as is basis.

Moisture was determined by weight loss after 16 h drying in a hot air oven at 102°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 content by Kjeldahl Block Digestion Method (KEL Plus, Model KES 6L, Pelican Equipments, Chennai, India.).

Ash was determined by weight loss after 2.5 h drying in a muffle furnace at 600°C. The amount of carbohydrate was calculated as 100 minus sum of the percentage of moisture, fat, protein and ash. Moisture, fat and protein contents of burger were determined on day 0, 10, 20 and 30 at 0-4°C and -20°C storage. The proximate composition was expressed in as-is-basis.

3.3.1.7. Mineral Composition

After wet digestion of 0.5g beef burger, Na, K, Ca and P contents were estimated and expressed in mg/ 100g of beef burger on as is basis. The Na and K contents were estimated using a Flame Photometer (Systronics flame photometer 128, Ahmadabad, India). The Ca content was determined using Atomic Absorption spectrophotometer (Perkin Elmer, 3110, US Instrument division, Norwalk, USA) at a wavelength of 422.7nm (Beaty and Kerbar, 1993). Phosphorous was determined by ANSA method (Fiske and Subbarow, 1925).

Effect of cooking on proximate composition of LFBB 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 beef burger formulation 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 = calories from fat + protein + 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 and Minerals of burger

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

Per cent daily value of protein in burger = per cent protein in burger/RDA of the protein. RDA of protein was taken as 60g (ICMR, 1990) and that of Na- 2400mg; K- 3500mg (Code of Federal Regulations, 1995), Ca-800mg and P- 800mg (NRC, 1989).

3.3.1.9. Purge Loss

Purge loss was determined by the method of Prabhu *et al.* (2004). Determination of purge (moisture) loss consisted of weighing each type of uncooked BB stored at 0-4°C and -20°C on d 10, 20 and 30. Two burgers were taken out of the package and carefully blotted with tissue paper to eliminate any liquid on the surface and weighed. The difference in weight expressed as percentage of initial weight prior to storage was reported as purge loss.

3.3.1.10. Thiobarbituric Acid Reactive Substances

TBARS in LFBB were determined by the extraction method of Witte *et al.* (1970) with a slight modification. The extraction supernatant was centrifuged (Etek 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, expressed as mg malonaldehyde per kg of burger were calculated by multiplying the absorbance with a factor of 5.2. The TBARS value of uncooked BB was measured on day 0, 10, 20 and 30 of storage at 0-4°C and -20°C under aerobic and vacuum packaging.

3.3.2. Sensory Evaluation

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

The burger after refrigerated thawing were low fat fried in refined sunflower oil, both sides for 4 min each to an internal temperature of 75 to 80°C. Panelists evaluated hot burger and recorded scores for the samples in an eight point Hedonic scale for appearance and color, flavour, texture, saltiness, juiciness, mouth coating and overall acceptability. The sensory evaluations were conducted on day 0, 10, 20 and 30 of storage at 0-4°C and -20°C, respectively.

3.4. COST OF PRODUCTION

The cost of production of the different burger formulations at laboratory level was calculated from the cost of meat and other ingredients.

3.5. STATISTICAL ANALYSIS

Data obtained were analysed by one way 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 beef burger

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

Attributes	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6	Sample 7	Sample 8	Sample 9
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

Low Fat Beef Burgers (LFBB) were formulated with two different fat levels, *viz.*, full fat (FF) 20 per cent and low fat (LF) 5 per cent. Seven formulations of LFBB with 5 per cent of fat were prepared with 0.5 per cent Carrageenan (CG), 1.5 per cent Tapioca starch (TS), 2 per cent pregelatinised pork skin collagen (PSC) and their blends as Fat Replacers (FR). Various physico-chemical, nutritional and organoleptic qualities and the shelf life of the aerobic and vacuum packaged LFBB were studied on d 0, 10, 20 and 30 of storage at 0-4°C and -20°C. The results obtained are narrated and supported by Tables and Figures in this Chapter.

4.1. PHYSICO-CHEMICAL CHARACTERISTICS OF LFBB

4.1.1. pH

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

The pH of uncooked beef burger in all formulations ranged from 6.06 ± 0.02 to 6.26 ± 0.01 . Among this, the formulation with PSC showed 6.26 ± 0.01 which was significantly ($P < 0.05$) higher than all other formulations except CG-TS and TS-PSC. The pH of FF control was 6.06 ± 0.02 which was significantly ($P < 0.05$) the lowest among all formulations.

pH of cooked burger in all formulations were significantly ($P < 0.05$) higher than their corresponding uncooked burgers and ranged from 6.18 ± 0.02 to 6.42 ± 0.02 . Cooked burgers with FF and LF controls were significantly ($P < 0.05$) acidic among all formulations.

pH of all the burgers were slightly acidic (low acid) before and after cooking although cooking significantly reduced the acidity in all formulations. The burgers with FR were the least acidic compared to FF and LF.

4.1.2. Cooking Characteristics

The percentage of cook yield (CY), cook loss (CL), dimensional shrinkage (DS), Fat retention percentage (FRP) and Moisture retention percentage (MRP) are given in Table 3 and trends were illustrated in Fig.4 and 5.

4.1.2.1. Cook Yield and Cook Loss

CY of burgers with CG-TS and CG-TS-PSC were 86.47 ± 0.09 and 85.84 ± 0.25 , respectively which were significantly ($P < 0.05$) the highest among all formulations. There was improvement in CY by the addition of FR. LFBB with combinations of FR showed significantly ($P < 0.05$) higher CY than those with single FR, evidently CG-TS and CG-TS-PSC.

The values for CL showed negative correlations with that of cooking yield. The percentage CL in FF and LF controls were 36.61 ± 0.31 and 32.23 ± 0.13 , respectively, which were significantly ($P < 0.05$) higher than in other formulations.

4.1.2.2. Dimensional Shrinkage

The DS of BB with CG-PSC and CG-TS-PSC was significantly ($P < 0.05$) lowest which were recorded as 13.09 ± 0.24 and 13.21 ± 0.89 , respectively, among all formulations.

LFBB with combinations of FR showed significantly ($P < 0.05$) lower DS than those with single FR.

4.1.2.3. Fat Retention Percentage

FRP were significantly ($P < 0.05$) highest for the burgers with CG-TS-PSC, which was 97.66 ± 0.43 , followed by the other formulations, viz., CG-PSC, LF, TS-PSC, CG-TS, CG, PSC, TS and FF, in the decreasing order of FRP.

4.1.2.4. Moisture Retention Percentage

Addition of blends of FR significantly ($P < 0.05$) increased MRP of burgers compared to single FR and controls. Among the blends, CG-PSC and CG-TS-PSC recorded MRP as 74.41 ± 0.15 and 74.36 ± 0.20 , respectively. This was followed by TS-PSC and CG-TS in the decreasing order of MRP.

4.1.3. Water Holding Capacity

The WHC expressed as percentage is given in Table 3 and the trend is shown in Fig.6.

Burgers with combinations of FR showed significantly ($P < 0.05$) higher WHC compared to single FR. The beef burgers with CG-TS-PSC were 95.36 ± 0.09 comparable to that of FF control which was 95.51 ± 0.05 and not significantly ($P > 0.05$) different. Among the single FR, TS showed significantly highest WHC of 94.73 ± 0.27 . LF control had the significantly ($P < 0.05$) lowest WHC among all.

4.1.4. Warner-Bratzler Shear Force

WBSF in Newton (N) of different formulations of LFBB are given in Table 3 and variations in the shear force are presented in Fig.7.

The WBSF of beef burgers with CG-TS-PSC was 5.30 ± 0.04 which did not significantly ($P > 0.05$) differ from FF which was 5.35 ± 0.03 . Although, both were significantly ($P < 0.05$) lower than the other formulations, the former with FR was the tenderest. Those burgers without any FR were the toughest with WBSF 6.73 ± 0.02 . Among the LFBB with combinations of FR, in the decreasing order of tenderness were, CG-TS, CG-PSC and TS-PSC. If PSC alone was used as FR, the burgers were significantly ($P < 0.05$) tenderer than CG and TS.

4.1.5. Colour

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

Table 3. pH, cooking characteristics, WHC and WBSF values of different formulations of low fat beef burger

Parameters	Formulations								
	FF	LF	CG	TS	PSC	CG-TS	CG-PSC	TS-PSC	CG-TS-PSC
pH before cooking	6.06 ^a ± 0.02	6.09 ^a ± 0.02	6.13 ^{abc} ± 0.02	6.12 ^{ab} ± 0.03	6.26 ^d ± 0.01	6.18 ^{bcd} ± 0.01	6.10 ^{ab} ± 0.03	6.21 ^{cd} ± 0.02	6.14 ^{abc} ± 0.03
pH after cooking	6.18 ^a ± 0.02	6.23 ^a ± 0.02	6.34 ^b ± 0.02	6.33 ^b ± 0.02	6.42 ^b ± 0.02	6.38 ^b ± 0.02	6.32 ^b ± 0.03	6.38 ^b ± 0.02	6.32 ^b ± 0.03
CY (%)	63.55 ^a ± 0.29	67.77 ^b ± 0.13	80.78 ^c ± 0.32	77.71 ^c ± 0.38	77.71 ^c ± 0.81	86.47 ^c ± 0.09	83.07 ^d ± 0.02	82.22 ^d ± 0.22	85.84 ^c ± 0.25
CL (%)	36.61 ^c ± 0.31	32.23 ^c ± 0.13	19.22 ^c ± 0.33	22.20 ^d ± 0.40	22.29 ^d ± 0.33	13.52 ^a ± 0.12	16.92 ^b ± 0.46	17.77 ^b ± 0.18	14.45 ^a ± 0.25
DS (%)	30.92 ^c ± 0.59	19.58 ^d ± 0.33	17.19 ^c ± 0.23	16.46 ^c ± 0.70	18.10 ^c ± 0.33	14.85 ^b ± 0.11	13.09 ^a ± 0.24	13.92 ^{ab} ± 0.29	13.21 ^a ± 0.89
FRP (%)	90.80 ^a ± 0.24	95.85 ^c ± 0.19	93.29 ^c ± 0.25	92.10 ^b ± 0.22	92.35 ^b ± 0.12	93.64 ^c ± 0.17	96.22 ^c ± 0.14	94.72 ^d ± 0.25	97.66 ^f ± 0.43
MRP (%)	68.31 ^c ± 0.13	66.36 ^a ± 0.15	68.31 ^c ± 0.22	67.52 ^b ± 0.16	68.48 ^c ± 0.23	71.41 ^d ± 0.14	74.41 ^f ± 0.15	72.27 ^e ± 0.16	74.36 ^f ± 0.20
WHC (%)	95.51 ^d ± 0.05	93.09 ^a ± 0.10	94.21 ^b ± 0.05	94.73 ^c ± 0.27	94.18 ^b ± 0.14	94.92 ^b ± 0.10	94.98 ^{cd} ± 0.23	94.94 ^{cd} ± 0.03	95.36 ^d ± 0.09
WBSF (N)	5.35 ^a ± 0.03	6.73 ^c ± 0.02	6.21 ^d ± 0.15	6.20 ^d ± 0.01	5.90 ^{bc} ± 0.03	5.83 ^b ± 0.02	5.94 ^c ± 0.01	6.11 ^d ± 0.01	5.30 ^a ± 0.04

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC;
 CY- Cook yield; CL- Cook Loss; DS- Dimensional Shrinkage; FRP- Fat Retention Percentage;
 MRP- Moisture Retention Percentage; WHC- Water Holding Capacity; N- Newton; WBSF- Warner-Bratzler Shear Force

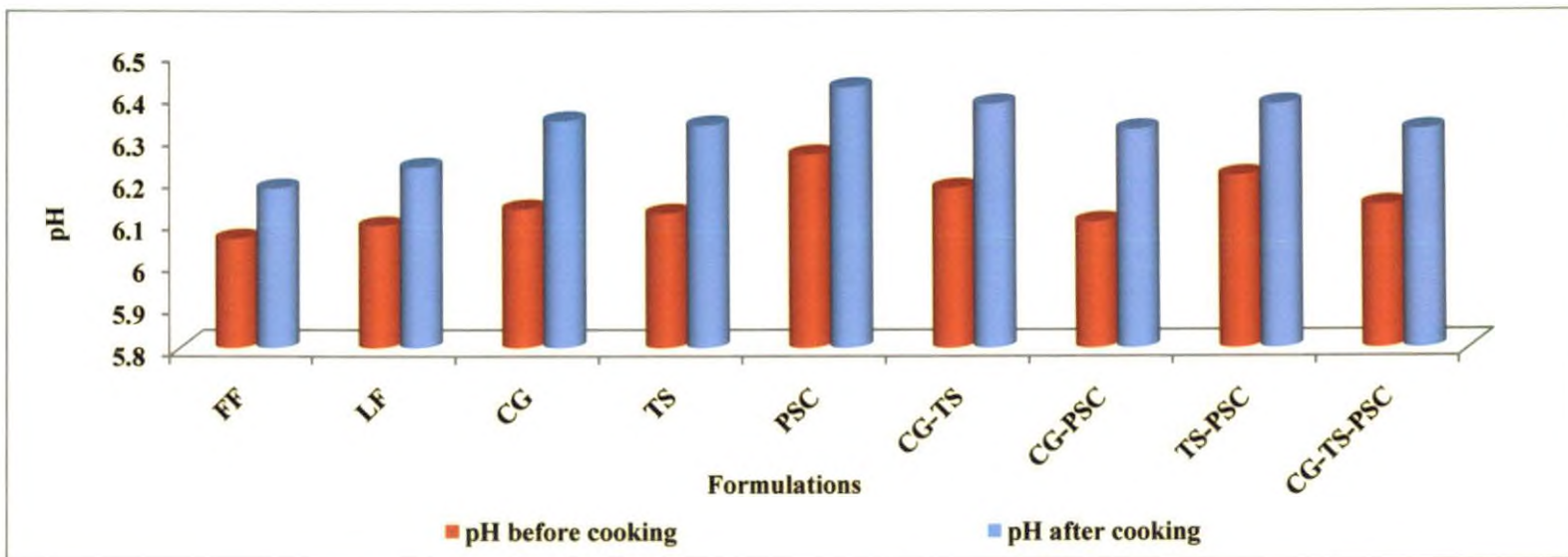


Fig 3. pH before and after cooking of low fat beef burger of different formulations

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; LFBB-Low fat beef burger

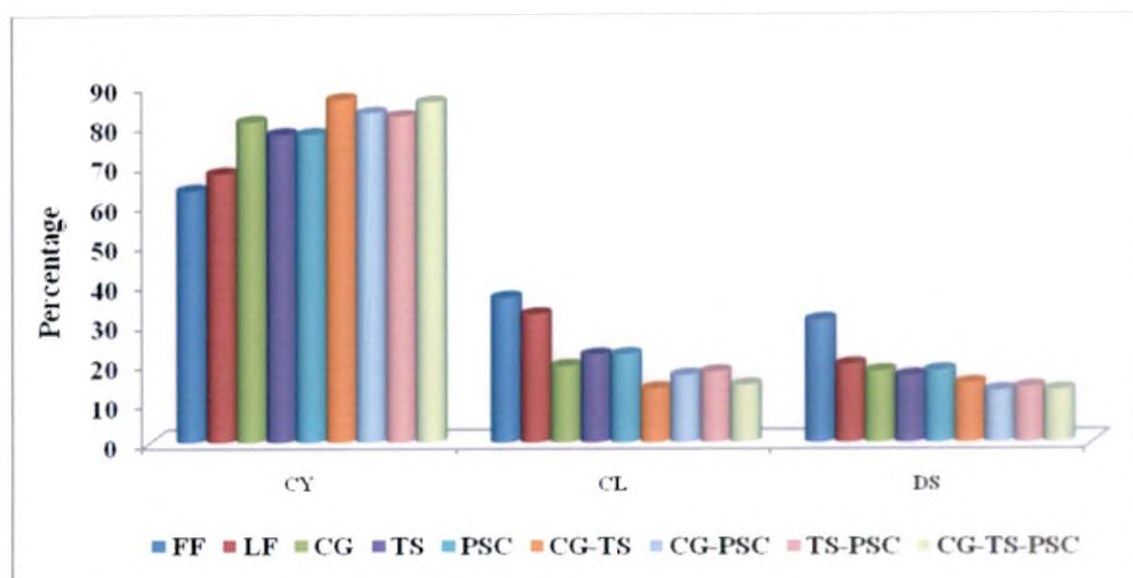


Fig. 4. Cook yield, cook loss and dimensional shrinkage of different formulations of low fat beef burger

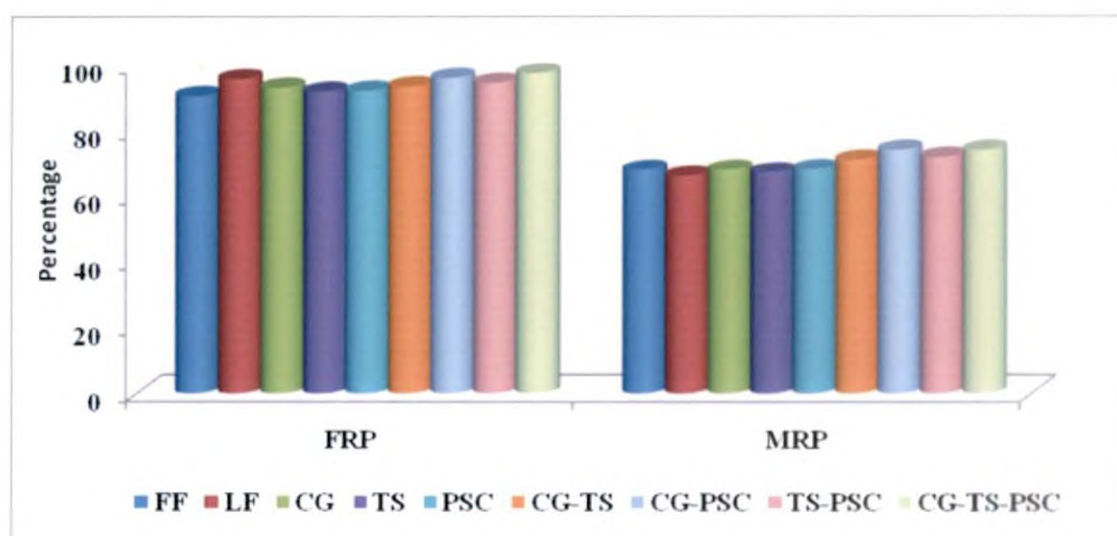
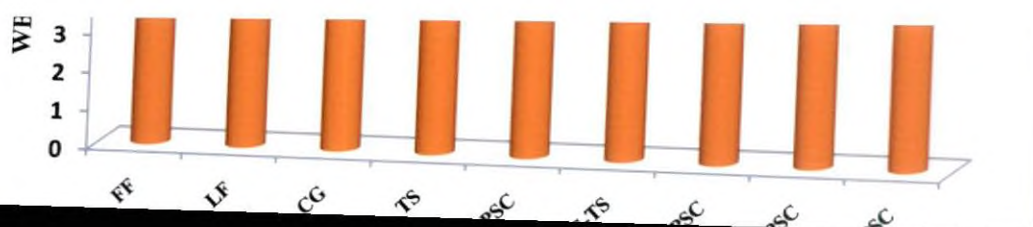


Fig. 5. Fat and moisture retention percentage of different formulations of low fat beef burger

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; CY- Cook yield; CL- Cook Loss; DS- Dimensional Shrinkage; FRP- Fat Retention Percentage; MRP- Moisture Retention Percentage



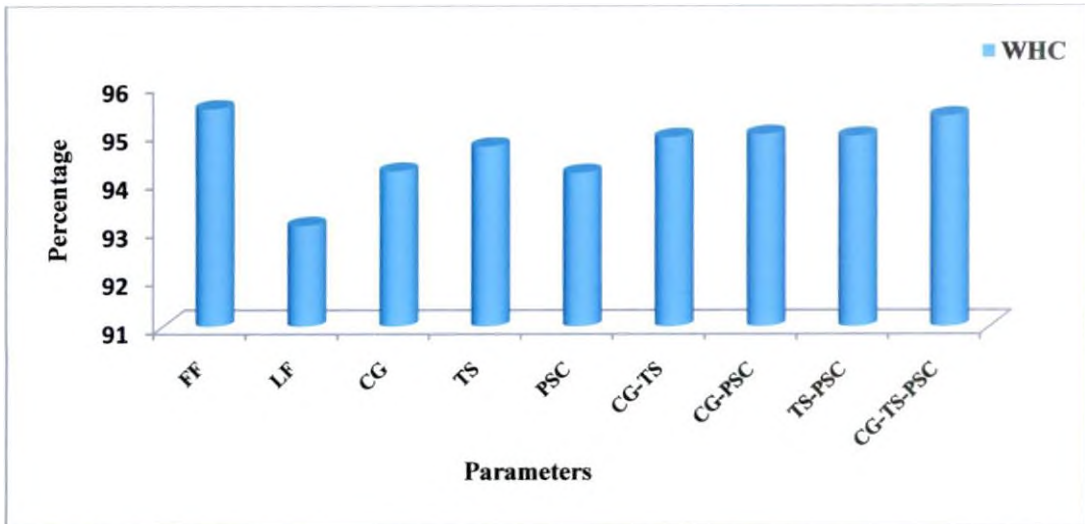


Fig.6. Water Holding Capacity of different formulation of low fat beef burger

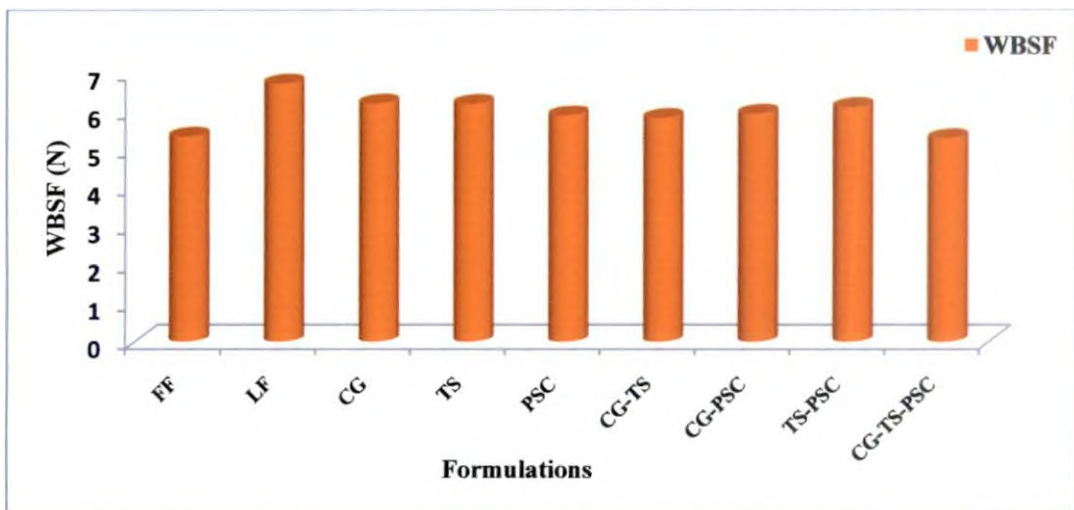


Fig. 7. Warner Bratzler Shear Force value of different formulations of low fat beef burger

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG
 & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC;
 WBSF- Warner Bratzler Shear Force; N – Newton.

The L* (lightness) values of burgers with CG-TS-PSC was 36.57 ± 0.38 and of FF control was 36.70 ± 0.61 which were significantly ($P < 0.05$) the highest of all formulations. The a* (redness) value was 7.25 ± 0.12 which was significantly ($P < 0.05$) the highest recorded in the burger with PSC compared to remaining formulations. The b* (yellowness) values of formulation with CG-TS-PSC was significantly ($P < 0.05$) the highest among all which was 10.65 ± 0.20 .

4.2. PROXIMATE AND MINERAL COMPOSITION

4.2.1. Lean Beef Trimming and Pork Skin Collagen

The proximate composition of lean beef trimmings and pork skin collagen are presented in Table 5 and in Fig.9.

Table 5. Proximate composition of lean beef trimming and pork skin collagen

Parameters (%)	Lean beef trimming	Pork skin collagen
Moisture	76.58 ± 0.37	72.52 ± 0.44
Protein	19.29 ± 0.30	25.61 ± 0.23
Fat	1.76 ± 0.01	0.58 ± 0.03
Carbohydrate	1.2 ± 0.01	0.38 ± 0.02
Ash	1.18 ± 0.01	0.58 ± 0.02

The fat content in the lean beef trimmings and pork skin collagen were 1.76 ± 0.01 and 0.58 ± 0.03 per cent, respectively.

4.2.2. Low Fat Beef Burger

4.2.2.1. Uncooked Beef Burger

Proximate and mineral composition in 100g of beef burger of all nine formulations before cooking are given in Table 6 and illustrated in Fig.10 and 11.

Proximate composition of LFBB with combinations of FR and single FR did not show any significant ($P > 0.05$) difference in moisture, protein, fat, carbohydrate and ash. In all LFBB the fat content ranged between 5.05 ± 0.08 and 5.85 ± 0.32 . But in FF formulations, fat was 20.53 ± 0.15 . Addition of FR in LFBB significantly ($P < 0.05$) increased protein content in all formulations.

Table 4. Hunter Lab colour values of different formulations of low fat beef burger

Formulations	Parameters		
	L*	a*	b*
FF	36.70 ^c ± 0.61	6.39 ^{ab} ± 0.59	7.78 ^c ± 0.30
LF	32.76 ^a ± 0.87	6.53 ^{ab} ± 0.11	6.75 ^a ± 0.21
CG	34.03 ^{ab} ± 0.40	6.01 ^{ab} ± 0.17	6.13 ^a ± 0.21
TS	34.45 ^{bc} ± 0.35	5.06 ^a ± 0.29	6.77 ^a ± 0.32
PSC	33.39 ^{ab} ± 0.48	7.25 ^c ± 0.12	7.17 ^{bc} ± 0.04
CG-TS	32.53 ^a ± 0.46	6.83 ^{ab} ± 0.43	7.09 ^c ± 0.17
CG-PSC	32.61 ^a ± 0.44	6.76 ^{ab} ± 0.24	6.98 ^b ± 0.34
TS-PSC	32.73 ^a ± 0.57	6.33 ^{ab} ± 0.31	7.86 ^c ± 0.29
CG-TS-PSC	36.57 ^c ± 0.38	6.75 ^{ab} ± 0.13	10.65 ^d ± 0.20

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

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

L*- Lightness; a*- Redness; b*- Yellowness

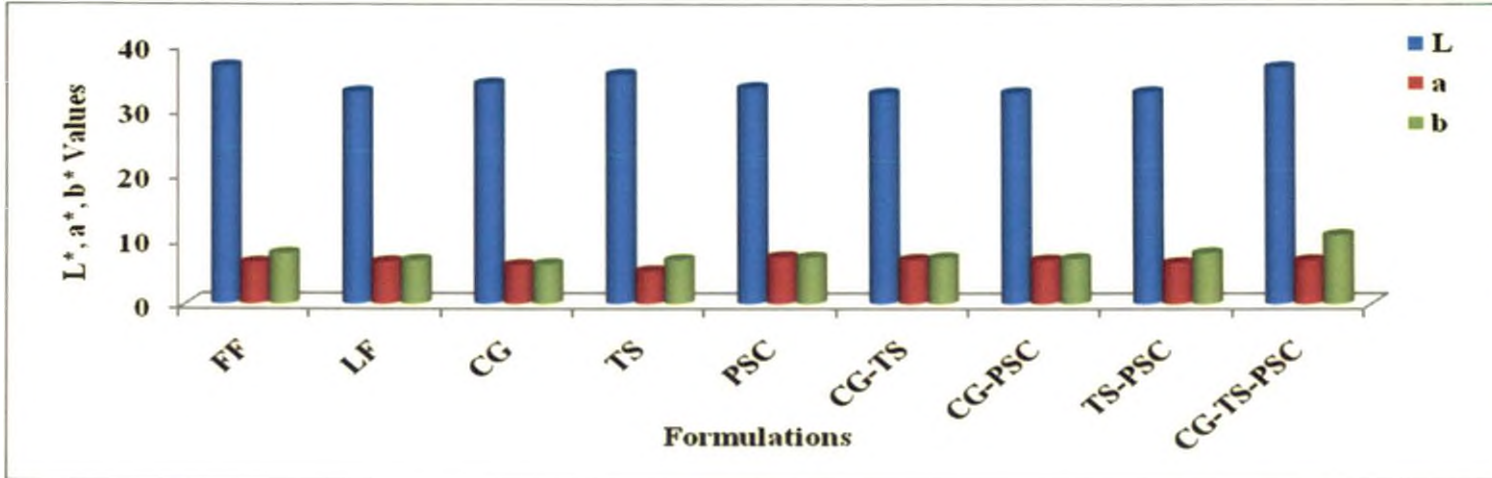


Fig. 8. Hunter L*, a* b* colour values of different formulations of low fat beef burger

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5%TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; WBSF- Warner Bratzler Shear Force Value;
 N – Newton; L*- Lightness; a*- Redness; b*- Yellowness.

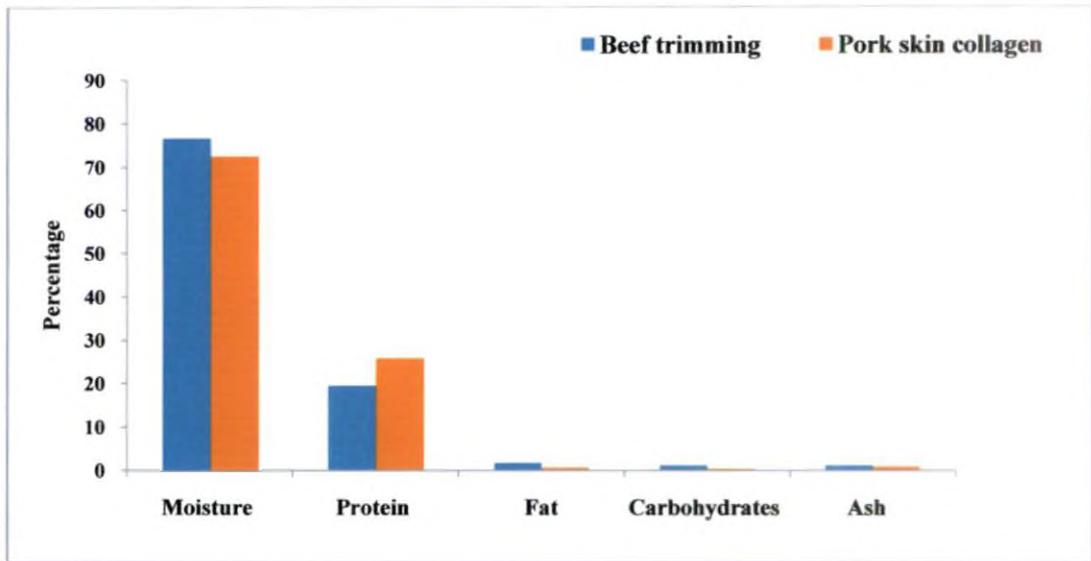


Fig.9. Proximate composition of lean beef trimmings and pork skin collagen

Ash content in beef burger ranged from 1.62 ± 0.04 to 2.06 ± 0.05 . CG-TS-PSC had significantly ($P < 0.05$) the highest mineral content among the other formulations. Na content was highest in CG-TS-PSC, K in CG-TS, Ca in CG and P in TS and was significantly ($P < 0.05$) the highest among all formulations.

4.2.2.2. Cooked Beef Burger

Proximate and mineral composition in 100g of all formulations of cooked beef burger of are given in Table 7 and illustrated in Fig.12 and 13.

Mean moisture percentage in all formulations ranged from 58.12 ± 0.04 to 68.14 ± 0.36 . Burgers with FF control was significantly ($P < 0.05$) the lowest and CG was significantly ($P < 0.05$) highest among all formulations.

The protein content of burgers with FR was significantly ($P < 0.05$) higher than in FF. The Fat content of FF burgers was significantly ($P < 0.05$) the highest, which was 19.83 ± 0.49 among all formulations. While the LFBB contained fat in the range of 5.45 ± 0.06 to 6.26 ± 0.18 only. Carbohydrate of burger with FF and TS-PSC showed significantly ($P < 0.05$) lower and CG showed significantly ($P < 0.05$) higher carbohydrate content.

The ash content of burger with TS-PSC and CG-TS-PSC were significantly ($P < 0.05$) higher which were 2.12 ± 0.02 and 2.14 ± 0.09 , respectively. Na and K content of beef burger with CG-TS-PSC was significantly ($P < 0.05$) the higher which was recorded as 946.3 ± 9.83 of Na and 572.1 ± 3.54 of K, respectively. Ca of burger with CG was 42.21 ± 0.73 which was significantly ($P < 0.05$) the highest among all formulation and P of burger with CG-TS-PSC was 216.5 ± 1.55 which was significantly ($P < 0.05$) the highest among all formulations.

Table 6. Proximate and mineral composition of different formulations of uncooked low fat beef burger

Formulations	Parameters, %								
	Moisture	Protein	Fat	Carbohy drate	Ash	Na (mg)	K (mg)	Ca (mg)	P (mg)
FF	59.98 ^a ± 0.25	16.82 ^a ± 0.55	20.53 ^c ± 0.15	1.03 ^a ± 0.35	1.62 ^a ± 0.04	749.7 ^a ± 1.06	468.0 ^b ± 3.48	36.22 ^{bc} ± 0.82	126.9 ^{ab} ± 0.91
LF	69.31 ^b ± 0.35	19.23 ^{bc} ± 0.21	5.67 ^{ab} ± 0.27	4.10 ^b ± 0.19	1.67 ^{ab} ± 0.06	758.5 ^a ± 2.91	483.5 ^{bc} ± 2.06	30.05 ^a ± 0.25	122.5 ^a ± 1.42
CG	69.43 ^b ± 0.95	17.72 ^b ± 0.78	5.05 ^a ± 0.08	5.97 ^b ± 0.08	1.81 ^{abc} ± 0.07	785.7 ^{bc} ± 1.88	507.7 ^{cde} ± 1.43	38.52 ^c ± 1.19	171.6 ^b ± 2.60
TS	67.82 ^b ± 0.47	19.13 ^{bc} ± 0.61	5.85 ^{ab} ± 0.32	5.42 ^b ± 0.08	1.75 ^{ab} ± 0.02	798.7 ^{cd} ± 1.25	505 ^{cd} ± 3.27	34.44 ^b ± 0.60	198.1 ^f ± 1.74
PSC	69.38 ^b ± 0.30	20.10 ^c ± 0.26	5.73 ^{ab} ± 0.16	3.03 ^{ab} ± 0.57	1.75 ^{ab} ± 0.06	825 ^e ± 1.43	525 ^{de} ± 3.90	34.32 ^b ± 1.32	181.9 ^d ± 2.33
CG-TS	68.14 ^b ± 0.50	17.78 ^b ± 0.67	5.26 ^b ± 0.30	5.94 ^{bcd} ± 0.14	1.86 ^{bcd} ± 0.06	756.5 ^a ± 1.05	584.2 ^e ± 2.52	35.16 ^{bc} ± 0.92	160.6 ^b ± 2.27
CG-PSC	68.55 ^b ± 0.61	19.29 ^{bc} ± 0.42	5.59 ^{ab} ± 0.25	4.61 ^b ± 0.19	1.94 ^{cde} ± 0.08	783 ^b ± 1.35	533.7 ^{ef} ± 2.01	37.00 ^{bc} ± 1.07	189.7 ^e ± 1.05
TS-PSC	68.09 ^b ± 0.41	20.07 ^c ± 0.59	5.61 ^{ab} ± 0.31	3.18 ^{ab} ± 0.43	2.03 ^{de} ± 0.06	804.7 ^d ± 1.03	434.7 ^a ± 4.60	28.32 ^a ± 0.13	156.9 ^b ± 2.40
CG-TS-PSC	67.46 ^b ± 0.51	20.54 ^c ± 0.30	5.45 ^{ab} ± 0.22	4.48 ^b ± 0.88	2.06 ^e ± 0.05	879.2 ^f ± 1.19	551.2 ^f ± 5.64	36.87 ^{bc} ± 0.26	187.4 ^{de} ± 265

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;

TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC- 0.5% CG, 1.5% TS & 2.0% PSC.

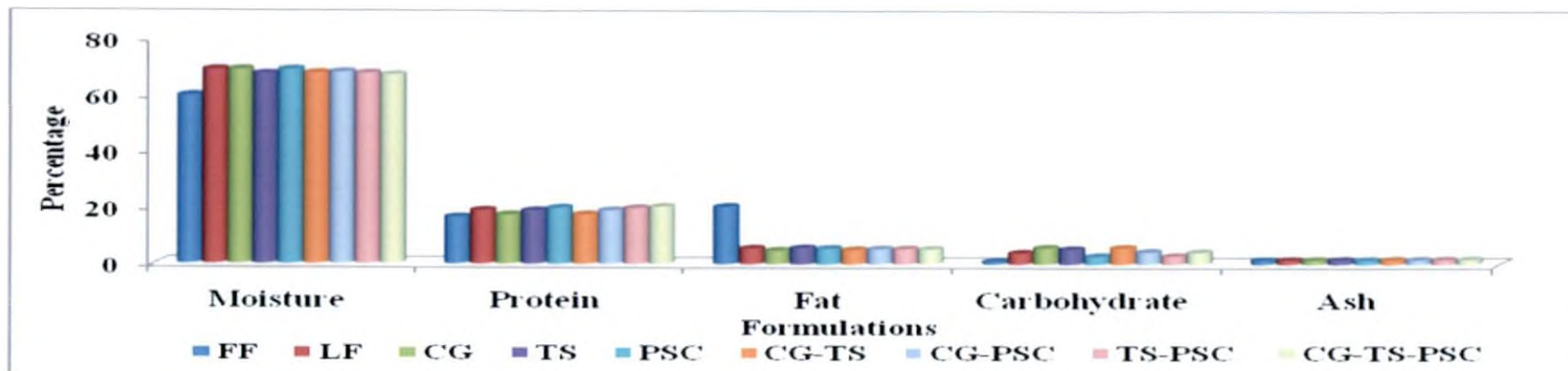


Fig. 10. Proximate composition of different formulations of uncooked low fat beef burger

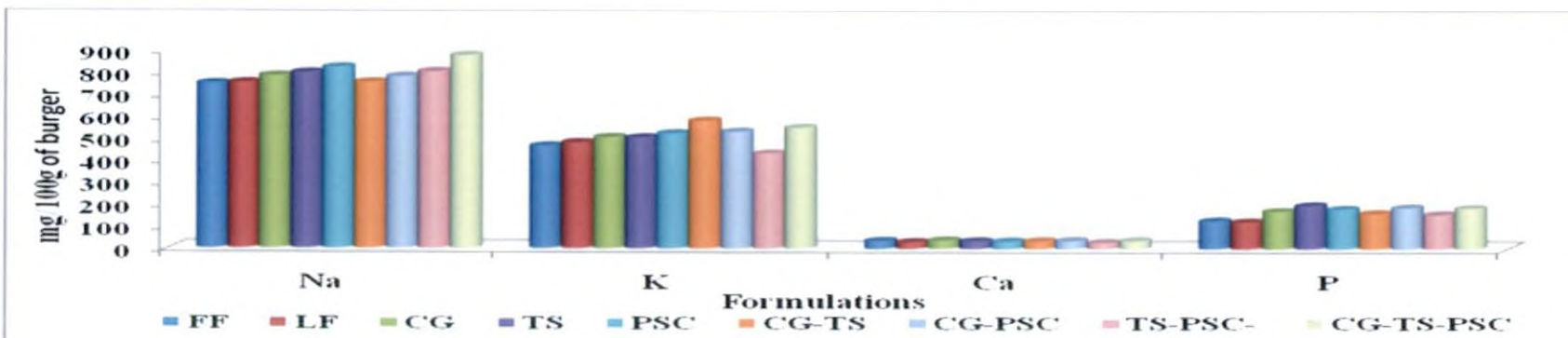


Fig.11. Mineral composition of different formulations of uncooked low fat beef burger

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

Table7. Proximate and mineral composition of different formulations of cooked low fat beef burger

Formulations	Parameters, %								
	Moisture	Protein	Fat	Carbohy drate	Ash	Na (mg)	K (mg)	Ca (mg)	P (mg)
FF	58.12 ^a ± 0.04	15.67 ^a ± 0.89	19.83 ^c ± 0.49	4.04 ^a ± 0.04	1.70 ^a ± 0.08	811 ^{bc} ± 3.62	482.7 ^a ± 4.28	37.83 ^b ± 0.94	143.3 ^b ± 1.15
LF	66.52 ^b ± 0.01	21.56 ^c ± 0.17	5.87 ^{ab} ± 0.19	4.54 ^c ± 0.03	1.74 ^a ± 0.03	773.6 ^a ± 6.17	497.4 ^b ± 1.60	34.68 ^b ± 0.75	134 ^a ± 2.56
CG	68.14 ^c ± 0.36	18.18 ^b ± 0.17	5.47 ^a ± 0.25	6.35 ^b ± 0.08	1.83 ^{ab} ± 0.03	808 ^{bc} ± 2.67	527.3 ^{bc} ± 0.22	42.21 ^d ± 0.73	142.5 ^d ± 2.78
TS	66.81 ^b ± 0.55	19.01 ^{ab} ± 0.86	6.26 ^{ab} ± 0.18	6.03 ^f ± 0.04	1.76 ^a ± 0.04	815 ^c ± 9.31	515.8 ^b ± 1.86	38.45 ^b ± 0.70	209.2 ^{ef} ± 2.97
PSC	66.34 ^b ± 0.30	21.59 ^c ± 0.34	6.09 ^{ab} ± 0.06	4.24 ^b ± 0.04	1.80 ^a ± 0.02	848.5 ^d ± 8.28	560.3 ^{cd} ± 7.80	37.97 ^b ± 0.35	205.2 ^c ± 1.93
CG-TS	66.45 ^b ± 0.39	20.22 ^{cd} ± 0.33	5.45 ^a ± 0.06	6.10 ^f ± 0.01	1.86 ^{ab} ± 0.08	793.2 ^b ± 4.65	618.8 ^c ± 2.70	36.70 ^{bc} ± 0.36	193.7 ^d ± 2.49
CG-PSC	66.71 ^b ± 0.53	20.61 ^{cd} ± 0.54	5.80 ^{ab} ± 0.05	4.93 ^d ± 0.12	1.99 ^{bc} ± 0.04	822.8 ^c ± 3.83	586.1 ^{de} ± 0.72	38.32 ^b ± 0.69	202.7 ^c ± 2.28
TS-PSC	67.07 ^{ab} ± 0.57	21.38 ^c ± 0.48	5.93 ^{ab} ± 0.15	3.99 ^a ± 0.01	2.12 ^c ± 0.08	850 ^d ± 5.89	481.1 ^a ± 0.67	31.93 ^a ± 1.38	182.2 ^c ± 6.44
CG-TS-PSC	66.72 ^b ± 0.53	20.00 ^{cd} ± 0.81	5.86 ^{ab} ± 0.28	5.36 ^e ± 0.03	2.14 ^c ± 0.09	946.3 ^e ± 9.83	572.1 ^e ± 3.54	38.49 ^b ± 1.33	216.5 ^b ± 1.55

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

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

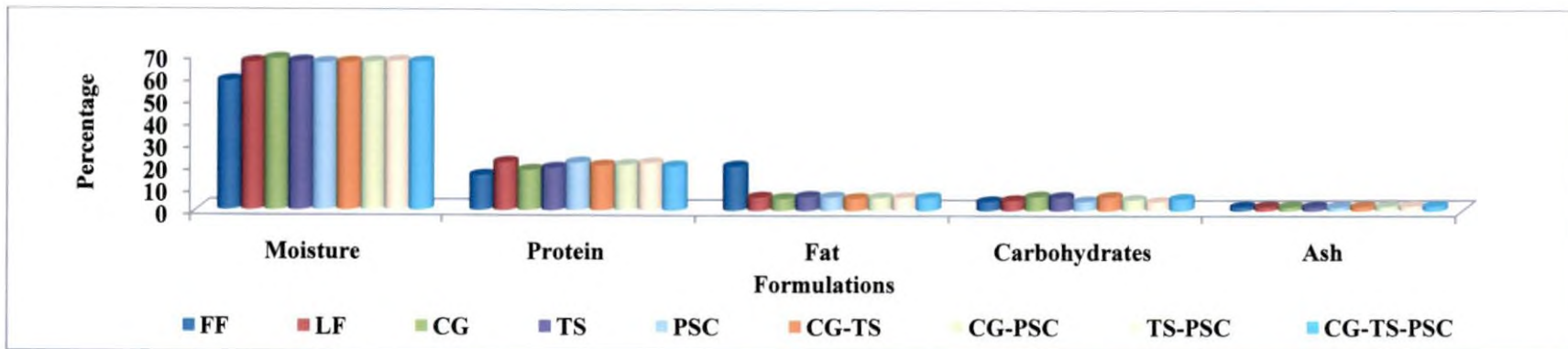


Fig. 12. Proximate composition of different formulations of cooked low fat beef burger

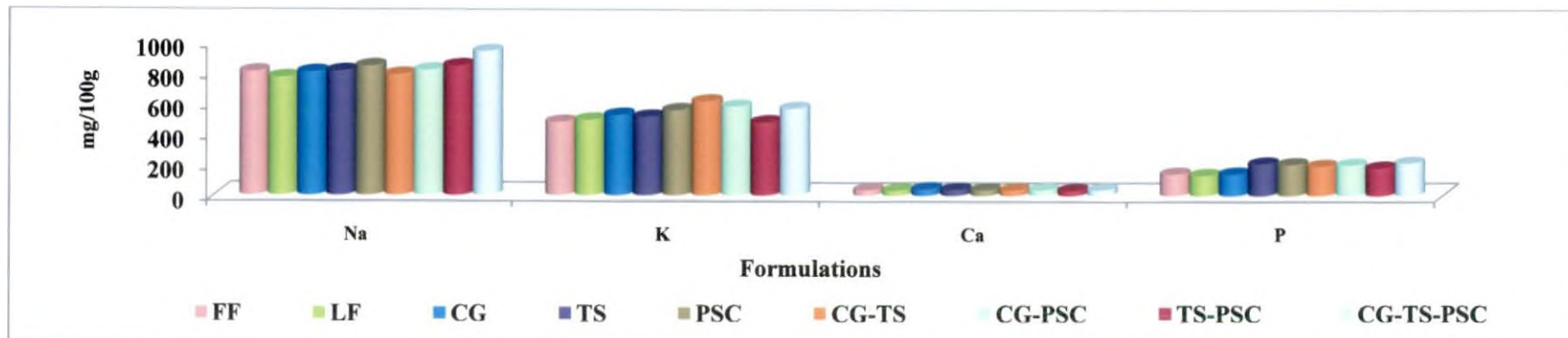


Fig.13. Mineral composition of different formulations of cooked low fat beef burger

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

4.2.2.3. Effect of Cooking on the Proximate and Mineral Composition of the Beef Burger

On comparison of the proximate and mineral composition of the uncooked and cooked beef burgers, an apparent reduction ($P < 0.05$) in the moisture content and a corresponding increase ($P < 0.05$) in the protein, fat, carbohydrate, ash, Na, K, Ca and P content.

4.3 NUTRITIONAL VALUE

4.3.1. Calorific Value of Nutrients and their Contribution to the Recommended Daily Allowance

Calorific value obtained from carbohydrate, protein and fat present in 100g of burgers of different formulations and their percentage contribution to the Recommended Daily Allowance (RDA) are given in the Table 8 and the trend is illustrated in Fig 14 and 15.

The total energy value in 100 g of LFBB with single FR ranged from 147.3 ± 0.23 to 158.1 ± 0.46 kCal while in combinations of FR from 154.3 ± 0.32 kCal to 154.3 ± 0.63 kCal. Both groups were significantly lower than FF control which contributed 257.3 ± 0.31 kCal. The RDA of calorific value 11.68 ± 0.04 per cent in FF could be reduced to the range of 6.36 ± 0.02 to 7.18 ± 0.01 per cent in LFBB, irrespective of single or combination of FR.

The percentage contribution to RDA of calorific value from carbohydrate was from 0.73 to 1.10. In the full fat BB, the calories from fat and their percentage contribution to RDA were 178.4 ± 0.23 kCal and 8.11 ± 0.04 per cent, which was significantly ($P < 0.05$) the highest. But in all formulations of LFBB, the contribution of fat to RDA of calorific value ranged from 2.22 ± 0.03 to 2.42 ± 0.07 . The burgers with CG-TS showed significantly ($P < 0.05$) lower percentage of RDA among the combinations of FR. While the burgers with TS showed significantly ($P < 0.05$) higher percentage RDA among those with single FR.

Table 8. Calorific value of nutrients and their per cent contribution to the RDA in different formulations of low fat beef burger

Formulations		Carbohydrate	Protein	Fat	Total
FF	kacl/100g	16.16 ^{ab} ± 0.73	62.68 ^a ± 0.44	178.4 ^c ± 0.23	257.3 ^d ± 0.31
	% of RDA	0.73 ^a ± 0.02	2.84 ^a ± 0.07	8.11 ^d ± 0.04	11.68 ^c ± 0.04
LF	kacl/100g	18.16 ^c ± 0.02	86.24 ^d ± 0.40	52.83 ^b ± 0.14	157.2 ^c ± 0.37
	% of RDA	0.82 ^a ± 0.04	3.92 ^c ± 0.01	2.40 ^b ± 0.06	7.14 ^{ab} ± 0.06
CG	kacl/100g	25.40 ^e ± 0.84	72.72 ^b ± 0.18	49.23 ^a ± 0.44	147.3 ^a ± 0.23
	% of RDA	1.15 ^d ± 0.03	3.30 ^b ± 0.04	2.23 ^a ± 0.01	6.36 ^a ± 0.02
TS	kacl/100g	24.12 ^c ± 0.24	76.04 ^c ± 0.25	56.34 ^d ± 0.51	156.5 ^c ± 0.32
	% of RDA	1.09 ^d ± 0.04	3.45 ^b ± 0.03	2.56 ^c ± 0.05	7.10 ^{ab} ± 0.06
PSC	kacl/100g	16.96 ^{ab} ± 0.47	86.36 ^d ± 0.45	54.8 ^d ± 0.19	158.1 ^c ± 0.46
	% of RDA	0.77 ^a ± 0.01	3.92 ^c ± 0.05	2.49 ^{bc} ± 0.03	7.18 ^{ab} ± 0.01
CG-TS	kacl/100g	24.4 ^c ± 0.15	80.88 ^c ± 0.26	49.05 ^a ± 0.45	154.3 ^b ± 0.32
	% of RDA	1.10 ^d ± 0.04	3.67 ^{bc} ± 0.03	2.22 ^a ± 0.03	6.99 ^{ab} ± 0.04
CG-PSC	Kacl/100g	19.72 ^c ± 0.53	82.44 ^c ± 0.47	52.2 ^b ± 0.30	154.3 ^b ± 0.63
	% of RDA	0.89 ^b ± 0.02	3.74 ^{bc} ± 0.05	2.37 ^b ± 0.04	7.00 ^{ab} ± 0.01
TS-PSC	kacl/100g	15.96 ^a ± 0.32	85.52 ^d ± 0.17	53.37 ^{bc} ± 0.17	154.3 ^b ± 0.32
	% of RDA	0.72 ^a ± 0.01	3.88 ^c ± 0.03	2.42 ^b ± 0.07	7.02 ^{ab} ± 0.07
CG-TS-PSC	kacl/100g	21.44 ^{cd} ± 0.35	80.00 ^c ± 0.26	52.74 ^{bc} ± 0.46	154.1 ^b ± 0.52
	% of RDA	0.97 ^c ± 0.02	3.63 ^{bc} ± 0.06	2.40 ^b ± 0.01	7.02 ^b ± 0.06

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG
& 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0%PSC

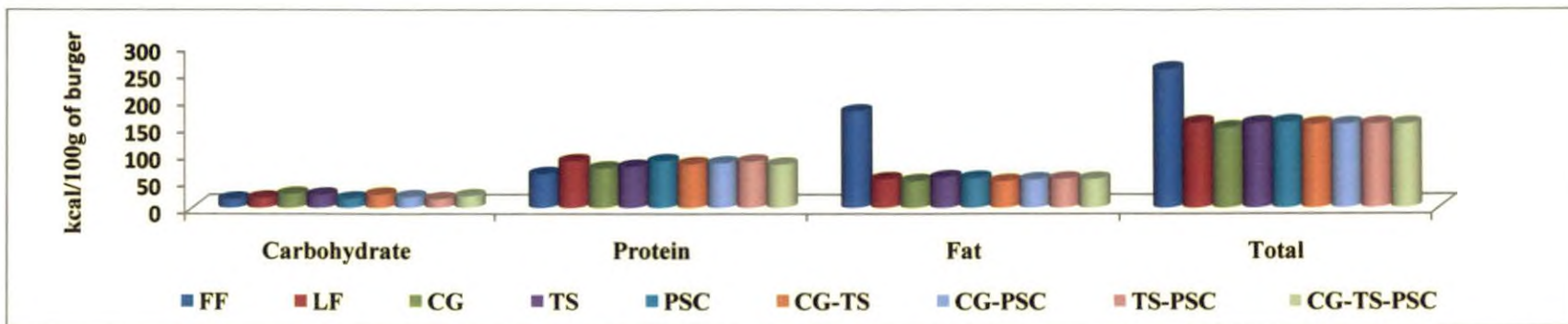


Fig. 14. Calorific value of nutrients in different formulations of low fat beef burger

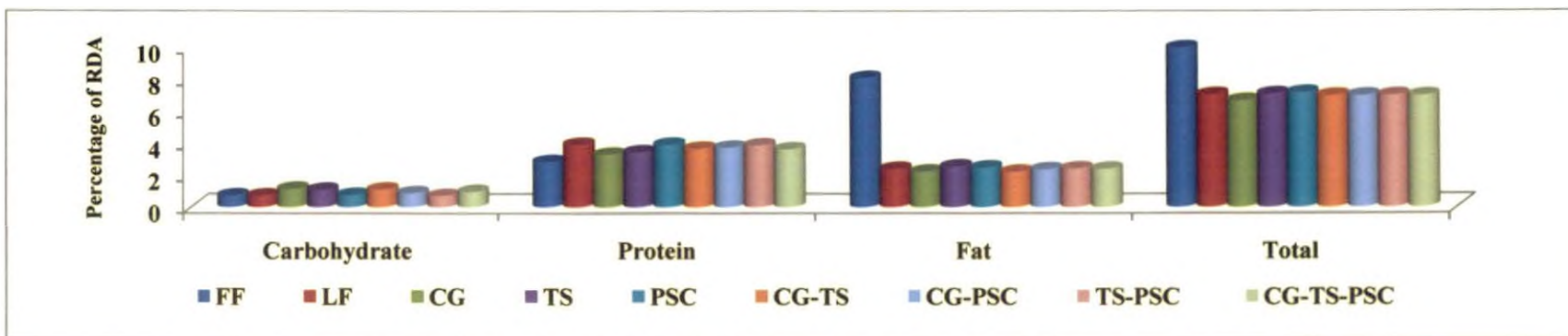


Fig.15. Contribution of nutrients to the RDA of calorific value of low fat beef burger

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen;

CG - 0.5% CG; TS - 1.5% TS; PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;

TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

Protein in burgers with PSC and LF showed significantly ($P < 0.05$) higher calorific value among all formulations. They were 86.36 ± 0.45 and 86.24 ± 0.40 , respectively and their percentage contribution of RDA was 3.92 ± 0.05 .

4.3.2. Per Cent Daily Values of Protein and Minerals

The percentage RDA of protein and minerals in the different formulations of LFBB is presented in Table 9 and illustrated in Fig. 16.

Table 9. Per cent RDA of protein and mineral of different formulations of low fat beef burger

Nutrients	Protein	Na	K	Ca	P
FF	26.10 ^a ± 0.75	33.79 ^{ab} ± 0.56	13.79 ^a ± 0.24	4.72 ^b ± 0.04	17.9 ^b ± 0.32
LF	35.93 ^d ± 0.62	32.23 ^a ± 0.96	14.21 ^b ± 0.17	4.33 ^b ± 0.07	16.7 ^a ± 0.17
CG	30.3 ^b ± 0.17	33.66 ^{ab} ± 0.74	15.06 ^{bc} ± 0.42	5.27 ^c ± 0.03	17.8 ^b ± 0.12
TS	31.68 ^b ± 0.35	33.95 ^{ab} ± 1.02	14.73 ^b ± 0.31	4.80 ^b ± 0.07	26.1 ^c ± 0.31
PSC	35.98 ^d ± 0.10	35.35 ^c ± 0.64	16.00 ^c ± 0.42	4.74 ^b ± 0.03	25.6 ^c ± 0.24
CG-TS	33.70 ^c ± 0.58	34.28 ^b ± 0.71	17.68 ^d ± 0.12	4.58 ^b ± 0.04	24.2 ^c ± 0.29
CG-PSC	34.35 ^{cd} ± 0.42	34.28 ^b ± 0.78	16.74 ^c ± 0.13	4.79 ^b ± 0.06	25.3 ^c ± 0.11
TS-PSC	35.63 ^d ± 0.21	35.41 ^c ± 0.51	13.74 ^a ± 0.31	3.99 ^a ± 0.01	22.7 ^{bc} ± 0.41
CG-TS-PSC	33.33 ^c ± 0.42	39.42 ^d ± 0.74	16.34 ^c ± 0.25	4.81 ^{ab} ± 0.04	27.0 ^d ± 0.34

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0%PSC

The LFBB with TS-PSC showed significantly ($P < 0.05$) higher percentage of 35.93 ± 0.62 daily value of protein among combinations of FR. While in burgers with PSC alone was 35.98 ± 0.62 and significantly ($P < 0.05$) the highest among the single FR.

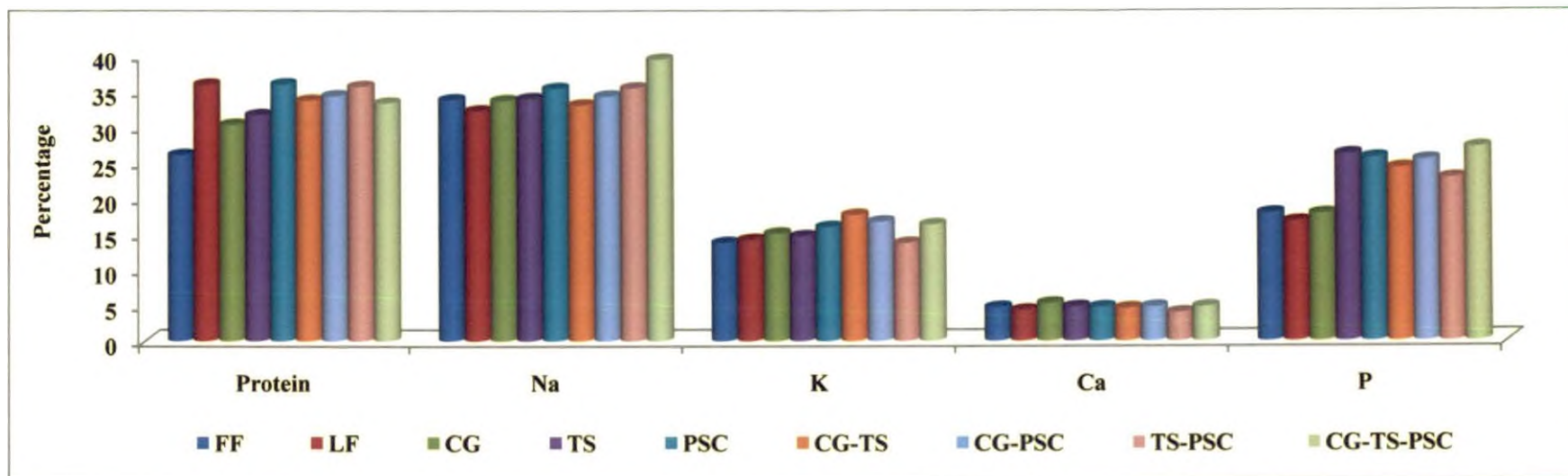


Fig.16. Per cent daily values of protein and minerals in low fat beef burger

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS;
PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;
CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; Na- Sodium; K- Potassium; Ca- Calcium; P- Phosphorous.

The burger with CG-TS-PSC contained significantly ($P < 0.05$) the highest Na and P among all formulations, which were 39.42 ± 0.74 and 27.0 ± 0.34 , respectively. K content in LFBB ranged from 13.74 ± 0.31 to 17.68 ± 0.12 . The Ca content of burger with CG showed significantly ($P < 0.05$) the highest compared to remaining formulations which were recorded as 5.27 ± 0.03 .

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

At $0-4^{\circ}\text{C}$, under AP and VP the BB could be stored up to d 10 only without spoilage. Similarly, at -20°C all the formulations of BB could be stored up to d 30 in both AP and VP. But FF control could be stored only up to d 20 in AP without spoilage.

4.4.1. Purge Loss

The effect of aerobic and vacuum packaging and the period of storage on PL of different formulations of burger on day 10 at $0-4^{\circ}\text{C}$ and d 10, 20 and 30 at -20°C , are presented in Table 10, 11 and illustrated in Fig.17 and 18, respectively.

The purge loss in LF control burgers packaged under aerobic and vacuum and stored at $0-4^{\circ}\text{C}$, was 1.04 ± 0.02 and 0.997 ± 0.02 per cent, respectively. But at -20°C it was 0.985 ± 0.01 and 0.940 ± 0.01 , respectively. This was significantly ($P < 0.05$) the highest among all formulations. BB with combinations of FR showed significantly ($P < 0.05$) lowest PL compared to single fat replacers in both types of packaging and storage. The PL of FF control in both types of packaging at $0-4^{\circ}\text{C}$ was same as that of BB with single FR. But at -20°C , the PL of FF control was same as that of burgers with combinations of FR.

The BB with PSC on d 10 in AP showed significantly ($P < 0.05$) higher PL compared to VP. Otherwise, packaging did not have any significant effect in PL under both types of storage.

Table 10. Effect of packaging and period of storage on purge loss of low fat beef burger stored at 0-4°C (%)

Formulations	Aerobic packaging	Vacuum packaging
	Period of storage, d	Period of storage, d
	10	10
FF	0.622 ^{bB} ±0.07	0.620 ^{bB} ±0.07
LF	1.04 ^{cB} ±0.02	0.997 ^{cB} ±0.02
CG	0.685 ^{bC} ±0.01	0.687 ^{bC} ±0.01
TS	0.622 ^{bC} ±0.02	0.622 ^{bC} ±0.02
PSC	0.650 ^{bB} ±0.01	0.617 ^{bA} ±0.01
CG-TS	0.417 ^{aCD} ±0.02	0.432 ^{aD} ±0.02
CG-PSC	0.467 ^{aD} ±0.04	0.477 ^{aD} ±0.04
TS-PSC	0.482 ^{aC} ±0.02	0.460 ^{aC} ±0.02
CG-TS-PSC	0.407 ^{aC} ±0.02	0.392 ^{aC} ±0.02

Table 11. Effect of packaging and period of storage on purge loss of low fat beef burger stored at -20°C (%)

Formulations	Aerobic packaging			Vacuum packaging		
	Period of storage, d			Period of storage, d		
	10	20	30	10	20	30
FF	0.557 ^{cB} ± 0.06	0.577 ^{cB} ± 0.06	S	0.535 ^{bAB} ± 0.06	0.485 ^{bA} ± 0.06	0.405 ^{cA} ± 0.04
LF	0.985 ^{cB} ± 0.01	1.18 ^{fC} ± 0.02	1.33 ^{dD} ± 0.03	0.940 ^{eA} ± 0.01	1.08 ^{eB} ± 0.01	1.23 ^{eC} ± 0.05
CG	0.682 ^{dC} ± 0.01	0.622 ^{deBC} ± 0.01	0.572 ^{bB} ± 0.02	0.667 ^{dC} ± 0.01	0.590 ^{cB} ± 0.00	0.527 ^{bA} ± 0.02
TS	0.625 ^{dC} ± 0.01	0.590 ^{dC} ± 0.03	0.502 ^{cB} ± 0.02	0.605 ^{dC} ± 0.02	0.557 ^{cB} ± 0.03	0.407 ^{bA} ± 0.02
PSC	0.665 ^{dB} ± 0.05	0.677 ^{eC} ± 0.02	0.865 ^{eD} ± 0.04	0.697 ^{eC} ± 0.03	0.737 ^{dC} ± 0.01	0.857 ^{eD} ± 0.04
CG-TS	0.397 ^{abC} ± 0.01	0.332 ^{abB} ± 0.02	0.302 ^{aB} ± 0.01	0.375 ^{aC} ± 0.01	0.325 ^{aB} ± 0.01	0.270 ^{aA} ± 0.02
CG-PSC	0.477 ^{bcD} ± 0.04	0.412 ^{bcC} ± 0.04	0.375 ^{abB} ± 0.04	0.420 ^{abC} ± 0.04	0.380 ^{abB} ± 0.03	0.340 ^{aA} ± 0.03
TS-PSC	0.497 ^{cC} ± 0.02	0.480 ^{cC} ± 0.03	0.395 ^{bB} ± 0.03	0.462 ^{bcC} ± 0.02	0.480 ^{ccC} ± 0.03	0.347 ^{bA} ± 0.05
CG-TS-PSC	0.385 ^{aC} ± 0.01	0.312 ^{aCB} ± 0.02	0.272 ^{aAB} ± 0.02	0.380 ^{aC} ± 0.01	0.395 ^{bcC} ± 0.02	0.255 ^{aA} ± 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).

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

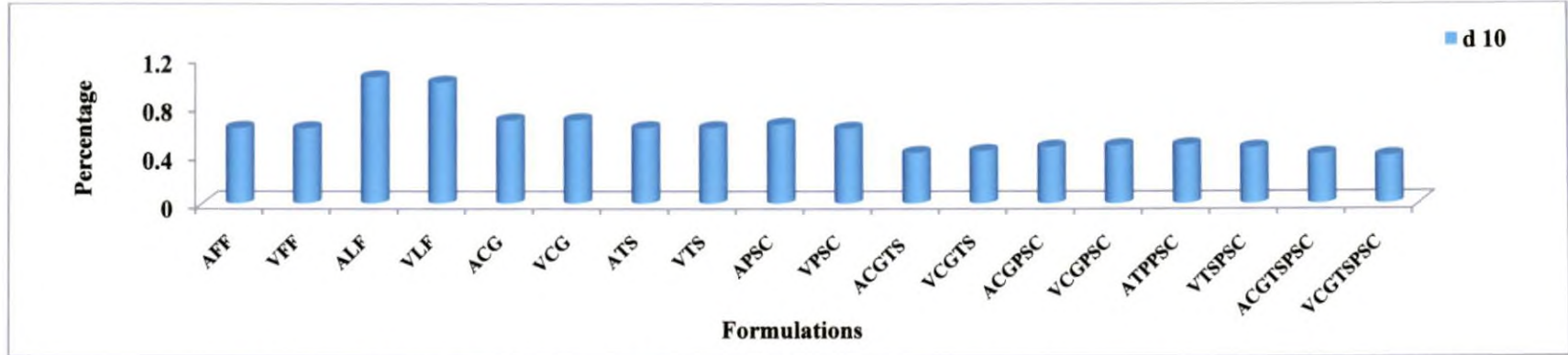


Fig.17. Effect of packaging and period of storage on purge loss of low fat beef burger stored at 0-4°C

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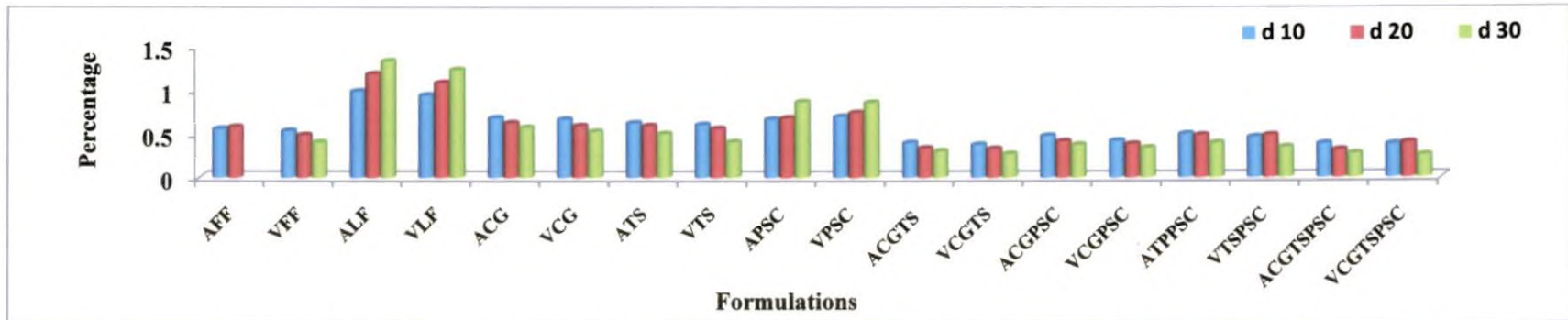


Fig. 18. Effect of packaging and period of storage on purge loss of low fat beef burger stored at -20°C

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen;

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;

TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; A- Aerobic packaging ;V- Vacuum packaging.

As the period of storage increased from day 10 to 30, the PL significantly ($P < 0.05$) decreased in all formulations except in BB with LF and PSC, in which it significantly ($P < 0.05$) increased.

4.4.2. TBARS Value

The change in TBARS value (mg malonaldehyde/kg of LFBB) of different formulations of beef burger packaged aerobically and under vacuum from day zero to d 10 at 0-4°C and on d, 10, 20 and 30 at -20°C are presented in Table 12, 13 and illustrated in Fig. 19 and 20.

In AP and VP at 0-4°C, the burgers with CG either in combination or alone showed significantly ($P < 0.05$) lower TBARS values. It ranged from 0.241 ± 0.00 to 0.274 ± 0.00 mg/kg on d 10 in AP. The remaining treatments showed significant ($P < 0.05$) increase in TBARS values as period of storage increased from 0 – 10 days, irrespective of the packaging.

All the burgers with CG stored at -20°C showed significant ($P < 0.05$) decrease in TBARS values in both AP and VP from d zero to d 30. In these, the values ranged from 0.252 ± 0.01 to 0.266 ± 0.01 mg/kg. Burgers with CG alone as FR recorded the lowest value. Among the combinations of FR, CG-PSC showed significantly ($P < 0.05$) the lowest value of 0.258 ± 0.01 mg/kg on d zero. In FF burgers the TBARS values significantly ($P < 0.05$) increased from d zero to d 20 in AP and the burgers spoiled on d 30. While in VP, although there was significant ($P < 0.05$) increase in the TBARS values, it could be stored up to 30 days without spoilage. The TBARS values of LFBB with CG in AP and VP at 0-4°C and at -20°C significantly ($P < 0.05$) decreased from d zero to d 30.

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

The Taste Panel scores of organoleptic qualities of different formulations of LFBB on the day of preparation, *i.e.*, day zero are given in Table 14 and illustrated in Fig.21.

Table 12. Effect of packaging and period of storage on TBARS of low fat beef burger stored at 0-4°C (mg malonaldehyde/ kg of burger)

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	0.319 ^{dA} ±0.03	0.350 ^{cB} ±0.01	0.319 ^{dA} ±0.03	0.340 ^{dB} ±0.01
LF	0.245 ^{aA} ±0.01	0.270 ^{bcBC} ±0.01	0.245 ^{aA} ±0.01	0.258 ^{bB} ±0.01
CG	0.252 ^{aC} ±0.01	0.241 ^{aAB} ±0.00	0.252 ^{aC} ±0.01	0.235 ^{aAB} ±0.01
TS	0.277 ^{cA} ±0.00	0.295 ^{dB} ±0.00	0.277 ^{cA} ±0.00	0.287 ^{cB} ±0.00
PSC	0.286 ^{cA} ±0.00	0.304 ^{dB} ±0.01	0.286 ^{cA} ±0.00	0.298 ^{cAB} ±0.01
CG-TS	0.272 ^{bcE} ±0.01	0.261 ^{bcD} ±0.01	0.272 ^{bcE} ±0.01	0.256 ^{bcC} ±0.01
CG-PSC	0.258 ^{abC} ±0.01	0.253 ^{abC} ±0.01	0.258 ^{abC} ±0.01	0.240 ^{abB} ±0.00
TS-PSC	0.276 ^{cA} ±0.00	0.292 ^{dc} ±0.01	0.276 ^{cA} ±0.00	0.292 ^{cc} ±0.00
CG-TS-PSC	0.278 ^{cB} ±0.00	0.274 ^{cB} ±0.00	0.278 ^{cB} ±0.00	0.264 ^{bA} ±0.00

Table 13. Effect of packaging and period of storage on TBARS of low fat beef burger stored at -20°C (mg malonaldehyde/ kg of burger)

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	0.319 ^{dA} ±0.03	0.344 ^{FB} ±0.01	0.347 ^{cB} ±0.01	S	0.319 ^{aD} ±0.03	0.362 ^{FB} ±0.01	0.381 ^{cC} ±0.01	0.390 ^{FC} ±0.01
LF	0.255 ^{aA} ±0.01	0.268 ^{cdB} ±0.01	0.290 ^{cC} ±0.01	0.305 ^{cC} ±0.01	0.245 ^{aA} ±0.01	0.262 ^{bc} ±0.01	0.304 ^{dc} ±0.01	0.320 ^{dc} ±0.01
CG	0.252 ^{aC} ±0.01	0.240 ^{aAB} ±0.01	0.229 ^{aA} ±0.01	0.219 ^{aA} ±0.01	0.252 ^{aC} ±0.01	0.236 ^{aAB} ±0.01	0.228 ^{aA} ±0.01	0.222 ^{aA} ±0.01
TS	0.277 ^{cA} ±0.00	0.286 ^{deAB} ±0.00	0.304 ^{cdBC} ±0.00	0.320 ^{cc} ±0.00	0.277 ^{cA} ±0.00	0.282 ^{deAB} ±0.00	0.301 ^{dBBC} ±0.00	0.311 ^{dc} ±0.00
PSC	0.286 ^{cA} ±0.00	0.299 ^{eb} ±0.00	0.319 ^{dBBC} ±0.00	0.334 ^{dc} ±0.01	0.286 ^{cA} ±0.00	0.294 ^{eAB} ±0.00	0.316 ^{dBBC} ±0.01	0.330 ^{dc} ±0.01
CG-TS	0.272 ^{bcE} ±0.01	0.258 ^{bcC} ±0.00	0.245 ^{ab} ±0.01	0.238 ^{aA} ±0.00	0.272 ^{bcE} ±0.01	0.251 ^{abcC} ±0.01	0.250 ^{bcC} ±0.01	0.255 ^{cc} ±0.01
CG-PSC	0.258 ^{abC} ±0.01	0.241 ^{abB} ±0.01	0.242 ^{ab} ±0.01	0.228 ^{aA} ±0.01	0.258 ^{abC} ±0.01	0.245 ^{abB} ±0.01	0.242 ^{abB} ±0.01	0.229 ^{abA} ±0.00
TS-PSC	0.276 ^{cA} ±0.00	0.284 ^{deB} ±0.00	0.299 ^{cC} ±0.01	0.315 ^{cdC} ±0.01	0.276 ^{cA} ±0.00	0.283 ^{deB} ±0.00	0.300 ^{dc} ±0.01	0.311 ^{dc} ±0.00
CG-TS-PSC	0.278 ^{cB} ±0.00	0.270 ^{cdB} ±0.00	0.268 ^{ba} ±0.00	0.266 ^{ba} ±0.01	0.278 ^{cB} ±0.00	0.265 ^{cdA} ±0.00	0.264 ^{cA} ±0.00	0.264 ^{bcA} ±0.01

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).

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

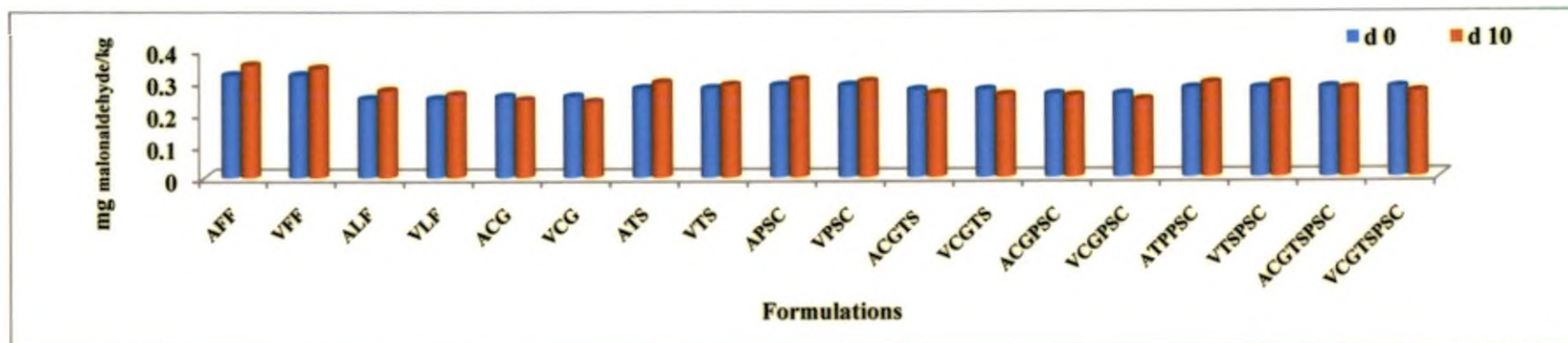


Fig.19. Effect of packaging and period of storage on TBARS of low fat beef burger stored at 0-4°C

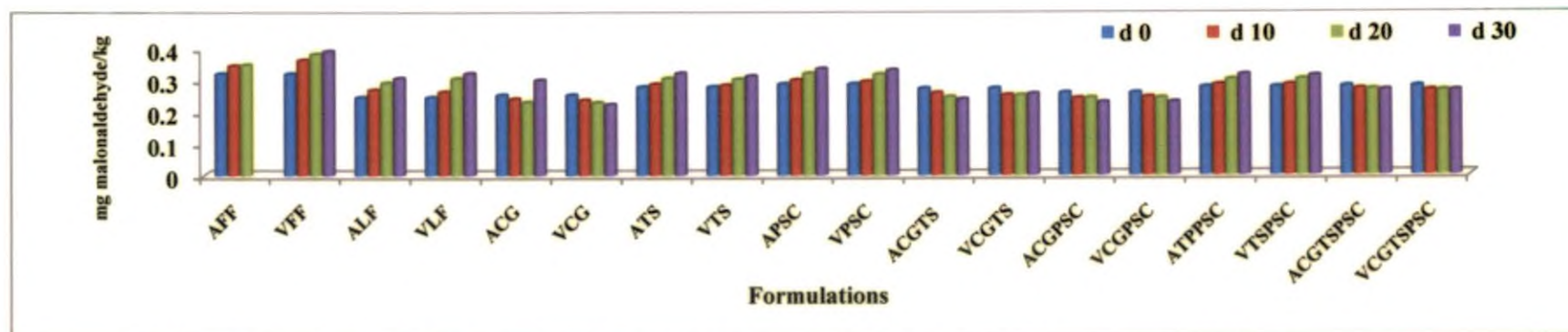


Fig.20. Effect of packaging and period of storage on TBARS of low fat beef burger stored at -20°C

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen;
 CG - 0.5% CG; TS - 1.5% TS;PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS;CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC;CG-TS-PSC - 0.5% CG,1.5% TS & 2.0% PSC; A- Aerobic packaging ;V- Vacuum packaging

Table 14. Sensory evaluation score of different formulations of low fat beef burger on the day of preparation

Attributes	Formulations								
	FF	LF	CG	TS	PSC	CG-TS	CG-PSC	TS-PSC	CG-TS-PSC
Appearance and colour	7.12 ^c ±0.08	6.37 ^b ±0.15	6.50 ^{bc} ±0.18	5.87 ^a ±0.27	7.00 ^d ±0.00	6.87 ^d ±0.08	6.62 ^{bcd} ±0.15	6.5 ^{bc} ±0.13	7.00 ^d ±0.00
Flavour	7.00 ^d ±0.00	6.00 ^a ±0.00	6.43 ^b ±0.04	6.37 ^b ±0.15	6.87 ^{cd} ±0.08	7.25 ^e ±0.09	6.75 ^{cd} ±0.09	6.62 ^{bc} ±0.08	7.00 ^d ±0.00
Texture	7.25 ^a ±0.09	6.12 ^a ±0.08	6.62 ^{bc} ±0.08	6.50 ^b ±0.09	6.68 ^{bc} ±0.13	6.93 ^{cd} ±0.11	6.75 ^{bc} ±0.09	6.87 ^c ±0.15	7.25 ^d ±0.13
Saltiness	6.87 ^c ±0.15	6.56 ^{ab} ±0.17	6.37 ^a ±0.08	6.37 ^a ±0.08	6.75 ^{bc} ±0.09	6.93 ^{cd} ±0.06	6.43 ^a ±0.11	6.31 ^a ±0.13	7.00 ^c ±0.00
Juiciness	7.18 ^f ±0.09	6.00 ^a ±0.00	6.43 ^b ±0.06	6.50 ^{bc} ±0.00	6.75 ^{cd} ±0.13	7.00 ^{de} ±0.09	6.62 ^{bc} ±0.12	6.56 ^{bc} ±0.14	7.06 ^e ±0.06
Mouth coating	7.25 ^f ±0.16	6.50 ^{abc} ±0.13	6.62 ^{bcd} ±0.15	6.12 ^a ±0.08	7.00 ^{de} ±0.00	6.87 ^{cd} ±0.08	6.50 ^{cd} ±0.00	6.62 ^{bcd} ±0.08	7.00 ^e ±0.00
Overall acceptability	7.25 ^f ±0.09	6.00 ^a ±0.00	6.50 ^c ±0.13	6.25 ^b ±0.09	6.87 ^{de} ±0.08	7.00 ^e ±0.00	6.75 ^{cde} ±0.09	6.62 ^{cd} ±0.08	7.25 ^f ±0.09

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;

TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

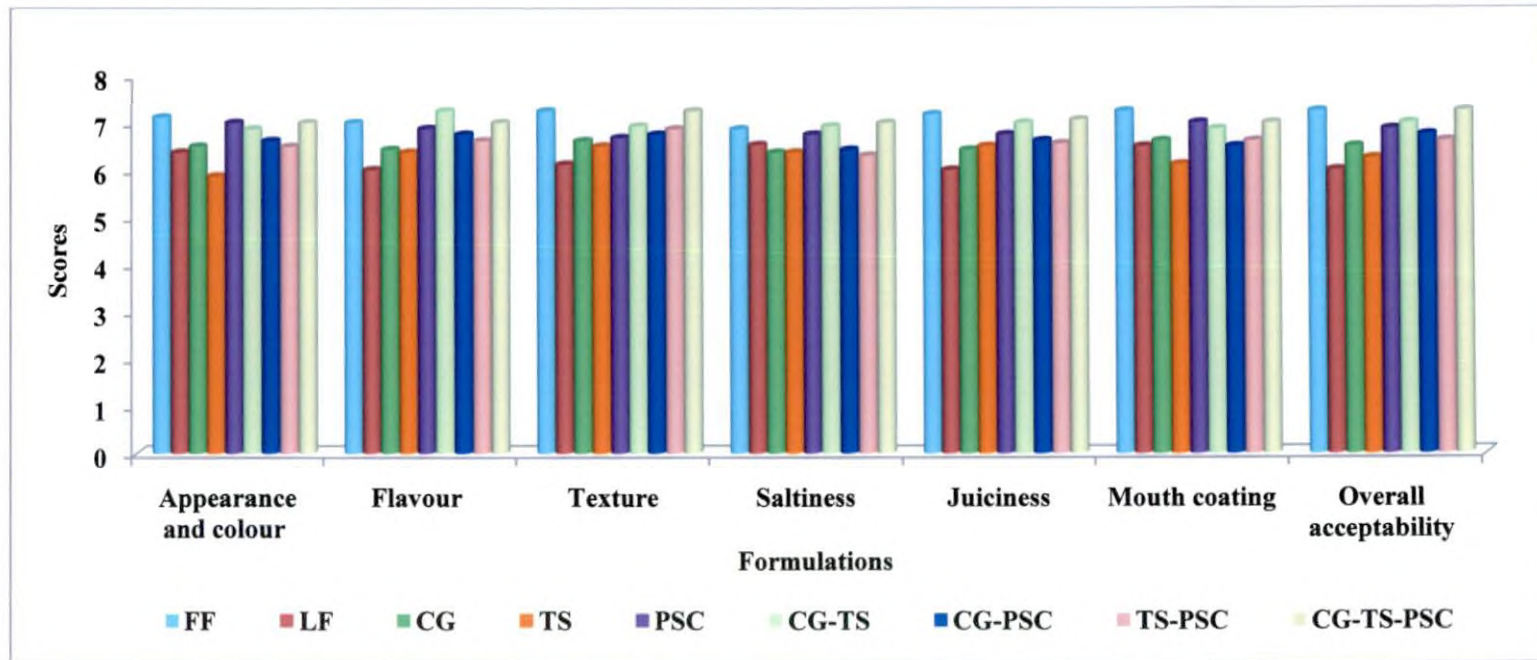


Fig.21. Sensory evaluation scores of different formulations of low fat beef burger on the day of preparation

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC.

On day zero, the appearance and colour of burgers with FF was 7.12 ± 0.08 . The burgers with CG-TS-PSC among combinations of FR and those with PSC with single FR scored 7.00 ± 0.00 . This was significantly ($P < 0.05$) higher than the other formulations.

Flavour scores were significantly ($P < 0.05$) highest for CG-TS with a score of 7.25 ± 0.09 followed by CG-TS-PSC and FF burgers. Both scored 7.00.

The burgers with CG-TS-PSC scored significantly ($P < 0.05$) higher values of 7.00 and above, for desirable texture and saltiness, very juicy, no mouth coating and very acceptable overall acceptability. The same trend was shown by FF, except for saltiness which was only moderately desirable with a score of 6.87 ± 0.15 . The overall acceptability of both was 7.25 ± 0.09 . The LFBB with CG-TS scored 7.00 for overall acceptability. This was significantly ($P < 0.05$) higher than other formulations except PSC and CG-PSC. Among single FR, the burgers with PSC showed significantly ($P < 0.05$) higher overall acceptability with a score of 6.87 ± 0.08 .

Sensory evaluation of the burgers at $0-4^{\circ}\text{C}$ could be stored without spoilage up to d 10 only in both AP and VP and therefore, observations were not recorded on day 20 and 30. Among the beef burger packaged aerobically and stored at -20°C , those with FF spoiled on d 30 and therefore, observations were not recorded on d 30.

4.4.3.1. Appearance and Colour

The appearance and colour of different formulations of beef burger under AP and VP from d zero to d 10 at $0-4^{\circ}\text{C}$ and on d, 10, 20 and 30 at -20°C are presented in Table 15 and 16 and illustrated in Fig. 22 and 23.

There was no significant ($P > 0.05$) difference in the appearance and colour of the burgers in any of the formulations on d 10 at $0-4^{\circ}\text{C}$ in AP and VP.

Table 15. Effect of packaging and period of storage on the appearance and colour of low fat beef burger stored at 0-4°C

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	7.12±0.08	7.00±0.00	7.12±0.08	7.12±0.08
LF	6.37±0.15	6.12±0.08	6.37±0.15	6.00±0.00
CG	6.50±0.18	6.50±0.18	6.50±0.18	6.62±0.08
TS	5.87±0.27	6.00±0.23	5.87±0.27	6.12±0.08
PSC	7.00±0.00	6.87±0.08	7.00±0.00	6.75±0.16
CG-TS	6.87±0.08	6.62±0.15	6.87±0.08	6.62±0.08
CG-PSC	6.62±0.15	6.50±0.18	6.62±0.15	6.87±0.08
TS-PSC	6.50±0.13	6.25±0.16	6.50±0.13	6.50±0.13
CG-TS-PSC	7.00±0.00	6.75±0.16	7.00±0.00	7.00±0.00

Table 16. Effect of packaging and period of storage on the appearance and colour of low fat beef burger stored at -20°C

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	7.12 ± 0.08	7.12 ± 0.08	7.00 ± 0.00	S	7.12 ± 0.08	7.00 ± 0.00	7.12 ± 0.08	6.62 ± 0.15
LF	6.37 ± 0.15	6.37 ± 0.15	6.12 ± 0.08	6.26 ± 0.13	6.37 ± 0.15	6.00 ± 0.00	6.06 ± 0.04	6.00 ± 0.13
CG	6.50 ± 0.18	6.87 ± 0.08	6.75 ± 0.09	6.50 ± 0.13	6.50 ± 0.18	6.62 ± 0.08	6.75 ± 0.09	6.12 ± 0.08
TS	5.87 ^A ± 0.27	6.25 ^C ± 0.21	6.12 ^B ± 0.20	5.62 ^D ± 0.15	5.87 ± 0.27	6.12 ± 0.08	6.12 ± 0.08	6.12 ± 0.08
PSC	7.00 ± 0.00	6.87 ± 0.08	6.62 ± 0.15	6.5 ± 0.13	7.00 ± 0.00	7.00 ± 0.00	6.87 ± 0.08	6.62 ± 0.08
CG-TS	6.87 ± 0.08	6.62 ± 0.15	6.45 ± 0.15	6.35 ± 0.25	6.87 ± 0.08	6.75 ± 0.08	6.75 ± 0.09	6.12 ± 0.08
CG-PSC	6.62 ± 0.15	6.87 ± 0.08	7.00 ± 0.13	6.75 ± 0.16	6.62 ± 0.15	7.00 ± 0.23	6.87 ± 0.08	6.75 ± 0.09
TS-PSC	6.50 ± 0.13	6.50 ± 0.13	6.50 ± 0.13	6.37 ± 0.15	6.50 ± 0.13	6.25 ± 0.09	6.25 ± 0.06	6.06 ± 0.04
CG-TS-PSC	7.00 ± 0.00	7.00 ± 0.00	6.87 ± 0.08	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00

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

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

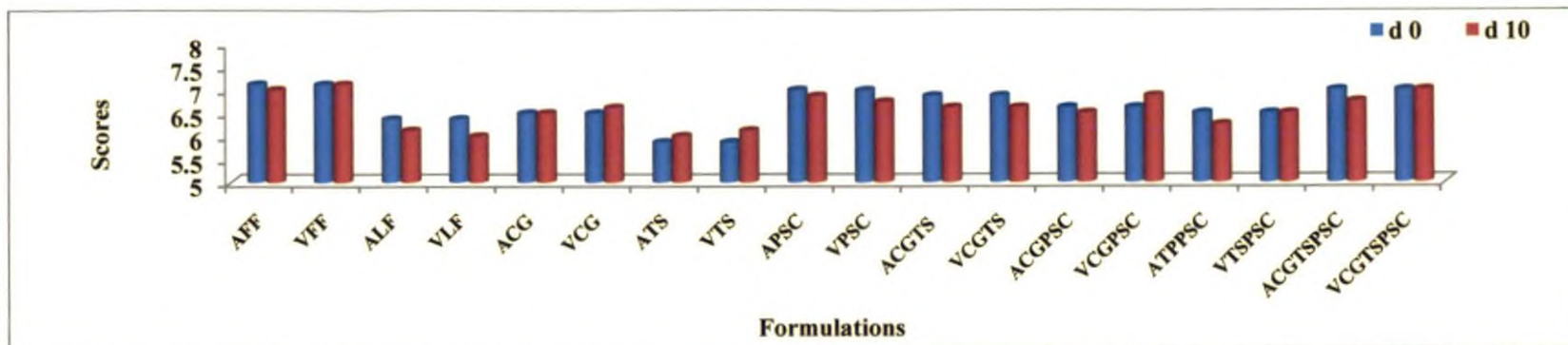


Fig.22. Effect of packaging and period of storage on appearance and colour of low fat beef burger stored at 0-4°C

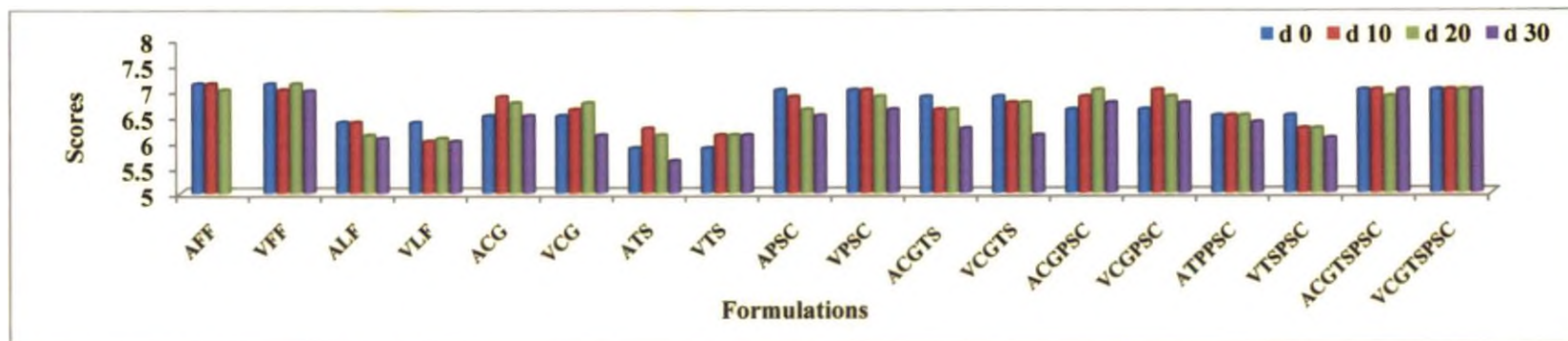


Fig.23. Effect of packaging and period of storage on appearance and colour of low fat beef burger stored at -20°C

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen;
 CG - 0.5% CG; TS - 1.5% TS; PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG& 2.0% PSC;
 TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG,1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

The LFBB with TS under AP and stored at -20°C showed significant ($P < 0.05$) decrease in appearance and colour score from 5.87 ± 0.27 to 5.62 ± 0.15 on d zero to d 30, respectively. All other formulations were not significantly different ($P > 0.05$) irrespective of the period of storage and type of packaging.

4.4.3.2. Flavour

The flavour scores of different formulations of beef burger packaged aerobically and under vacuum from d zero to d 10 at $0-4^{\circ}\text{C}$ and on d, 10, 20 and 30 at -20°C are presented in Table 17, 18 and illustrated in Fig. 24 and 25.

AP and VP did not significantly ($P > 0.05$) affect the flavour score of the BB stored at $0-4^{\circ}\text{C}$ irrespective of the days of storage from d zero to d 10.

The burger with CG-TS, CG-PSC and TS-PSC, among combinations of FR, stored at -20°C and packaged under AP and VP showed significant ($P < 0.05$) decrease in flavour score on d 30. The other treatments did not show any significant ($P > 0.05$) difference on storage up to 30 days.

4.4.3.3. Texture

The effect of packaging and period of storage on the texture scores of LFBB stored at $0-4^{\circ}\text{C}$ up to d 10 and -20°C for d 10, 20 and 30 are presented in Table 19 and 20 and trend are illustrated in Fig. 26 and 27, respectively.

There was no significant ($P > 0.05$) difference in the texture among all formulations of BB stored at $0-4^{\circ}\text{C}$ under AP and VP on d10.

The burger with CG-TS and CG stored at -20°C under AP and VP showed significant ($P < 0.05$) decrease in texture score on d 30. All other treatments except LF did not show any significant ($P > 0.05$) difference in AP and VP on storage from d zero to d 30. LF burger in VP recorded significant ($P > 0.05$) decrease in the texture score on d 30.

Table 17. Effect of packaging and period of storage on the flavour of low fat beef burger stored at 0-4°C

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	6.87 ± 0.08
LF	6.00 ± 0.00	6.00 ± 0.00	6.00 ± 0.00	5.87 ± 0.08
CG	6.43 ± 0.04	6.25 ± 0.09	6.43 ± 0.04	6.37 ± 0.08
TS	6.37 ± 0.15	6.12 ± 0.08	6.37 ± 0.15	6.12 ± 0.08
PSC	6.87 ± 0.08	6.62 ± 0.08	6.87 ± 0.08	6.75 ± 0.08
CG-TS	7.25 ± 0.09	6.87 ± 0.08	7.25 ± 0.09	7.00 ± 0.00
CG-PSC	6.75 ± 0.09	6.62 ± 0.15	6.75 ± 0.09	6.62 ± 0.08
TS-PSC	6.62 ± 0.08	6.37 ± 0.08	6.62 ± 0.08	6.37 ± 0.08
CG-TS-PSC	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00

Table 18. Effect of packaging and period of storage on the flavour of low fat beef burger stored at -20°C

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	S	7.00 ± 0.00	7.00 ± 0.00	6.75 ± 0.09	6.62 ± 0.15
LF	6.00 ± 0.00	6.00 ± 0.00	5.93 ± 0.01	5.62 ± 0.15	6.00 ± 0.00	6.12 ± 0.08	5.87 ± 0.08	5.62 ± 0.15
CG	6.43 ± 0.04	6.50 ± 0.00	6.25 ± 0.09	6.12 ± 0.08	6.43 ± 0.04	6.37 ± 0.08	6.25 ± 0.09	6.06 ± 0.04
TS	6.37 ± 0.15	6.62 ± 0.08	6.00 ± 0.00	6.00 ± 0.00	6.37 ± 0.15	6.25 ± 0.09	6.25 ± 0.16	6.00 ± 0.00
PSC	6.87 ± 0.08	7.00 ± 0.00	6.62 ± 0.15	6.37 ± 0.15	6.87 ± 0.08	6.75 ± 0.09	6.62 ± 0.08	6.12 ± 0.08
CG-TS	7.25 ^D ± 0.09	7.00 ^C ± 0.00	6.87 ^B ± 0.08	6.62 ^A ± 0.08	7.25 ^D ± 0.09	7.00 ^C ± 0.00	6.68 ^B ± 0.12	6.25 ^A ± 0.09
CG-PSC	6.75 ^D ± 0.09	6.62 ^C ± 0.15	6.25 ^B ± 0.09	6.12 ^A ± 0.08	6.75 ^D ± 0.09	6.37 ^C ± 0.15	6.37 ^B ± 0.15	5.75 ^A ± 0.09
TS-PSC	6.62 ^D ± 0.08	6.62 ^C ± 0.08	6.37 ^B ± 0.08	6.00 ^A ± 0.13	6.62 ^C ± 0.08	6.56 ^{BC} ± 0.10	6.56 ^{BC} ± 0.09	6.12 ^A ± 0.08
CG-TS-PSC	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	6.87 ± 0.08	6.62 ± 0.08

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

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

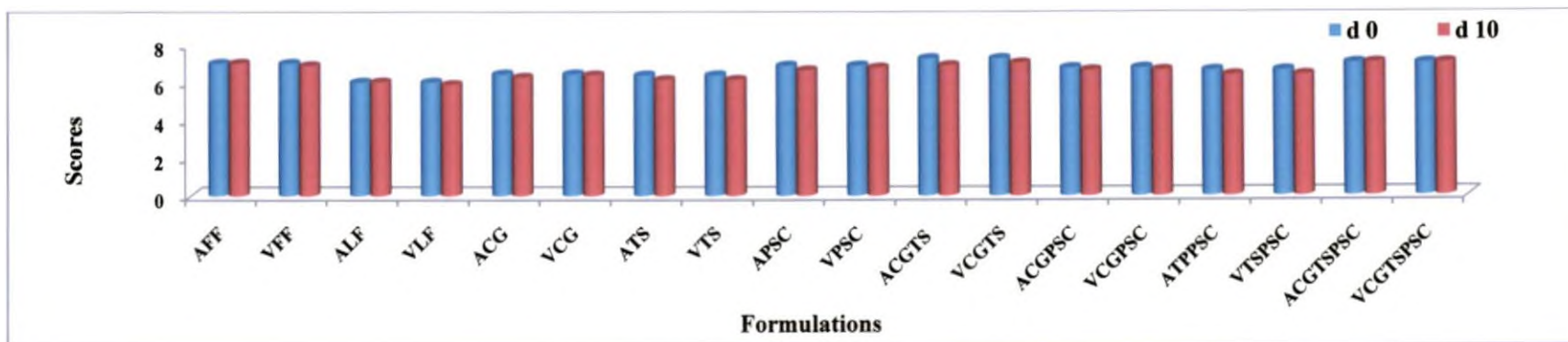


Fig.24. Effect of packaging and period of storage on the flavour of low fat beef burger stored at 0-4°C

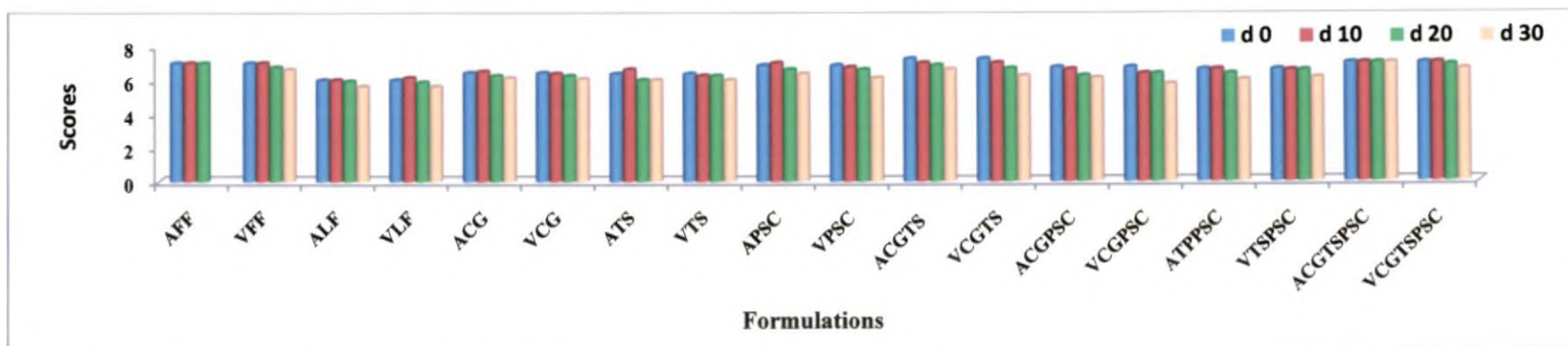


Fig.25. Effect of packaging and period of storage on the flavour of low fat beef burger stored at -20°C

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS;
 PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;
 CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

Table 19. Effect of packaging and period of storage on the texture of low fat beef burger stored at 0-4°C

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	7.25±0.09	6.87±0.12	7.25±0.09	7.12±0.08
LF	6.12±0.08	6.12±0.08	6.12±0.08	6.06±0.06
CG	6.62±0.08	6.62±0.08	6.62±0.08	6.46±0.09
TS	6.50±0.09	6.43±0.14	6.50±0.09	6.37±0.08
PSC	6.68±0.13	6.50±0.16	6.68±0.13	6.53±0.15
CG-TS	6.93±0.11	6.87±0.08	6.93±0.11	6.75±0.09
CG-PSC	6.75±0.09	6.62±0.08	6.75±0.09	6.62±0.12
TS-PSC	6.87±0.15	6.56±0.14	6.87±0.15	6.56±0.14
CG-TS-PSC	7.25±0.13	7.12±0.08	7.25±0.13	7.06±0.06

Table 20. Effect of packaging and period of storage on the texture of low fat beef burger stored at -20°C

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	7.25 ± 0.09	6.93 ± 0.06	6.81 ± 0.09	S	7.25 ± 0.09	7.00 ± 0.09	6.62 ± 0.08	6.33 ± 0.15
LF	6.12 ± 0.08	6.12 ± 0.08	6.15 ± 0.12	6.62 ± 0.08	6.12 ± 0.08	6.12 ± 0.12	5.75 ± 0.16	5.56 ± 0.17
CG	6.62 ^D ± 0.08	6.43 ^C ± 0.14	6.21 ^B ± 0.08	6.09 ^A ± 0.20	6.62 ^D ± 0.08	6.46 ^C ± 0.09	6.18 ^{AB} ± 0.09	6.18 ^{AB} ± 0.13
TS	6.50 ± 0.09	6.50 ± 0.09	6.18 ± 0.09	6.31 ± 0.16	6.50 ± 0.09	6.50 ± 0.13	6.21 ± 0.08	6.12 ± 0.08
PSC	6.68 ± 0.13	6.62 ± 0.12	6.25 ± 0.13	6.37 ± 0.12	6.68 ± 0.13	6.37 ± 0.15	6.31 ± 0.13	6.28 ± 0.15
CG-TS	6.93 ^D ± 0.11	6.81 ^C ± 0.09	6.53 ^B ± 0.15	6.46 ^A ± 0.16	6.93 ^D ± 0.11	6.62 ^{BC} ± 0.09	6.37 ^{AB} ± 0.11	6.25 ^A ± 0.12
CG-PSC	6.75 ± 0.09	6.75 ± 0.09	6.56 ± 0.13	6.43 ± 0.14	6.75 ± 0.09	6.62 ± 0.12	6.25 ± 0.08	6.56 ± 0.13
TS-PSC	6.87 ± 0.15	6.62 ± 0.13	6.46 ± 0.13	6.31 ± 0.09	6.87 ± 0.15	6.56 ± 0.17	6.37 ± 0.20	5.96 ± 0.15
CG-TS-PSC	7.25 ± 0.13	7.06 ± 0.09	7.00 ± 0.09	6.62 ± 0.12	7.25 ± 0.13	7.12 ± 0.12	6.87 ± 0.12	6.81 ± 0.13

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

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0%
PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

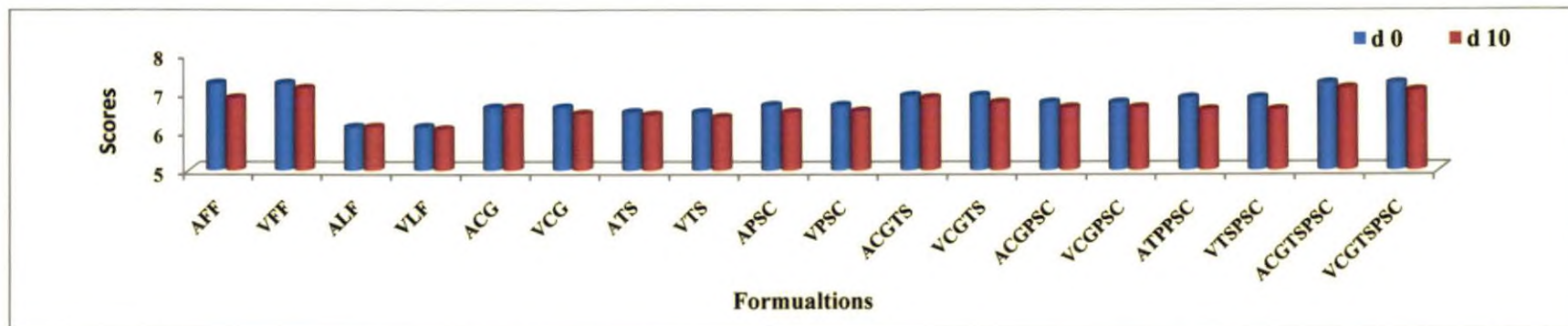


Fig.26. Effect of packaging and period of storage on texture value of low fat beef burger stored at 0-4°C

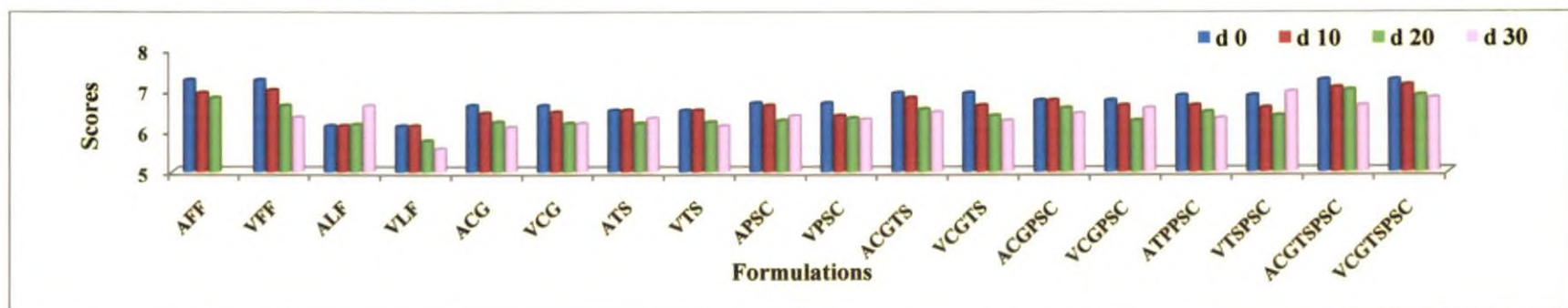


Fig.27. Effect of packaging and period of storage on texture value low fat beef burger stored at -20°C

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS;
 PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;
 CG-TS-PSC - 0.5% CG,1.5% TS & 2.0% PSC; A- Aerobic packaging ;V- Vacuum packaging.

4.4.3.4. *Saltiness*

The effect of packaging and period of storage on the saltiness scores of LFBB stored at 0-4°C and -20°C are presented in Table 21 and 22 and trend are depicted in Fig. 28 and 29, respectively.

There was no significant ($P > 0.05$) difference in the saltiness among all formulations of BB stored at 0-4°C under AP and VP on d10.

The burger with TS-PSC stored at -20°C under AP showed significantly ($P < 0.05$) higher saltiness score on d 30 compared to VP. The saltiness score in the formulation with CG-PSC and TS-PSC among combinations of FR showed significantly ($P < 0.05$) lower score at d 30 in VP compared to AP.

4.4.3.5. *Juiciness*

The effect of packaging and period of storage on the juiciness scores of LFBB stored at 0-4°C and -20°C are presented in Table 23 and 24 and trend are depicted in Fig. 30 and 31, respectively.

There was no significant ($P > 0.05$) difference in the juiciness among all formulations of BB stored at 0-4°C and -20°C under AP on d10 and in VP on d 10, 20 and 30.

4.4.3.6. *Mouth Coating*

The effect of packaging and period of storage on the mouth coating scores of LFBB stored at 0-4°C and -20°C are presented in Table 25 and 26 and trend are illustrated in Fig. 32 and 33, respectively.

The scores for mouth coating for all burgers packaged aerobically and under vacuum and stored at 0-4°C and -20°C were not significantly ($P > 0.05$) affected by the type of packaging and period of storage.

Table 21. Effect of packaging and period of storage on the saltiness of low fat beef burger stored at 0-4°C

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	6.87±0.15	6.81±0.16	6.87±0.15	6.68±0.16
LF	6.53±0.17	6.50±0.16	6.53±0.17	6.59±0.16
CG	6.37±0.08	6.25±0.06	6.37±0.08	6.37±0.12
TS	6.37±0.08	6.25±0.09	6.37±0.08	6.37±0.08
PSC	6.75±0.09	6.68±0.13	6.75±0.09	6.75±0.09
CG-TS	6.93±0.06	6.93±0.06	6.93±0.06	6.93±0.06
CG-PSC	6.43±0.11	6.28±0.08	6.43±0.11	6.43±0.11
TS-PSC	6.31±0.13	6.31±0.13	6.31±0.13	6.43±0.14
CG-TS-PSC	7.00±0.00	7.00 ^A ±0.00	7.00±0.00	6.87±0.08

Table 22. Effect of packaging and period of storage on the saltiness of low fat beef burger stored at -20°C

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	6.87 ± 0.15	6.75 ± 0.13	6.62 ± 0.15	S	6.87 ± 0.15	6.81 ± 0.13	6.81 ± 0.13	6.56 ± 0.17
LF	6.53 ± 0.17	6.50 ± 0.16	6.50 ± 0.16	6.18 ± 0.09	6.53 ± 0.17	6.43 ± 0.14	6.31 ± 0.13	6.18 ± 0.18
CG	6.37 ± 0.08	6.37 ± 0.08	6.25 ± 0.09	6.03 ± 0.21	6.37 ± 0.08	6.37 ± 0.12	6.37 ± 0.08	6.12 ± 0.08
TS	6.37 ± 0.08	6.37 ± 0.08	6.00 ± 0.00	6.37 ± 0.12	6.37 ± 0.08	6.37 ± 0.08	6.18 ± 0.09	6.00 ± 0.16
PSC	6.75 ± 0.09	6.68 ± 0.13	6.43 ± 0.11	6.15 ± 0.15	6.75 ± 0.09	6.18 ± 0.13	6.31 ± 0.13	6.15 ± 0.08
CG-TS	6.93 ± 0.06	6.93 ± 0.06	6.62 ± 0.12	6.43 ± 0.17	6.93 ± 0.06	6.75 ± 0.16	6.53 ± 0.15	6.18 ± 0.09
CG-PSC	6.43 ± 0.11	6.43 ± 0.11	6.12 ± 0.12	6.12 ± 0.08	6.43 ± 0.11	6.50 ± 0.13	6.31 ± 0.09	6.06* ± 0.06
TS-PSC	6.31 ± 0.13	6.37 ± 0.12	6.37 ± 0.12	6.12* ± 0.08	6.31 ± 0.13	6.25 ± 0.13	6.31 ± 0.13	6.06* ± 0.11
CG-TS-PSC	7.00 ± 0.00	6.93 ± 0.06	6.68 ± 0.09	6.75 ± 0.13	7.00 ± 0.00	6.87 ± 0.08	6.50 ± 0.13	6.28 ± 0.08

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

* represents significance difference between two packaging system in the same time period ($P < 0.05$)

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0%PSC; S- Spoiled.

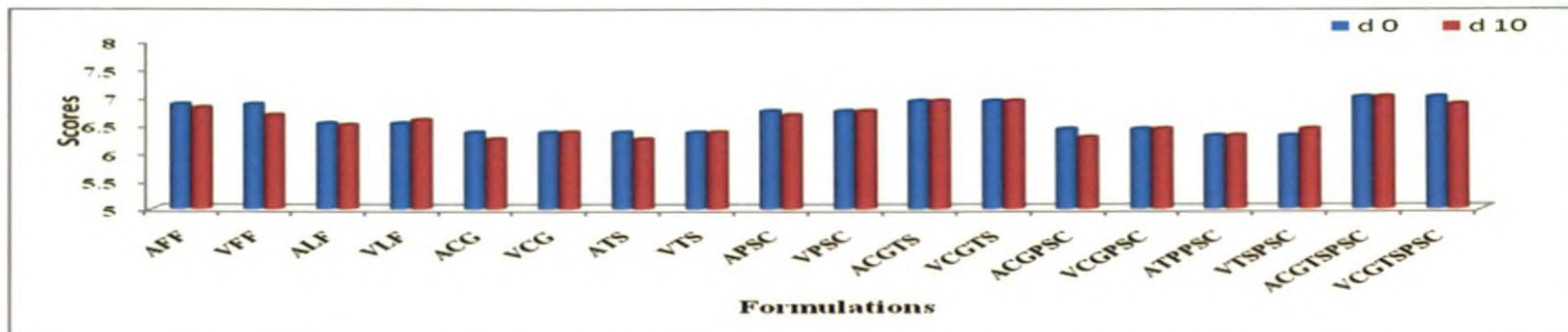


Fig.28. Effect of packaging and period of storage on saltiness of low fat beef burger stored at 0-4°C

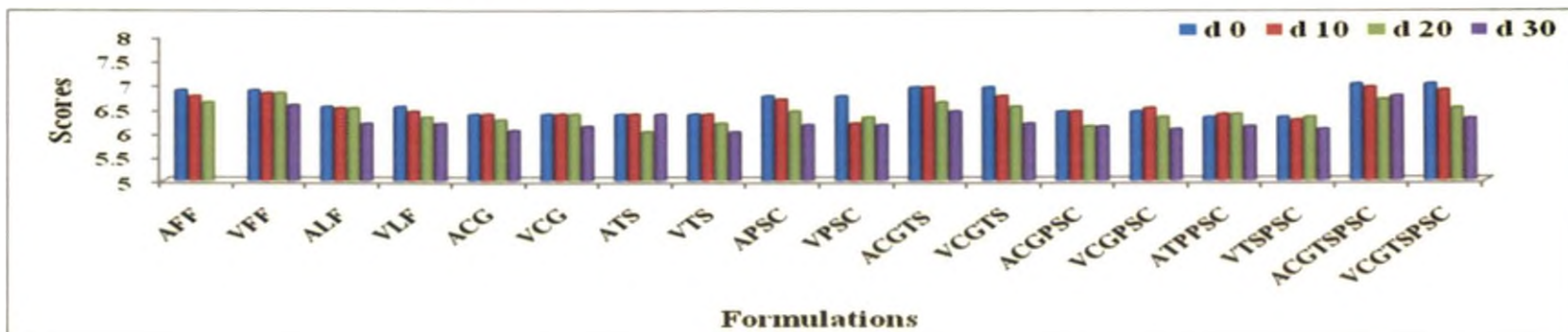


Fig.29. Effect of packaging and period of storage on saltiness of low fat beef burger stored at -20°C

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS;
 PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;
 CG-TS-PSC - 0.5% CG,1.5% TS & 2.0% PSC; A- Aerobic packaging ;V- Vacuum packaging.

Table 23. Effect of packaging and period of storage on the juiciness of low fat beef burger stored at 0-4°C

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	7.18±0.09	7.00±0.00	7.18±0.09	7.06±0.06
LF	6.00±0.00	6.00±0.00	6.00±0.00	6.18±0.13
CG	6.43±0.06	6.56±0.14	6.43±0.06	6.50±0.09
TS	6.50±0.00	6.50±0.00	6.50±0.00	6.50±0.00
PSC	6.75±0.13	6.68±0.13	6.75±0.13	6.87±0.08
CG-TS	7.00±0.09	7.00±0.09	7.00±0.09	6.83±0.14
CG-PSC	6.62±0.12	6.62±0.12	6.62±0.12	6.62±0.12
TS-PSC	6.56±0.14	6.56±0.11	6.56±0.14	6.75±0.09
CG-TS-PSC	7.06±0.06	7.06±0.06	7.06±0.06	7.00±0.00

Table 24. Effect of packaging and period of storage on the juiciness of low fat beef burger stored at -20°C

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	7.18 ± 0.09	7.07 ± 0.06	6.87 ± 0.00	S	7.18 ± 0.09	7.00 ± 0.00	7.12 ± 0.12	7.00 ± 0.18
LF	6.00 ± 0.00	6.00 ± 0.00	6.03 ± 0.03	6.21 ± 0.08	6.00 ± 0.00	6.06 ± 0.17	6.25 ± 0.09	6.18 ± 0.09
CG	6.43 ± 0.06	6.56 ± 0.06	6.75 ± 0.09	6.75 ± 0.13	6.43 ± 0.06	6.56 ± 0.06	6.62 ± 0.08	6.81 ± 0.13
TS	6.50 ± 0.00	6.50 ± 0.00	6.62 ± 0.08	6.68 ± 0.16	6.50 ± 0.00	6.56 ± 0.06	6.68 ± 0.13	6.84 ± 0.10
PSC	6.75 ± 0.13	6.75 ± 0.13	6.87 ± 0.08	6.88 ± 0.16	6.75 ± 0.13	6.81 ± 0.08	6.62 ± 0.08	6.75 ± 0.15
CG-TS	7.00 ± 0.09	6.93 ± 0.06	6.81 ± 0.09	6.93 ± 0.06	7.00 ± 0.09	7.00 ± 0.09	6.81 ± 0.13	6.75 ± 0.15
CG-PSC	6.62 ± 0.12	6.62 ± 0.12	6.68 ± 0.16	6.84 ± 0.14	6.62 ± 0.12	6.62 ± 0.12	6.68 ± 0.12	6.75 ± 0.16
TS-PSC	6.56 ± 0.14	6.50 ± 0.13	6.50 ± 0.09	6.75 ± 0.09	6.56 ± 0.14	6.56 ± 0.14	6.56 ± 0.16	6.87 ± 0.15
CG-TS-PSC	7.06 ± 0.06	7.00 ± 0.00	6.87 ± 0.08	6.87 ± 0.12	7.06 ± 0.06	6.93 ± 0.01	6.93 ± 0.06	7.06 ± 0.11

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

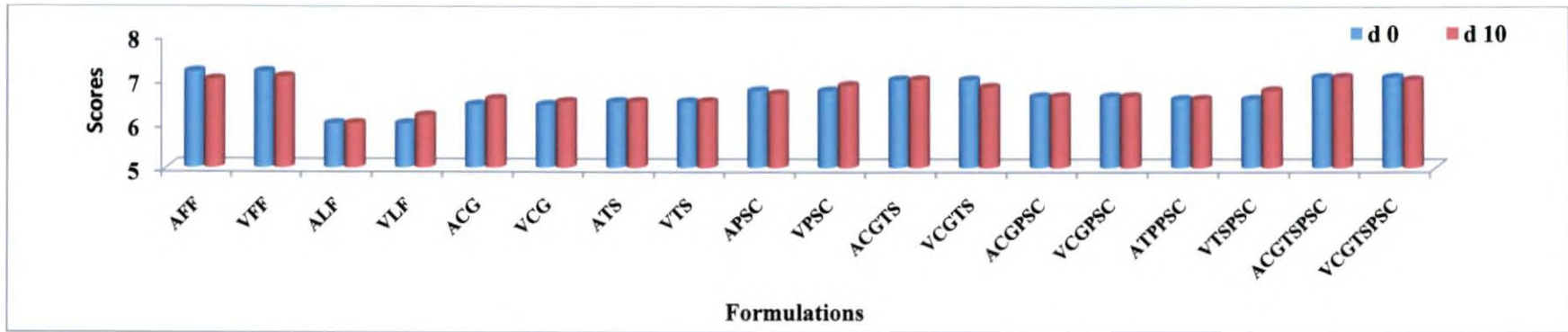


Fig.30. Effect of packaging and period of storage on juiciness of low fat beef burger stored at 0-4°C

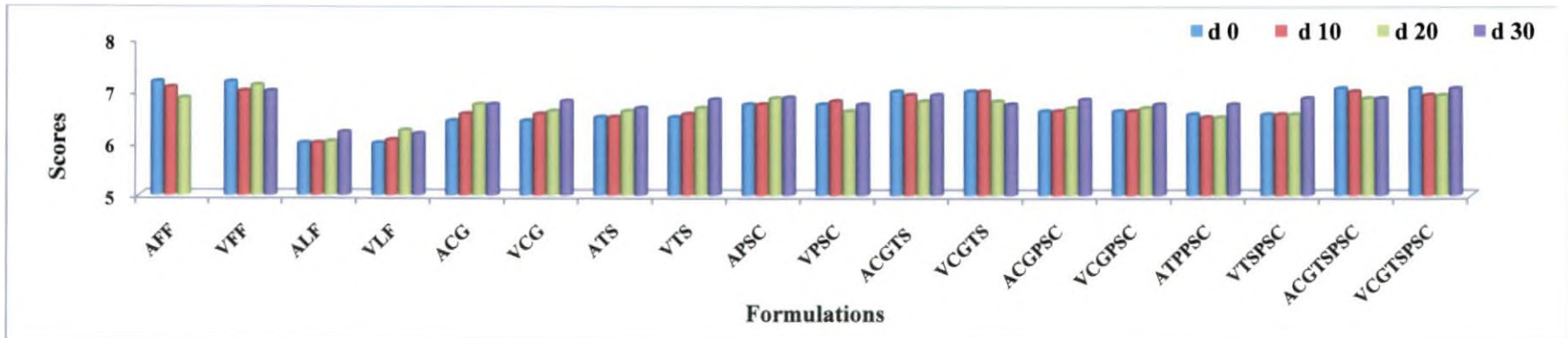


Fig.31. Effect of packaging and period of storage on juiciness of low fat beef burger stored at -20°C

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS; PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG 1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

Table 25. Effect of packaging and period of storage on mouth coating of low fat beef burger stored at 0-4°C

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	7.25±0.16	7.00±0.00	7.25±0.16	7.00±0.00
LF	6.50±0.13	6.37±0.15	6.50±0.13	6.12±0.08
CG	6.62±0.15	6.50±0.13	6.62±0.15	6.50±0.13
TS	6.12±0.08	6.12±0.08	6.12±0.08	6.00±0.13
PSC	7.00±0.00	6.75±0.16	7.00±0.00	6.68±0.12
CG-TS	6.87±0.08	6.62±0.15	6.87±0.08	7.00±0.00
CG-PSC	6.50±0.00	6.25±0.09	6.50±0.00	6.37±0.08
TS-PSC	6.62±0.08	6.50±0.00	6.62±0.08	6.62±0.08
CG-TS-PSC	7.00±0.00	7.00±0.00	7.00±0.00	7.00±0.00

Table 26. Effect of packaging and period of storage on mouth coating of low fat beef burger stored at -20°C

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	7.25 ± 0.16	7.00 ± 0.00	6.87 ± 0.08	S	7.25 ± 0.16	7.00 ± 0.00	6.75 ± 0.09	6.50 ± 0.13
LF	6.50 ± 0.13	6.37 ± 0.15	6.12 ± 0.08	5.62 ± 0.15	6.50 ± 0.13	6.25 ± 0.09	6.18 ± 0.07	6.00 ± 0.13
CG	6.62 ± 0.15	6.50 ± 0.13	6.50 ± 0.13	5.87 ± 0.20	6.62 ± 0.15	6.37 ± 0.08	6.37 ± 0.08	6.25 ± 0.09
TS	6.12 ± 0.08	6.25 ± 0.09	6.00 ± 0.00	6.00 ± 0.00	6.12 ± 0.08	6.37 ± 0.08	6.25 ± 0.09	6.12 ± 0.08
PSC	7.00 ± 0.00	7.00 ± 0.00	6.62 ± 0.08	6.37 ± 0.08	7.00 ± 0.00	7.00 ± 0.00	6.62 ± 0.08	6.37 ± 0.08
CG-TS	6.87 ± 0.08	6.75 ± 0.09	6.62 ± 0.08	6.25 ± 0.09	6.87 ± 0.08	7.00 ± 0.00	6.75 ± 0.09	6.37 ± 0.08
CG-PSC	6.50 ± 0.00	6.50 ± 0.00	6.25 ± 0.09	6.12 ± 0.08	6.50 ± 0.00	6.50 ± 0.00	6.25 ± 0.09	6.12 ± 0.08
TS-PSC	6.62 ± 0.08	6.62 ± 0.08	6.37 ± 0.08	6.25 ± 0.09	6.62 ± 0.08	6.62 ± 0.08	6.25 ± 0.09	6.06 ± 0.04
CG-TS-PSC	7.00 ± 0.00	7.00 ± 0.00	6.75 ± 0.09	6.37 ± 0.15	7.00 ± 0.00	7.00 ± 0.00	6.87 ± 0.08	6.75 ± 0.09

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0%
PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoilage.

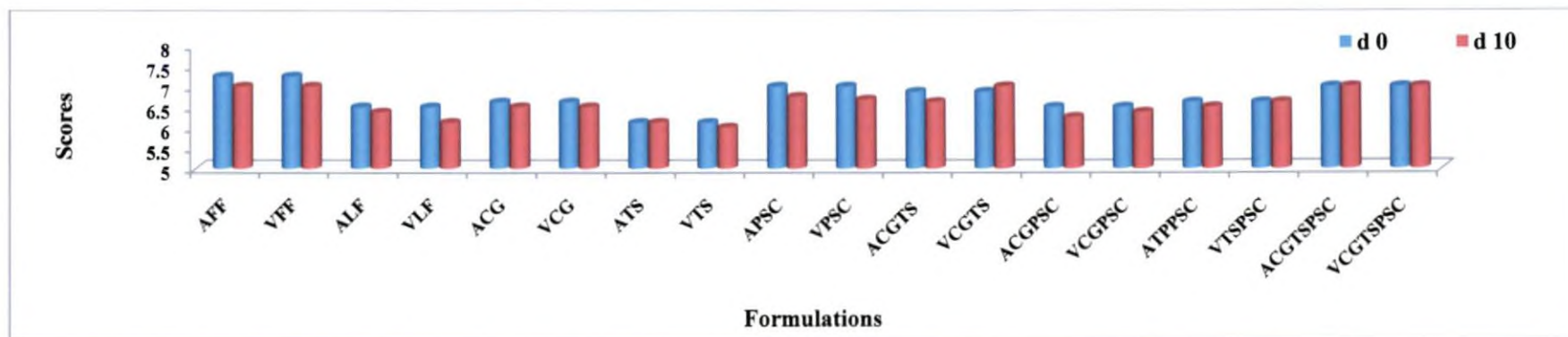


Fig. 32. Effect of packaging and period of storage on mouth coating of low fat beef burger stored at 0-4°C

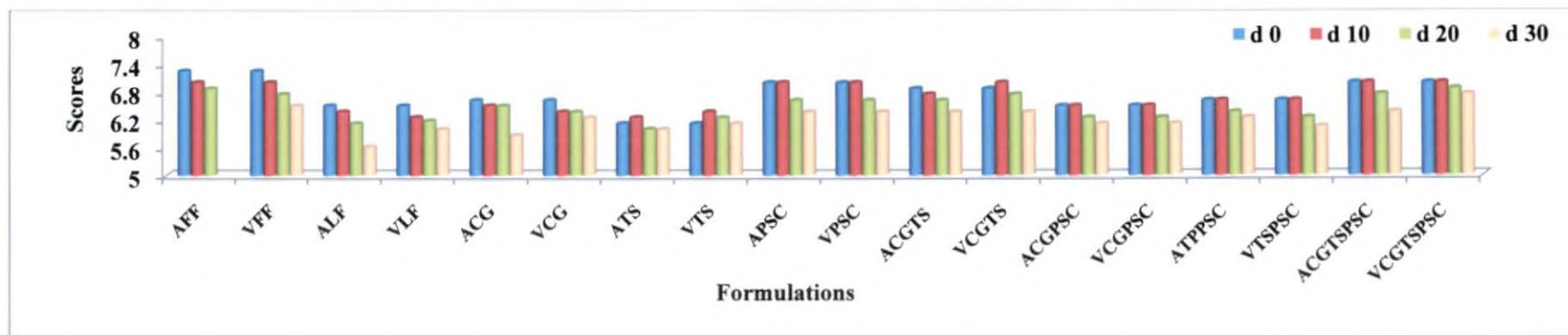


Fig. 33. Effect of packaging and period of storage on mouth coating of low fat beef burger stored at -20°C

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS;

PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;

CG-TS-PSC - 0.5% CG,1.5% TS & 2.0% PSC;A- Aerobic packaging ;V- Vacuum packaging.

Table 29. Effect of packaging and period of storage on the moisture of low fat beef burger stored at 0-4°C (%)

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	59.98 ^{aA} ±0.25	60.17 ^{aB} ±0.40	59.98 ^{aA} ±0.25	60.49 ^{aA} ±0.30
LF	69.31 ^{bB} ±0.35	69.99 ^{dA} ±0.75	69.31 ^{bB} ±0.35	70.03 ^{cA} ±0.69
CG	69.43 ^{bB} ±0.95	69.02 ^{cbA} ±0.64	69.43 ^{bB} ±0.95	69.18 ^{cdeA} ±0.65
TS	67.82 ^{bA} ±0.47	67.92 ^{bcA} ±0.46	67.82 ^{bA} ±0.47	67.38 ^{bA} ±0.34
PSC	69.38 ^B ±0.30	68.72 ^{cdA} ±0.14	69.38 ^B ±0.30	69.30 ^{deA} ±0.26
CG-TS	68.14 ^{bA} ±0.50	68.05 ^{bcA} ±0.34	68.14 ^{bA} ±0.50	67.89 ^{bcA} ±0.32
CG-PSC	68.55 ^{bAB} ±0.61	68.02 ^{bcA} ±0.43	68.55 ^{bAB} ±0.61	68.44 ^{bcdA} ±0.34
TS-PSC	68.09 ^{bA} ±0.41	68.75 ^{cdA} ±0.27	68.09 ^{bA} ±0.41	68.80 ^{cdA} ±0.27
CG-TS-PSC	67.46 ^{bA} ±0.51	67.08 ^{bA} ±0.43	67.46 ^{bA} ±0.51	67.34 ^{baA} ±0.38

Table 30. Effect of packaging and period of storage on the moisture of low fat beef burger stored at -20°C (%)

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	59.98 ^{aA} ±0.25	60.33 ^{aA} ±0.47	60.33 ^{aB} ±0.50	S	59.98 ^{aA} ±0.25	60.39 ^{aA} ±0.38	60.42 ^{aA} ±0.35	59.95 ^{aB} ±0.35
LF	69.31 ^{bB} ±0.35	70.50 ^{eb} ±0.70	70.50 ^{cb} ±0.80	70.04 ^{aC} ±0.78	69.31 ^{bB} ±0.35	70.22 ^{eA} ±0.61	70.10 ^{eA} ±0.50	69.89 ^{dB} ±0.59
CG	69.43 ^{bB} ±0.95	69.12 ^{dB} ±0.49	69.12 ^{dB} ±0.63	68.86 ^{bcA} ±0.64	69.43 ^{bB} ±0.95	69.88 ^{deA} ±0.52	69.48 ^{deA} ±0.50	69.42 ^{cA} ±0.42
TS	67.82 ^{bA} ±0.47	67.62 ^{bcA} ±0.32	67.62 ^{bcA} ±0.40	67.68 ^{abA} ±0.41	67.82 ^{bA} ±0.47	68.02 ^{deB} ±0.31	68.05 ^{bcA} ±0.38	67.44 ^{bA} ±0.48
PSC	69.38 ^{bB} ±0.30	69.12 ^{dA} ±0.19	69.20 ^{dA} ±0.23	68.84 ^{bcB} ±0.19	69.38 ^B ±0.30	68.26 ^{bcC} ±0.53	68.78 ^{cdB} ±0.33	68.40 ^{bcA} ±0.18
CG-TS	68.14 ^{bA} ±0.50	67.19 ^{bcd} ±0.30	67.91 ^{bed} ±0.31	67.55 ^{abA} ±0.38	68.14 ^{bA} ±0.50	68.45 ^{bcB} ±0.35	68.18 ^{bcD} ±0.31	68.38 ^{bcB} ±0.40
CG-PSC	68.55 ^{bAB} ±0.61	68.25 ^{bcd} ±0.43	68.25 ^{bcd} ±0.48	67.67 ^{abB} ±0.51	68.55 ^{bAB} ±0.61	68.41 ^{bcA} ±0.39	68.24 ^{bcA} ±0.38	68.37 ^{bcA} ±0.31
TS-PSC	68.09 ^{bA} ±0.41	68.91 ^{cdB} ±0.27	68.91 ^{cdB} ±0.27	68.40 ^{abA} ±0.27	68.09 ^{bA} ±0.41	68.72 ^{cdB} ±0.30	68.59 ^{cdB} ±0.33	68.10 ^{bcA} ±0.34
CG-TS-PSC	67.46 ^{bA} ±0.51	67.41 ^{bB} ±0.37	67.41 ^{bB} ±0.37	66.95 ^{bA} ±0.48	67.46 ^{bA} ±0.51	67.20 ^{bA} ±0.42	67.23 ^{bA} ±0.39	67.08 ^{bA} ±0.37

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled

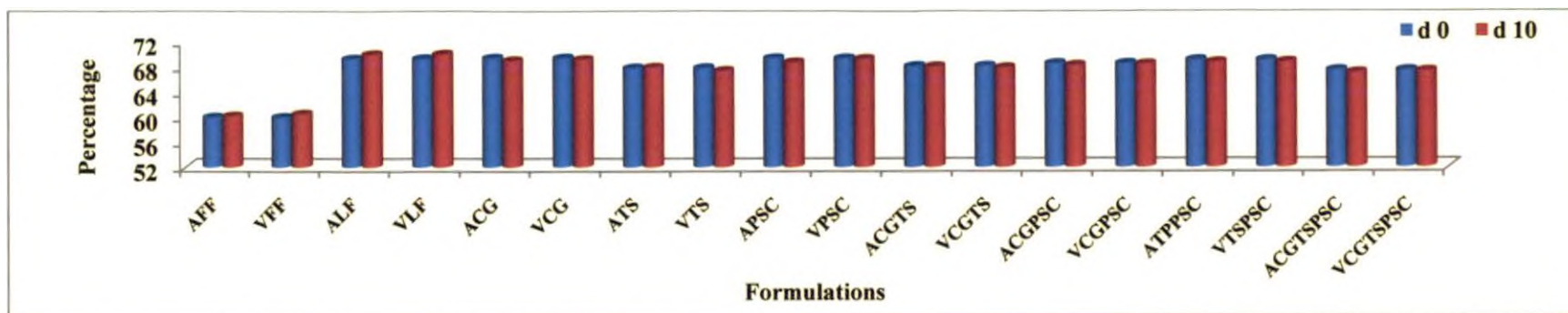


Fig.36. Effect of packaging and period of storage on moisture of low fat beef burger stored at 0-4°C

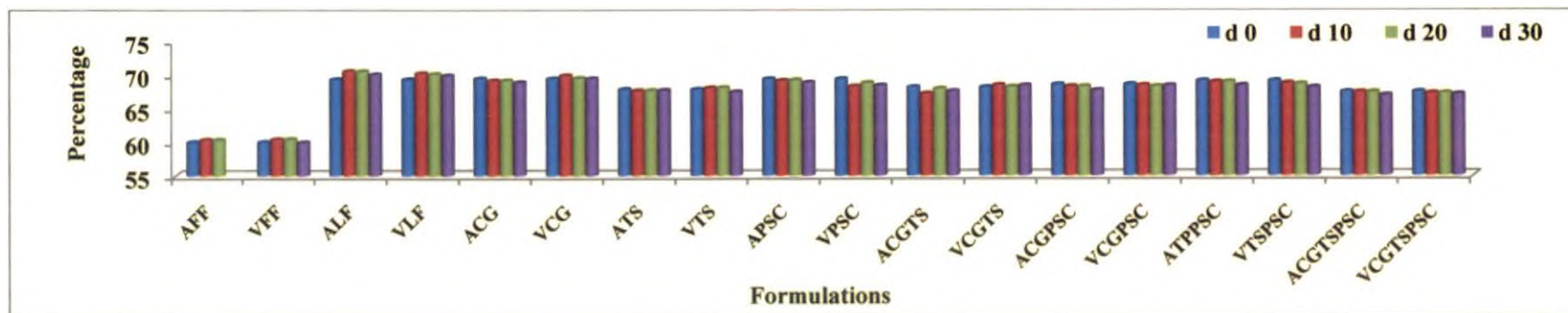


Fig.37. Effect of packaging and period of storage on moisture of low fat beef burger stored at -20°C

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS; PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG 1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

At -20°C under AP, the burger with FF and LF showed significant ($P < 0.05$) in moisture as period of storage increased from d zero to d 30. TS-PSC also showed the same trend. But PSC, CG-PSC and CG-TS-PSC showed significant ($P < 0.05$) decrease in moisture.

Burgers with TS and PSC under VP showed significant ($P < 0.05$) decrease in moisture on increase in storage period to d 30. But LF showed same trend as in AP.

There was a no significant ($P < 0.05$) difference in the moisture content in all of BB irrespective of the packaging and storage at $0-4^{\circ}\text{C}$ and -20°C .

4.4.4.2. Protein

The effect of packaging and period of storage on the protein of LFBB stored at $0-4^{\circ}\text{C}$ and -20°C are presented in Table 31 and 32 and trend are illustrated in Fig. 38 and 39, respectively.

Under AP at $0-4^{\circ}\text{C}$ only, the burgers with PSC, CG-PSC and TS-PSC showed significant ($P < 0.05$) increase in protein on d 10 while others were not different. In VP, none of the formulations were significantly ($P < 0.05$) different. CG and CG-TS showed significantly ($P < 0.05$) lower protein on d 10 in AP compared to that in VP. Remaining formulations did not show any significant ($P > 0.05$) difference between the days of storage.

At -20°C in AP, CG-TS showed significant ($P < 0.05$) increase in protein on d 10, d 20 and d 30. Other formulations did not show any significant difference ($P > 0.05$). Under VP, only FF, LF and PSC showed significant ($P < 0.05$) increase in protein content as the period of storage increased. On d 20 BB with TS showed significantly ($P < 0.05$) higher protein in VP compared to AP. TS-PSC and CG-TS-PSC showed significantly ($P < 0.05$) higher protein content in AP compared to VP on d 30.

Table 31. Effect of packaging and period of storage on the protein of low fat beef burger stored at 0-4°C (%)

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	16.82 ^a ±0.55	17.07 ^a ±0.20	16.82 ^a ±0.55	17.20 ^a ±0.21
LF	19.23 ^b ±0.21	19.57 ^{bc} ±0.13	19.23 ^b ±0.21	19.69 ^{cd} ±0.11
CG	17.72 ^a ±0.78	17.89 ^{c*} ±0.51	17.72 ^a ±0.78	18.28 ^{ab*} ±0.47
TS	19.13 ^b ±0.61	19.21 ^b ±0.38	19.13 ^b ±0.61	19.32 ^{bc} ±0.43
PSC	20.10 ^{bcA} ±0.26	20.92 ^{cdB} ±0.17	20.10 ^{bc} ±0.26	20.39 ^{cd} ±0.15
CG-TS	17.78 ^a ±0.67	17.41 ^{a*} ±0.44	17.78 ^a ±0.67	17.90 ^{a*} ±0.43
CG-PSC	19.29 ^{bA} ±0.42	20.17 ^{bcB} ±0.35	19.29 ^b ±0.42	19.42 ^c ±0.45
TS-PSC	20.07 ^{bcA} ±0.59	21.10 ^{dB} ±0.34	20.07 ^{bc} ±0.59	20.49 ^{cd} ±0.47
CG-TS-PSC	20.54 ^c ±0.30	21.75 ^d ±0.26	20.54 ^c ±0.30	20.77 ^d ±0.18

Table 32. Effect of packaging and period of storage on the protein of low fat beef burger stored at -20°C (%)

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	16.82 ^a ±0.55	17.03 ^a ±0.24	17.26 ^a ±0.25	S	16.82 ^{aA} ±0.55	17.12 ^{aB} ±0.24	17.61 ^{aC} ±0.27	17.66 ^{aC} ±0.21
LF	19.23 ^b ±0.21	19.45 ^{bc} ±0.19	19.57 ^{bcd} ±0.19	19.71 ^{bcd} ±0.25	19.23 ^{bA} ±0.21	19.60 ^{bC} ±0.12	19.62 ^{aC} ±0.25	20.47 ^{cdB} ±0.54
CG	17.72 ^a ±0.78	17.87 ^a ±0.53	18.17 ^a ±0.50	18.30 ^a ±0.61	17.72 ^a ±0.78	18.23 ^a ±0.53	18.65 ^a ±0.55	18.73 ^{ab} ±0.44
TS	19.13 ^b ±0.61	19.44 ^{bc} ±0.48	19.23 ^{abc} ±0.38	19.51 ^{abc} ±0.39	19.13 ^b ±0.61	19.47 ^b ±0.50	19.72 ^{a*} ±0.54	19.53 ^{bc} ±0.14
PSC	20.10 ^{bc} ±0.26	20.39 ^{cd} ±0.17	20.31 ^{cdc} ±0.14	20.39 ^{dc} ±0.11	20.10 ^{bcA} ±0.26	20.45 ^{bb} ±0.14	20.69 ^{aC} ±0.17	20.98 ^{dB} ±0.41
CG-TS	17.78 ^{aA} ±0.67	18.27 ^{abB} ±0.51	18.51 ^{abC} ±0.51	18.73 ^{abC} ±0.49	17.78 ^a ±0.67	17.94 ^a ±0.43	18.14 ^a ±0.44	18.13 ^a ±0.50
CG-PSC	19.29 ^b ±0.42	19.62 ^c ±0.42	19.61 ^{bcd} ±0.32	19.82 ^{cd} ±0.38	19.29 ^b ±0.42	19.48 ^b ±0.47	19.72 ^a ±0.47	19.98 ^{bcd} ±0.54
TS-PSC	20.07 ^{bc} ±0.59	20.33 ^{cd} ±0.46	20.40 ^{dc} ±0.39	20.74 ^{cde*} ±0.55	20.07 ^{bc} ±0.59	20.42 ^b ±0.50	20.67 ^a ±0.54	20.53 ^{cd} ±0.23
CG-TS-PSC	20.54 ^c ±0.30	21.01 ^e ±0.26	20.95 ^e ±0.22	21.30 ^{e*} ±0.30	20.54 ^c ±0.30	20.58 ^b ±0.14	20.64 ^b ±0.82	20.97 ^d ±0.18

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

* represents significance difference between two packaging system in the same time period (P<0.05)

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen
CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S-Spoiled.

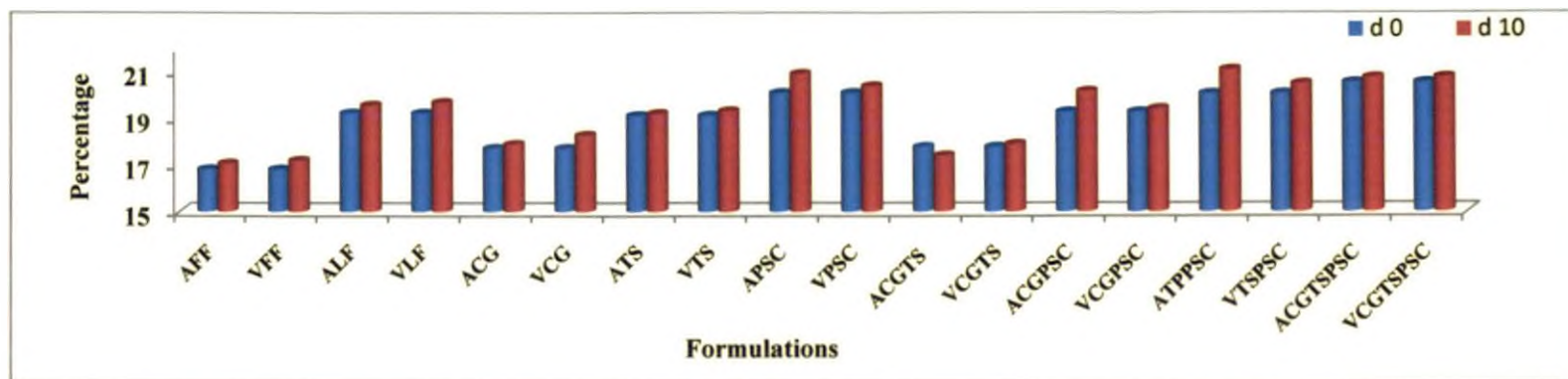


Fig. 38. Effect of packaging and period of storage on protein of low fat beef burger stored at 0-4°C

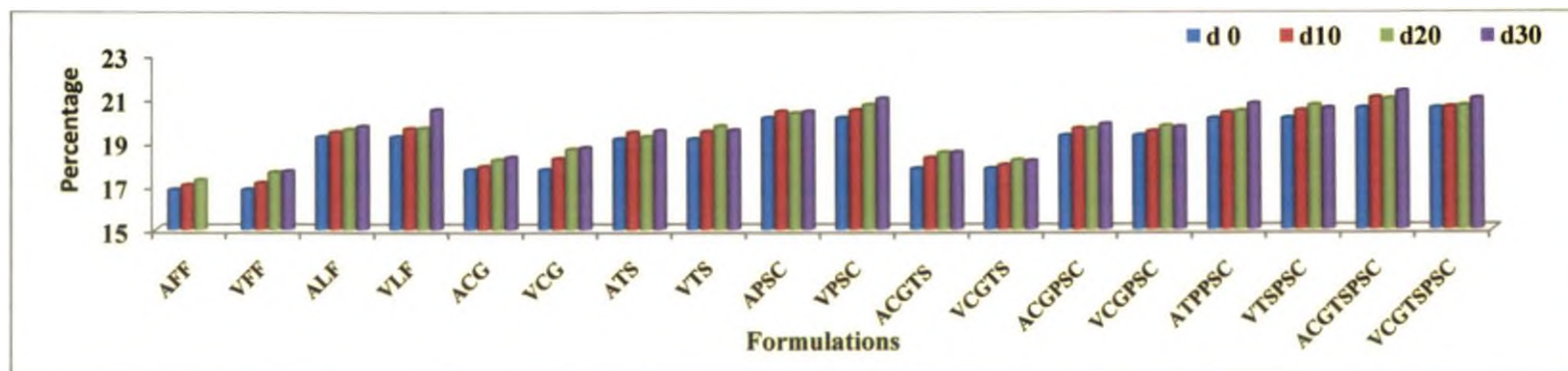


Fig.39. Effect of packaging and period of storage on protein of low fat beef burger stored at -20°C

FF- Full Fat; LF- Low Fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS;
 PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC;
 CG-TS-PSC - 0.5% CG 1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

Table 33. Effect of packaging and period of storage on the fat of low fat beef burger stored at 0-4°C (%)

Formulations	Aerobic packaging		Vacuum packaging	
	Period of storage, d		Period of storage, d	
	0	10	0	10
FF	20.53 ^a ±0.15	21 ^a .00±0.22	20.53 ^a ±0.15	20.96 ^a ±0.24
LF	5.67±0.27	5.64±0.16	5.67±0.27	5.69±0.16
CG	5.05±0.08	5.34±0.12	5.05±0.08	5.35±0.10
TS	5.85±0.32	5.64±0.20	5.85±0.32	5.63±0.20
PSC	5.73±0.16	5.69±0.14	5.73±0.16	5.66±0.14
CG-TS	5.26±0.30	5.86±0.24	5.26±0.30	5.89±0.26
CG-PSC	5.59±0.25	5.37±0.16	5.59±0.25	5.40±0.17
TS-PSC	5.61±0.31	5.65±0.24	5.61±0.31	5.56±0.19
CG-TS-PSC	5.45±0.22	5.63±0.21	5.45±0.22	5.76±0.26

Table 34. Effect of packaging and period of storage on the fat of low fat beef burger stored at -20°C (%)

Formulations	Aerobic packaging				Vacuum packaging			
	Period of storage, d				Period of storage, d			
	0	10	20	30	0	10	20	30
FF	20.53 ^a ± 0.15	20.96 ^a ± 0.21	21.10 ^a ± 0.23	S	20.53 ^a ± 0.15	21.03 ^a ± 0.21	21.10 ^a ± 0.21	21.33 ^a ± 0.14
LF	5.67 ± 0.27	5.57 ± 0.11	5.67 ± 0.11	5.78 ± 0.10	5.67 ± 0.27	5.65 ± 0.15	5.78 ± 0.14	5.88 ± 0.14
CG	5.05 ± 0.08	5.40 ± 0.11	5.42 ± 0.10	5.61 ± 0.12	5.05 ± 0.08	5.36 ± 0.10	5.44 ± 0.10	5.55 ± 0.11
TS	5.85 ± 0.32	5.68 ± 0.19	5.76 ± 0.19	5.77 ± 0.20	5.85 ± 0.32	5.65 ± 0.20	5.72 ± 0.20	5.80 ± 0.20
PSC	5.73 ± 0.16	5.73 ± 0.14	5.80 ± 0.16	5.89 ± 0.17	5.73 ± 0.16	5.70 ± 0.14	5.80 ± 0.14	5.87 ± 0.14
CG-TS	5.26 ± 0.30	5.91 ± 0.26	6.00 ± 0.26	6.09 ± 0.25	5.26 ± 0.30	5.92 ± 0.16	5.89 ± 0.27	5.96 ± 0.28
CG-PSC	5.59 ± 0.25	5.40 ± 0.16	5.52 ± 0.14	5.58 ± 0.16	5.59 ± 0.25	5.39 ± 0.17	5.61 ± 0.13	5.66 ± 0.13
TS-PSC	5.61 ± 0.31	5.88 ± 0.19	5.65 ± 0.19	5.74 ± 0.19	5.61 ± 0.31	5.60 ± 0.18	5.73 ± 0.17	5.79 ± 0.17
CG-TS-PSC	5.45 ± 0.22	5.65 ± 0.20	5.73 ± 0.23	5.82 ± 0.24	5.45 ± 0.22	5.69 ± 0.21	5.78 ± 0.21	6.01 ± 0.23

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

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

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen

CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0%

PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC; S- Spoiled.

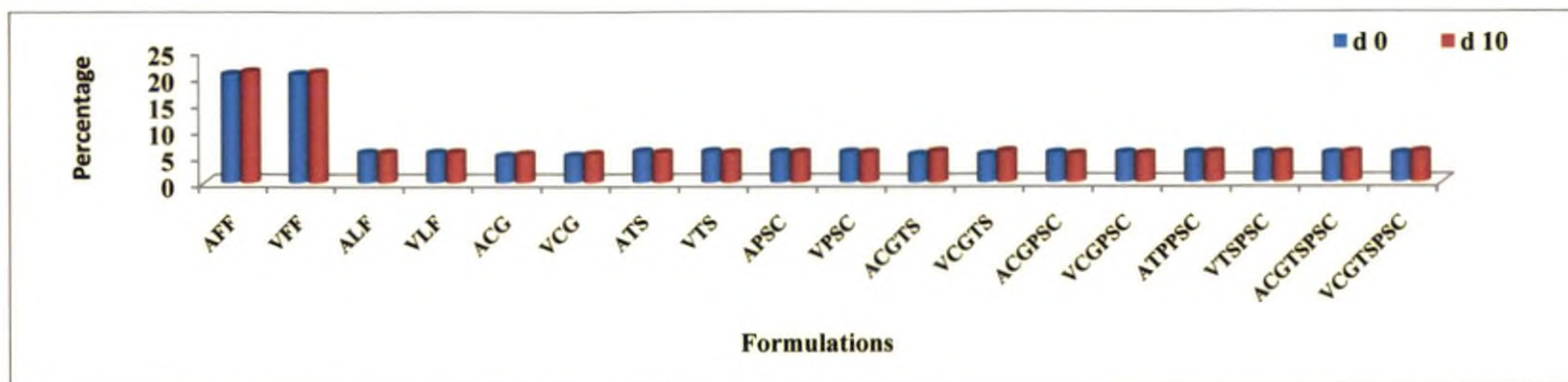
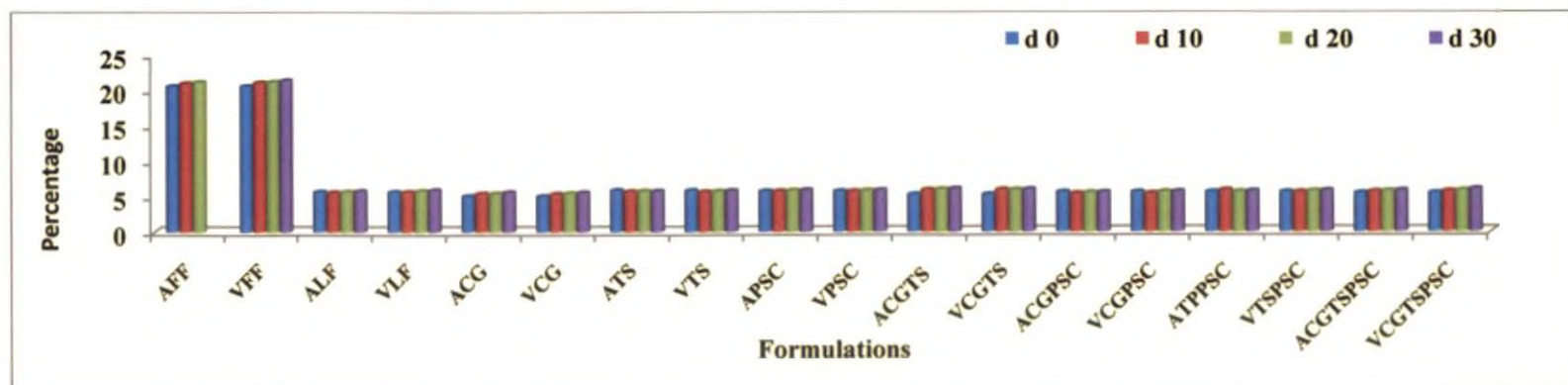


Fig.40. Effect of packaging and period of storage on fat of low fat beef burger stored at 0-4°C



41. Effect of packaging and period of storage on fat of low fat beef burger stored at -20°C

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS; PSC - 2%PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG 1.5% TS & 2.0% PSC; A- Aerobic packaging; V- Vacuum packaging.

4.4.4.3. Fat

The effect of packaging and period of storage on the fat of LFBB stored at 0-4°C and -20°C are presented in Table 33 and 34 and trend are illustrated in Fig.40 and 41, respectively.

The beef burgers showed no significant ($P < 0.05$) difference in fat content during the entire period of storage at 0-4°C from d zero to d 10 in AP and VP and also at -20°C from d zero to d 30.

4.5 COST OF PRODUCTION

The cost of production of 1 kg uncooked LFBB of different formulations is presented in Table. 35

Table.35. Cost of production of different formulations of low fat beef burger

Formulations	Cost of Productions (Rs/kg burger)
FF	114
LF	114
CG	113
TS	112
PSC	113
CG-TS	111
CG-PSC	113
TS-PSC	112
CG-TS-PSC	111

FF- Full fat; LF- Low fat; CG- Carrageenan; TS- Tapioca Starch; PSC- Pork Skin Collagen; CG - 0.5% CG; TS - 1.5% TS; PSC - 2% PSC; CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0%PSC.

Discussion

DISCUSSION

The study was taken up with the objective of developing a palatable and economic formulary for Low Fat Beef Burger (LFBB) with Carrageenan (CG), Tapioca starch (TS), Pregelatinised pork skin collagen (PSC) and their blends as fat replacers (FR) based on the physical and chemical parameters, cooking characteristics, Warner-Bratzler Shear Force (WBSF), Hunter L*, a*, b* values, proximate composition, nutritive value, organoleptic qualities of the BB. The keeping quality of LFBB under aerobic and vacuum packaging (AP and VP) on storage at 0-4°C and -20°C were also assessed by the purge loss, Thiobarbituric Acid Reactive Substances (TBARS) and effect on proximate composition. Seven different formulations of LFBB were prepared with 5 per cent fat and CG, TS, PSC and their blends as FR. Full fat 20 per cent and low fat 5 per cent BB without FR were used as controls. The results are discussed in this Chapter.

5.1. PHYSICO-CHEMICAL CHARACTERISTICS OF LOW FAT BEEF BURGER

5.1.1. pH

pH of cooked burger in all formulations were significantly ($P < 0.05$) higher than their corresponding uncooked burgers and ranged from 6.18 ± 0.02 in FF to 6.42 ± 0.02 in PSC. Cooked burgers with FF and LF controls were significantly ($P < 0.05$) acidic compared to others with FR. Cooking significantly ($P < 0.05$) reduced the acidity of all the burgers. However, the addition of CG, PSC and TS as FR in LFBB significantly ($P < 0.05$) reduced the acidity in cooked burgers compared to FF and LF controls.

An increase in pH after thermal processing was also observed by Claus and Hunt (1989) in low fat, high added water bologna formulated with texture modifying agents. They suggested the reason for the higher pH as inactivation of enzymes during cooking.

Ho *et al.* (1995) also reported an increase in pH due to addition of 0.4 per cent CG and ascribed it to the basic properties of CG.

Selvakumar (2009) also observed an increase in the pH of cooked low fat frankfurter on addition of PSC as FR and Osburn *et al.* (1997) in reduced fat bologna. They opined that increased pH of collagen resulted in higher pH of the batter. Eilert *et al.* (1993) in meat batters manufactured with modified beef connective tissue also reported similar observations and attributed it to the higher pH of the connective tissue.

5.1.2. Cooking Characteristics

5.1.2.1. Cook Yield and Cook Loss

Cook yield of CG-TS and CG-TS-PSC was significantly ($P < 0.05$) higher *i.e.*, 86.47 and 85.84 per cent, respectively among all formulations. LFBB with combinations of FR showed significantly ($P < 0.05$) higher CY than those with single FR, evidently CG-TS and CG-TS-PSC. Correspondingly, the percentage CL in FF and LF controls were 36.61 and 32.23 per cent, respectively. The CL showed inverse relationship with cooking yield.

Troutt *et al.* (1992) also showed that beef burger containing 30 per cent fat had the greatest CL. In low fat ground beef products cook loss increased as fat content decreased from 20 per cent to 7-8 per cent (Hoelscher *et al.* 1987; Cannel *et al.* 1989; Bullock *et al.* 1994). Serdaroglu and Degirmencoglu (2004) also reported that the CY 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.

Significant ($P < 0.05$) improvement in CY due to addition of CG in LFBB is in agreement with the findings of Kumar and Sharma (2004) in low fat pork patties and also Suman (2001) in low fat buffalo patties. Similarly, Egbert *et al.*

(1991) reported that beef burgers containing CG had significantly ($P < 0.05$) lower total CL than 20 per cent fat beef burger.

Significant ($P < 0.05$) improvement in CY due to increasing levels of PSC in LFBB 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 Adegbaaju (2001) in pork sausages and in low fat frankfurter by Selvakumar (2009). Allen *et al.* (1999) observed significantly ($P < 0.05$) increased CY in LFBB with blends of TS and CG

The reduction in CL with subsequent increase in the CY 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 meat products. Collagen work synergistically with myofibrillar structures in meat proteins to bind water.

5.1.2.2. Dimensional Shrinkage

The burger with CG-PSC and CG-TS-PSC showed significantly ($P < 0.05$) the lowest DS of 13.09 and 13.21 per cent, respectively among all formulations. But FF had significantly ($P < 0.05$) the highest DS of 30.92 per cent. LFBB with blends of CG, TS and PSC as FR showed significantly ($P < 0.05$) lower DS than those with either single FR or controls.

Similar result are reported by Troy *et al.* (1999) in LFBB on addition of 0.5 per cent CG and 1 - 1.5 per cent TS and their blends significantly ($P < 0.05$) reduced the diameter by 18.9 - 22.5 per cent as result of high fat loss and moisture during cooking. El-Magoli *et al.* (1996) reported that reduction in fat level from 22 to 11 per cent reduced shrinkage in low fat ground beef patties and

Serdaroglu and Degirmencioglu (2004) in turkish meat balls formulated with 20 per cent fat.

The DS during the cooking process was due to the denaturation of the meat proteins with loss of water and fat which could be reduced with the addition of blends of CG, TS and PSC at the level of 0.5, 1.5 and 2 per cent, respectively in LFBB.

5.1.2.3. Fat Retention Percentage

CG-TS-PSC formulation had the significantly ($P < 0.05$) highest FRP of 97.66 followed by CG-PSC; TS-PSC, CG and CG-TS, TS, in the decreasing order of FRP. On the contrary, BB with FF had the lowest FRP of 90.80 per cent. In order to increase FRP it is ideal to incorporate CG, TS, PSC or their blends in LFBB.

This observation is supported by Sheard *et al.* (1998) who reported that in high fat burgers, the fat loss was much more than in low fat products regardless of the method of cooking. Khalil (2000) studied the quality characteristics of low fat beef patties modified with corn starch and water and observed significant ($P < 0.05$) increase in fat retention on cooking with increase in replacement of fat.

The improvement in FRP was reported while reducing the fat content from around 20 to 10 per cent and with the addition of CG (EI-Magoli, 1996; Suman, 2001; Kumar and Sharma, 2004). Collagen from various sources has been used as an ingredient to improve fat retention (Webster *et al.*, 1982; Jobling, 1984).

5.1.2.4. Moisture Retention Percentage

The blends of CG, TS and PSC as FR showed higher MRP compared to single FR. Among the blends, CG-PSC and CG-TS-PSC recorded MRP of 74.41 and 74.36, respectively. LF showed significantly ($P < 0.05$) the lowest percentage of 66.36.

Same results are reported by Kumar and Sharma (2004) in low fat ground beef patties by addition of 0.5 per cent of CG. Foegeding and Ramsey (1986) opined that Kappa and iota CG at levels of < 1 per cent was the most beneficial for holding water. LFBB containing TS had the ability to bind and retain the moisture (Egbert *et al.* 1991; Hart and Price, 1993; Desmond *et al.* 1998).

5.1.3. Water Holding Capacity

The WHC of LFBB with CG-TS-PSC was 95.36 per cent equivalent to that of BB with FF. As a result of the increased WHC these burgers were more succulent and juicy. Burgers with blends of FR showed significantly higher WHC compared to single FR. Among single FR, TS showed significantly highest WHC of 94.73 per cent.

LFBB without any FR had the significantly lowest WHC among all. Allen *et al.* (1999) also reported that decreasing fat content from 30 to 5 per cent significantly decreased WHC. Hart and Price (1993) and Troy *et al.* (1999) reported that LFBB containing TS increased their WHC. Starch favors formations of stronger heat induced structure through swelling of starch granules embedded in protein gel matrix and increasing water binding. Same observation was made by Khalil (2000).

Ulu (2004) found that the addition of CG at levels of 0.5–1 per cent in low-fat meatballs improved their WHC which is in concurrence with the present results. CG being a hydrocolloid consisting of two sulphate groups per repeat unit of disaccharide, it can improve the water retention in meat products by the formation of water-protein complex as suggested by Cofrades *et al.* (2000)

Webster *et al.* (1982) also reported the ability of collagen to increase the WHC in processed products. Schnell (1999) reported that PSC incorporation significantly ($P < 0.05$) increased the WHC in low fat bologna. PSC improves protein functionality and WHC through the immobilization of free water.

5.1.4. Warner-Bratzler Shear Force

The BB with CG-TS-PSC was the tenderest with the WBSF value 5.30 N equivalent to FF which was 5.35 N. This indicated that LFBB with a blend of the FR were more succulent than and as tender as the FF burger. Among the LFBB with combinations of FR, in the decreasing order of tenderness were, CG-TS, CG-PSC and TS-PSC. If PSC alone was used as FR, the burgers were significantly ($P < 0.05$) tenderer than CG and TS. The LFBB with 5 per cent fat and without any FR were the toughest with WBSF 6.73. This showed that fat has an inverse relationship with WBSF as fat is less resistant to shear force compared to hard proteinateous matrix.

Troy *et al.* (1999) also reported that higher WBSF value for BB with 4 – 8 per cent fat compared to those with 20 per cent fat. Addition of CG in different formulations of LFBB significantly ($P < 0.05$) reduced the shear force values. Egbert *et al.* (1991) observed decrease in shear force in cooked patties on adding 0.5 per cent CG. Addition of TS in LFBB gave desirable tenderness of burger which significantly reduced shear force (Hart and Price, 1993; Bullock, 1995; Desmond *et al.* 1998; Allen *et al.* 1999). Chavez (1983) observed an increased collagen level in hamburger/patties which decreased the mean shear force. Osburn *et al.* (1997) and Osburn and Mandigo (1998) also observed increased water binding and gelling ability of collagen which improved the texture by diluting the stronger binding myofibrillar proteins in low fat formulations.

5.1.5. Colour

The L^* (lightness) values of cooked burgers with CG-TS-PSC was 36.57 and of FF control was 36.70 which were not significantly ($P > 0.05$) different. The a^* (redness) values of LFBB with FR and FF control were less reddish (more bluish) than PSC. The b^* (yellowness) values of formulations with PSC, CG-TS, TS-PSC were also significantly similar to FF burgers and were less yellowish (more greenish). This indicated that 5 per cent LFBB with blends of CG, TS and PSC as FR were as acceptable as 20 per cent FF BB. The LFBB with CG-TS-



PSC was lighter than the remaining formulations. Replacement of lean meat with PSC reduced the amount of myoglobin responsible for the red colour which resulted in light red BB. Cooking decreased the redness of the products which was expected due to the denaturation of the myoglobin.

Osburn *et al.* (1997) and Selvakumar (2009) opined that addition of connective tissue gels may help lighten product colour in reduced fat comminuted meat products. Arganosa *et al.* (1987) also observed increase in collagen level and reduction in lean tissue resulted in higher L^* value of patties. The patties formulated with starch were possibly darker because of a higher degree of non enzymatic browning reaction in meat due to reactivity of starch with protein (Khalil, 2000).

5.2. PROXIMATE AND MINERAL COMPOSITION

5.2.1. Lean Beef Trimmings and Pork Skin Collagen

The percentage of moisture, protein, fat, carbohydrate and ash content of lean beef trimming were 76.58 ± 0.37 , 19.29 ± 0.30 , 1.76 ± 0.01 , 1.2 ± 0.01 and 1.18 ± 0.01 , respectively and in PSC 72.52 ± 0.44 , 25.61 ± 0.23 , 0.58 ± 0.03 , 0.38 ± 0.01 and 0.58 ± 0.02 , respectively. Fat content in the beef trimmings and PSC were < 1.76 per cent. Similar results are reported by Selvakumar (2009) also.

5.2.2. Uncooked and Cooked Low Fat Beef Burger

Proximate composition of different formulations of uncooked LFBB with FR did not show any significant ($P > 0.05$) difference in moisture, protein, fat, carbohydrate and ash. Moisture content in FF was 59.98 per cent which was significantly ($P < 0.05$) the lowest among all. In LFBB, the fat content ranged between 5.05 and 5.85 per cent while in FF formulations, fat was 20.53 per cent. Addition of FR in LFBB significantly ($P < 0.05$) increased protein and mineral content in all formulations in uncooked BB. Burger with CG-TS-PSC had significantly ($P < 0.05$) the highest mineral content especially Na. Whereas, highest content K in CG-TS, Ca in CG and P in TS.

On comparison of the proximate and mineral composition of the uncooked and cooked BB, significant ($P < 0.05$) reduction in the moisture content and a corresponding increase in the protein, fat, carbohydrate, ash, Na, K, Ca and P content was noticed. It is observed that the addition of FR in LFBB significantly ($P < 0.05$) increased the protein and mineral content and a blend of CG, TS and PSC was more acceptable.

The data on moisture and fat depicted an inverse relationship. The results are in agreement with that of Troy *et al.* (1999) in LFBB, Kumar and Sharma (2004) in low fat pork patties, Khalil (2000) in low fat beef patties, Desmond *et al.* (1998) in LFBB. They also reported that the FF control had lower moisture and greater fat content than low fat treatments.

Addition of PSC in LFBB significantly ($P < 0.05$) increased the protein content compared to LF formulations. Similar result was reported by Selvakumar (2009) in formulations of low fat frankfurter.

Cierach *et al.* (2009) also reported increased mineral content in BB with hydrocolloid CG as a result of higher content of potassium, sodium, magnesium and calcium sulphate esters of galactose and 3, 6-anhydrogalactose, copolymers.

5.3. NUTRITIONAL VALUE

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

The total RDA of calorific value 11.68 per cent in FF could be reduced to the range of 6.36 to 7.18 per cent in LFBB, with either single or blends of FR. But in all formulations of LFBB, the contribution of fat to RDA of calorific value ranged from 2.22 to 2.42 only. The percentage contribution of carbohydrate in LFBB to RDA of calorific value was from 0.72 to 1.15 and protein from 3.30 to 3.92.

The contribution of calories from fat to the RDA was far below the

recommended 30 per cent (NRC, 1989; WHO, 2003) in all formulation of LFBB with blends of FR. 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 and minerals

The LFBB with TS-PSC showed significantly ($P < 0.05$) higher percentage of 35.63 daily value of protein among combinations of FR, while in burgers with PSC alone was 35.98 per cent among the single FR. More than one third of the daily requirement of protein is obtained from 100g of LFBB.

The burger with CG-TS-PSC contained significantly ($P < 0.05$) the highest Na and P among all formulations, which were 39.42 and 27.0 per cent, respectively. K content in LFBB ranged from 13.74 to 17.68 per cent. LFBB with CG had the significantly ($P < 0.05$) highest Ca content of 5.27 per cent. LFBB with FR are good sources of Na, K and P but not of Ca.

RDA of protein was taken as 60g (ICMR, 1990) and that of Na- 2400mg; K- 3500mg (Code of Federal Regulations, 1995), Ca-800mg and P- 800mg (NRC, 1989).

5.4. EFFECT OF PACKAGING AND STORAGE ON QUALITY OF LFBB

At 0-4°C, under AP and VP the BB could be stored up to d 10 only without spoilage. Similarly, at -20°C all the formulations of BB could be stored up to d 30 in both AP and VP. But FF control could be stored only up to d 20 in AP without spoilage.

5.4.1. Purge Loss

At 0-4°C, LFBB without FR had significantly ($P < 0.05$) the highest PL in AP and VP systems and days of storage. The blends of FR showed significantly ($P < 0.05$) lower PL compared to the single FR in both types of packaging and

storage. CG-TS-PSC showed the lowest PL of 0.39 per cent on d 10 at 0 - 4°C under VP. Same trend was observed in VP. The BB with PSC on d 10 in AP showed significantly ($P < 0.05$) higher PL compared to VP. Otherwise, packaging did not have any significant ($P > 0.05$) effect on PL at 0-4°C.

At -20°C as the period of storage increased from d 10 to d 30, the PL significantly ($P < 0.05$) decreased in all formulations except in BB with LF and PSC, which significantly ($P < 0.05$) increased. LFBB with blends of FR showed significantly ($P < 0.05$) lower PL compared to single FR in both types of packaging as in 0-4°C. CG-TS-PSC and CG-TS showed the lowest PL of 0.38 and 0.39 per cent, respectively under AP. Similar results were obtained in VP.

These observations indicate that LFBB with CG-TS-PSC could reduce purge loss significantly ($P < 0.05$) at 0-4°C in both AP and VP and could be stored up to d 10 only. At -20°C the LFBB with FR could be stored up to d 30 in both AP and VP with significantly ($P < 0.05$) lower PL. However, LFBB with blends of FR, viz., CG-TS-PSC and CG-TS had the lowest PL. Lowering of fat from 20 per cent to 5 per cent in BB without FR will lead to significant ($P < 0.05$) increase in PL in AP and VP and storage at 0-4°C and -20°C. The CG, PSC and TS could retain considerable portion of added water in LFBB and thereby reduced the PL on storage.

These observations are supported by the findings of Cierach *et al.* (2009) and Candogan and Kolsarici (2003) who reported that addition of 0.3-0.7 per cent CG in LFBB significantly ($P < 0.05$) decreased PL. Shand *et al.* (1994) also found that kappa carrageenan addition at 0.5 and 1.0 per cent level to structured beef role reduced purge in vacuum packaged slices during refrigerated storage. Prabhu *et al.* (2004) reported that use of PSC at 1.5 per cent and above significantly ($P < 0.05$) reduced PL after eight weeks of refrigerated storage. The present results agree with Webster *et al.* (1982), who reported that the PSC is able to increase WHC in processed products.

5.4.2. TBARS value

At 0-4°C, in AP and VP, the LFBB with CG either in combination with other FR or alone showed significantly ($P < 0.05$) lower TBARS values. The remaining treatments showed significantly ($P < 0.05$) higher TBARS values as period of storage increased from 0 - 10 days, irrespective of the packaging system. The values were significantly ($P < 0.05$) lesser than that of FF control in which the TBARS values significantly ($P < 0.05$) increased with days of irrespective of the packaging and temperature of storage.

LFBB with CG stored at -20°C showed significant ($P < 0.05$) decrease in TBARS values in both AP and VP from d zero to d 30. Burgers with CG alone as FR recorded the lowest value. Among the combinations of FR, CG-PSC showed significantly ($P < 0.05$) the lowest value of 0.228 mg/kg on d 30 under AP. In FF burgers the TBARS values significantly ($P < 0.05$) increased from d zero to d 20 in AP and the burgers spoiled on d 30. While in VP, although there was significant ($P < 0.05$) increase in the TBARS values, it could be stored up to 30 days without spoilage. The TBARS in other formulations in AP and VP significantly ($P < 0.05$) increased with the period of storage.

The results indicate that addition of CG as FR either alone in blends with TS and PSC in LFBB could reduce the development of TBARS and oxidative rancidity till d 10 on storage at 0-4°C and d 30 at -20°C under AP and VP. VP did not have any additional effect over AP of increasing the keeping quality of LFBB more than 10 days at 0-4°C and 30 days at -20°C.

The results are in agreement with Nazeera (2007) who also revealed that there was no significant ($P < 0.05$) difference between the aerobically and vacuum packaged low fat restructured turkey loaves during storage. On the contrary, some research workers observed lower TBARS values for vacuum packaged samples compared to aerobically packaged sample (Lynch *et al.* 1986; Ahn *et al.* 1992; Ho *et al.* 1995; and Nam and Ahn, 2001).

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. Moreover, the low fat content and the presence of onion in the formulary would have synergistically acted with CG in reducing the TBARS. Younathan *et al.* (1980) also reported the presence of numerous flavones in onion making them effective antioxidants.

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

5.4.3.1. Appearance and Colour

On day zero, the appearance and colour score of LFBB with CG-TS-PSC and PSC as FR was very good and same as that of BB with FF. All other LFBB were good except those with TS which was fair only. But on storage at 0-4°C under AP and VP none of the formulations showed significant ($P < 0.05$) difference in the appearance and colour of burgers on d 10. But at -20°C, the LFBB with TS under AP showed significant ($P < 0.05$) decrease in appearance and colour score from 5.87 to 5.62 on d zero to d 30, respectively may be due to increased brown colour as a result of caramelisation. All other formulations were not significantly ($P > 0.05$) different irrespective of the period of storage and types of packaging.

The results indicate that LFBB with blends of CG, TS and PSC as FR would be very good as FF beef burger with 20 per cent fat. In AP and VP LFBB retained the appearance and colour on storage at 0-4°C for 10 days and at -20°C for 30 days. The uniform brown colour of the cooked burger may be due to the Maillard reaction.

Arganosa *et al.* (1986) also reported similar result of uniform brown colour on the surface of cooked patties. They found no difference in colour in low fat pork sausage with collagen on storage.

5.4.3.2. Flavour

On the day of preparation the flavour of the LFBB with CG-TS and CG-TS-PSC were very intense, same as that of FF burger. Other formulations were moderately intense only. AP and VP did not affect the flavour score of the BB stored at 0-4°C irrespective of the days of storage from d zero to d 10. The burger with CG-TS, CG-PSC and TS-PSC, among combinations of FR, stored at -20°C and packaged under AP and VP showed significant ($P < 0.05$) decrease in flavour score on d 30. The other treatments did not show any significant ($P < 0.05$) difference on storage up to 30 days. The result shows that the LFBB with CG-TS-PSC maintained the flavour in both packaging systems up to 10 days at 0-4°C and at -20°C for 30 days. LFBB without FR was only moderately intense in flavour.

The results are in agreement with that of Huffman and Egbert (1990) who reported that there was no difference in beef flavour intensity over a range of 5 – 20 per cent in fat. Khalil (2000) observed that flavour intensity score were not affected replacing fat with water/starch.

5.4.3.3. Texture

On zero day, texture of LFBB with CG-TS-PSC was very desirable and same as that of FF. The remaining formulation scored moderately desirable texture only. There was no significant ($P > 0.05$) difference in the texture among all formulations of BB stored at 0-4°C under AP and VP on d10. LF burger on d zero was moderately desirable and after storage for 30 days it deteriorated to slightly desirable in VP. The results indicate that LFBB with blends of CG, TS and PSC particularly CG-TS-PSC was very desirable and had a keeping quality of 10 days at 0-4°C and 30 days at -20°C under AP and VP. LFBB without addition of FR was the toughest among all.

Brewer *et al.* (1992) reported that the burger with mixture of CG and TS had a rubbery texture similar to FF in LFBB. In low fat meat balls addition of CG

improved the texture (Ulu, 2004). Allen *et al.* (1999) reported that addition of TS in LFBB resulted in more succulent and tender product.

5.4.3.4. Saltiness

The burgers with CG-TS-PSC scored significantly ($P < 0.05$) very desirable score for saltiness while other formulations were moderately desirable only on the day of preparation. There was no significant ($P > 0.05$) difference in saltiness among all formulations of BB stored at 0-4°C under AP and VP on d10. Even during storage at -20°C under AP and VP the saltiness remained moderately desirable up to d 30 in LFBB. The method of packaging and period of storage did not affect saltiness score of the BB either at 0-4°C and -20°C.

5.4.3.5. Juiciness

The burgers with CG-TS-PSC scored significantly ($P < 0.05$) higher score of very juicy as in the case of FF on d zero and on storage. LFBB without FR scored the lowest value and was less juicy. There was no significant ($P > 0.05$) difference in the juiciness among all formulations of BB stored at 0-4°C and -20°C under AP on d10 and in VP on d 10, 20 and 30. AP and VP and the period of storage did not affect the juiciness of LFBB on storage at 0-4°C and -20°C and therefore, could be stored up to d 10 and d 30, respectively.

Similar results are obtained by Desmond *et al.* (1998) and reported that most of the low fat formulations had similar levels of juiciness to the FF control indicating that a number of ingredients retained the appropriate amount of moisture to ensure a juicy product.

5.4.3.6. Mouth Coating

The LFBB with FR and FF control were not significantly ($P > 0.05$) different and scored practically nil mouth coating. The type of packaging and period of storage did not enhance mouth coating.

5.4.3.7. Overall Acceptability

There was no significant ($P > 0.05$) difference in the overall acceptability among all formulations of AP and VP burgers stored at $0-4^{\circ}\text{C}$ on d 10 and at -20°C on d 10, 20 and 30.

The scores for all the seven sensory attributes for organoleptic evaluation of CG-TS-PSC were 7 and above while in the case of FF only six traits scored 7. The BB with CG-TS had only three traits with a score of 7 and above. All other formulations were with a score between 6 -7 for all the traits.

The results indicate that the overall acceptability of the LFBB in the decreasing order of choice by the Taste Panelists was CG-TS-PSC, FF, CG-TS, CG-PSC, TS-PSC, PSC, CG, TS and LF without any FR. The very acceptable nature of CG-TS-PSC formulation might be due to its synergistic effect of fat replacers CG, TS and PSC. Similarly, among single fat replacers LFBB with PSC scored higher overall acceptability.

Similar results were 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 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

The moisture content in the LFBB with CG-TS-PSC, CG-TS, TS-PSC and TS did not show any significant ($P < 0.05$) difference as the period of storage increased from d zero to d 10 both AP and VP at $0-4^{\circ}\text{C}$.

At -20°C under AP, the burger with CG-TS-PSC, CG-TS, CG-PS, TS-PSC, PSC and TS did not show any significant ($P < 0.05$) difference as the period of storage increased on d zero to d 30. Similarly, in the case of VP the LFBB with

FR except the PSC showed no significantly ($P < 0.05$) difference. But addition of PSC showed significantly ($P < 0.05$) increase in moisture content. Similarly Nazeera (2007) reported the same decreasing trend in the moisture content of low fat restructured turkey loaves during its storage and Selvakumar (2009) in low fat frankfurter.

The LFBB with FR either alone or in blends in both AP and VP could be stored up to d 10 at 0-4°C and up to d 30 at -20°C without significant ($P < 0.05$) reduction in protein content.

The beef burgers showed no significant ($P < 0.05$) difference in fat content during the entire period of storage at 0-4°C from d zero to d 10 in AP and VP and also at -20°C from d zero to d 30.

The results indicate that the LFBB with FR packaged either aerobically or vacuum can be stored at 0-4°C for 10 days and at -20°C for 30 days without significant decrease in proximate composition.

The LFBB with CG, TS, PSC and their blends as FR are developed economically with very acceptable overall acceptability, cook yield, nutritional quality, reduced purge loss and oxidative rancidity and shelf life up to 10 days at 0-4°C and 30 days at -20°C under AP and VP. The best LFBB with overall acceptability was CG-TS-PSC followed by CG-TS, CG-PSC, TS-PSC, PSC, CG and TS. Blends of FR are better than single FR, particularly CG-TS-PSC, as they increase CY, FRP, MRP, WHC, sensory attributes and reducing the pH, CL, DS, WBSF, PL and TBARS. The cost of production was calculated at laboratory level only and further investigations with production of large quantity are required for commercial production.

Summary

SUMMARY

Many researchers proved that high fat intake is associated with increased risk for obesity, colon cancer, high blood cholesterol, non-insulin diabetes and coronary heart disease. Therefore, the health conscious meat consumers prefer low fat meat products. Manufacturing meat products with fat replacers (FR) enable to reduce fat and to alleviate the problems with the reduction of fat in products. Therefore, the present study was undertaken with the objectives of developing a palatable and economic formulary for low fat beef burger (LFBB) with carrageenan (CG), tapioca starch (TS), pregelatinised pork skin collagen (PSC) and their blends as fat replacers (FR) and to assess its physico-chemical parameters, cooking characteristics, proximate composition, nutritional value, textural and organoleptic qualities and shelf life under aerobic (AP) and vacuum packaging (VP) at 0-4°C and -20°C and its cost of production.

Beef burgers (BB) are formulated at two different fat levels, *viz.*, full fat (FF) 20 per cent and low fat (LF) 5 per cent as controls. Seven formulations of LFBB with 5 per cent fat are prepared with 0.5 per cent CG, 1.5 per cent TS, 2 per cent PSC and their blends, *viz.*, CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC as FR.

Beef burgers (BB) are prepared as per the formularies by mixing appropriate amount of minced lean beef trimmings with tallow, salt, black pepper, onion, garlic, ginger, green chilli, rusk, ice flakes and fat replacers. The chilled burger mix was manually moulded and packaged aerobically in HDPE and in vacuum in polyethylene-polyamide (PEPA) pouches. The packaged BB are stored at 0 – 4°C and at -20°C for further studies.

Various physico-chemical parameters, *viz.*, pH, cook yield (CY), cook loss (CL), fat retention percentage (FRP), moisture retention percentage (MRP),

dimensional shrinkage (DS), water holding capacity (WHC), Warner-Bratzler Shear Force (WBSF), Hunter L^* , a^* , b^* colour values, proximate and mineral composition and nutritional value, purge loss (PL), Thiobarbituric Acid Reactive Substances (TBARS) value and sensory qualities are assessed on d 0, 10, 20 and 30 of storage at 0-4°C and -20°C or till spoilage, whichever is earlier. Six trials of the experiment were conducted.

pH of cooked burger in all formulations were significantly ($P < 0.05$) higher ($P < 0.05$) than their corresponding uncooked burgers and ranged from 6.18 in FF to 6.42 in PSC. Cooking significantly ($P < 0.05$) reduced the acidity of all the burgers. However, by the addition of CG, TS and PSC as FR a very low acid LFBB could be prepared compared to FF and LF controls.

CY of burgers with CG-TS and CG-TS-PSC were 86.47 and 85.84 per cent, respectively which were significantly ($P < 0.05$) the highest among all formulations. LFBB with combinations of FR showed significantly ($P < 0.05$) higher CY than those with single FR and correspondingly lesser CL. The percentage CL in FF and LF controls were significantly ($P < 0.05$) higher than in other formulations.

The DS in LFBB with CG-PSC and CG-TS-PSC was significantly ($P < 0.05$) the lowest with 13.09 and 13.21 per cent, respectively. The DS during the cooking process may be due to the denaturation of the meat proteins with loss of water and fat which could be reduced with the addition of blends of CG, TS and PSC at the level of 0.5, 1.5 and 2 per cent, respectively in LFBB.

FRP in CG-TS-PSC formulation was significantly ($P < 0.05$) the highest with 97.66 per cent followed by CG-PSC, TS-PSC, CG and CG-TS, TS. On the contrary, BB with FF had the lowest FRP of 90.80 per cent. Similarly, MRP was significantly ($P < 0.05$) higher in CG-TS-PSC and CG-PSC with 74.36 and 74.41

per cent, respectively. In order to increase FRP and MRP in LFBB, it is ideal to incorporate blends of CG, TS and PSC compared to single FR.

The WHC of LFBB with CG-TS-PSC was 95.36 per cent equivalent to that of FF and the burgers were more succulent and juicy. Burgers with blends of FR showed significantly ($P < 0.05$) higher WHC compared to single FR. Starch favors formations of stronger heat induced structure through swelling of starch granules embedded in protein gel matrix which increased water binding. CG can improve the water retention by the formation of water-protein complex. PSC improves protein functionality and WHC through the immobilization of free water.

The LFBB with CG-TS-PSC was most succulent and tender with WBSF value 5.30 N equivalent to FF, which was 5.35 N. But those without any FR were the toughest with WBSF 6.73 N. Among the LFBB with blends of FR, in the decreasing order of tenderness were, CG-TS, CG-PSC and TS-PSC. PSC significantly ($P < 0.05$) reduced the WBSF in LFBB. The water binding and gelling ability of collagen improved the texture of LF formulations by diluting the myofibrillar proteins. Fat has an inverse relationship with WBSF as fat is less resistant to shear force compared to hard proteinaceous matrix.

Cooked LFBB with blends of FR, especially CG-TS-PSC was lighter, less reddish (more bluish) and less yellowish (more greenish) and is comparable to FF BB. Replacement of lean meat with PSC reduced the amount of myoglobin responsible for the red colour which resulted in light red BB. Cooking decreased the redness of products which may be due to the denaturation of the myoglobin.

Fat content in the beef trimmings and PSC were < 1.76 per cent. Burger with CG-TS-PSC had significantly ($P < 0.05$) the highest mineral content, especially Na. Highest content of K was in CG-TS, Ca in CG and P in TS. On comparison of the proximate and mineral composition of the uncooked and

cooked BB, significant ($P < 0.05$) reduction in the moisture content and a corresponding increase in the protein, fat, carbohydrate, ash, Na, K, Ca and P content were noticed. The addition of FR significantly ($P < 0.05$) increased the protein and mineral content and BB with CG-TS-PSC was more acceptable.

The RDA of total calorific value 11.68 per cent in FF could be reduced to the range of 6.36 to 7.18 per cent in LFBB, with FR. The contribution of fat to RDA of calorific value ranged from 2.22 to 2.42 only. The contribution of calories from fat to the RDA was below the recommended 30 per cent in all formulations.

The LFBB with TS-PSC had significantly ($P < 0.05$) higher percentage of 35.63 daily value of protein among blends of FR, while in burgers with PSC alone was 35.98 per cent. More than one third of the daily requirement of protein is obtained from 100g of LFBB. The burger with CG-TS-PSC contained significantly ($P < 0.05$) the highest Na and P among all formulations, which were 39.42 and 27.0 per cent, respectively. K content in LFBB ranged from 13.74 to 17.68 per cent. LFBB with CG had the significantly ($P < 0.05$) highest Ca content of 5.27 per cent. LFBB with FR are good sources of Na, K and P but not of Ca.

At 0-4°C, under AP and VP the BB could be stored up to d 10 only while at -20°C up to d 30. But FF control could be stored only up to d 20 in AP without spoilage at -20°C. The PL in LFBB with CG-TS-PSC and CG-TS was significantly ($P < 0.05$) the lowest at 0-4°C in AP and VP. Blends of FR were more efficient in reducing PL. Lowering of fat from 20 to 5 per cent in BB without FR will lead to significant ($P < 0.05$) increase in PL in AP and VP and on storage at 0-4°C and -20°C. The CG, PSC and TS could retain considerable portion of added water in LFBB and thereby reduced the PL on storage.

The addition of CG either alone in blends with TS and PSC in LFBB could significantly ($P < 0.05$) reduce the development of TBARS and oxidative rancidity till d 10 on storage at 0-4°C and d 30 at -20°C under AP and VP. VP did not have

any additional effect over AP of increasing the keeping quality of LFBB more than 10 days at 0-4°C and 30 days at -20°C. TBARS values determined were lower than the acceptable range of 1mg malonaldehyde/kg for oxidative rancidity. Moreover, the low fat content and the presence of onion in the formulary would have synergistically acted with CG in reducing the TBARS.

On sensory evaluation on d zero, the LFBB with CG-TS-PSC scored significantly ($P < 0.05$) higher values of 7.00 and above for very good appearance and colour, very intense flavour, very desirable texture, juiciness, practically nil mouth coating and very acceptable overall acceptability similar to FF burger. But saltiness was very desirable than in FF. LFBB without FR was the toughest, least juicy and with lowest flavour among all. The uniform brown colour of the cooked burger may be due to the Maillard reaction.

In AP and VP, LFBB with CG-TS-PSC retained all the sensory attributes and proximate composition even on storage at 0-4°C for 10 days and at -20°C for 30 days. The overall acceptability of the LFBB in the decreasing order of choice was CG-TS-PSC, FF, CG-TS, CG-PSC, TS-PSC, PSC, CG, TS and LF without any FR. The very acceptable nature of CG-TS-PSC formulation might be due to the synergistic effect of fat replacers.

The LFBB with 5 per cent fat and CG (0.5%), TS (1.5%), PSC (2%) and their blends as FR are developed economically with very acceptable overall acceptability, CY, nutritional quality, reduced PL and oxidative rancidity and shelf life up to 10 days at 0-4°C and 30 days at -20°C under AP and VP. The best LFBB with overall acceptability was CG-TS-PSC followed by CG-TS, CG-PSC, TS-PSC, PSC, CG and TS. Blends of FR are better than single FR, particularly CG-TS-PSC, as they increase CY, FRP, MRP, WHC, sensory attributes and decrease pH, CL, DS, WBSF, PL and TBARS. The cost of production of LFBB with CG-TS-PSC was about Rs.111/= per kg.

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FORMULATION OF LOW FAT BEEFBURGER WITH FAT REPLACERS

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ABSTRACT

Health conscious meat consumers prefer low fat meat products due to increasing incidents of high fat induced diseases. Manufacturing meat products with fat replacers (FR) enable to reduce fat and to alleviate the problems with the reduction of fat in products. Therefore, the present study was undertaken with the objectives of developing a palatable and economic formulary for low fat beef burger (LFBB) with carrageenan (CG), tapioca starch (TS), pregelatinised pork skin collagen (PSC) and their blends as FR and to assess its pH, cooking characteristics, proximate composition, nutritional value, textural and organoleptic qualities and shelf life under aerobic (AP) and vacuum packaging (VP) at 0-4°C and -20°C and its cost of production. Beef burgers (BB) are formulated at two different fat levels, *viz.*, full fat (FF) 20 per cent and low fat (LF) 5 per cent as controls. Seven formulations of LFBB with 5 per cent fat are prepared with 0.5 per cent CG, 1.5 per cent TS, 2 per cent PSC and their blends, *viz.*, CG-TS - 0.5% CG & 1.5% TS; CG-PSC - 0.5% CG & 2.0% PSC; TS-PSC - 1.5% TS & 2.0% PSC; CG-TS-PSC - 0.5% CG, 1.5% TS & 2.0% PSC as FR.

BB are prepared as per the formularies with minced lean beef trimmings, tallow, salt, spices and condiments, rusk, ice flakes and FR. They are packaged aerobically in HDPE and in vacuum in polyethylene-polyamide (PEPA) pouches. pH, cook yield (CY), cook loss (CL), fat retention percentage (FRP), moisture retention percentage (MRP), dimensional shrinkage (DS), water holding capacity (WHC), Warner-Bratzler Shear Force (WBSF), Hunter L*, a*, b* colour values, proximate and mineral composition and nutritional value, purge loss (PL), Thiobarbituric Acid Reactive Substances (TBARS) value and sensory qualities are assessed on d 0, 10, 20 and 30 of storage at 0-4°C and -20°C or till spoilage, whichever is earlier. Six trials of the experiment were conducted.

Cooking reduced the acidity of all the burgers. By the addition of FR a significantly ($P < 0.05$) very low acid cooked LFBB could be prepared. CY of

burgers with CG-TS-PSC was significantly ($P < 0.05$) the highest with 85.84 per cent. LFBB with blends of FR significantly ($P < 0.05$) increased CY and correspondingly reduced CL. The DS in LFBB with CG-TS-PSC was significantly ($P < 0.05$) the lowest with 13.21 per cent. Addition of blends of FR holds water and fat in LFBB and reduces DS during cooking. FRP and MRP in CG-TS-PSC formulation was significantly ($P < 0.05$) the highest with 97.66 and 74.36 per cent, respectively due to blends of CG, TS and PSC.

The WHC of LFBB with CG-TS-PSC was 95.36 per cent and WBSF value 5.30 N comparable to FF and the burgers were significantly ($P < 0.05$) most succulent, juicy and tender with the addition of blends of FR compared to tougher BB without FR. According to Hunter L^* , a^* , b^* values, LFBB with blends of FR, especially CG-TS-PSC was lighter, less reddish (more bluish) and less yellowish (more greenish) and comparable to FF burger.

Fat content in the beef trimmings and PSC were < 1.76 per cent. Cooking significantly ($P < 0.05$) reduced moisture content with a corresponding increase in the protein, fat, carbohydrate and ash. The percentage total calorific value of LFBB ranged from 6.36 to 7.18 of the Recommended Dietary Allowance (RDA). The contribution of fat to RDA of calorific value was from 2.22 to 2.42 per cent only, which was below the recommended 30 per cent. More than one third of the daily requirement of protein is obtained from 100g of LFBB. LFBB with FR are good sources of Na, K and P but not of Ca.

Blends of FR in LFBB, especially CG-TS-PSC, were more efficient in significantly ($P < 0.05$) reducing PL and TBARS value on storage at $0-4^{\circ}\text{C}$ for 10 days and at -20°C for 30 days in AP and VP. TBARS values were lower than the acceptable range of 1mg malonaldehyde/kg for oxidative rancidity. The low fat content and the presence of onion containing antioxidants in the formulary would have synergistically acted with CG in reducing the TBARS.

On sensory evaluation on zero day, the LFBB with CG-TS-PSC scored significantly higher ($P < 0.05$) values of 7.00 and above for very good appearance and colour, very intense flavour, very desirable texture, juiciness, practically nil mouth coating and very acceptable overall acceptability similar to FF burger. But saltiness was very desirable than in FF. The LFBB with CG-TS-PSC in AP and VP retained all the sensory attributes and proximate composition even on storage. The very acceptable nature of CG-TS-PSC formulation might be due to the synergistic effect of fat replacers.

The LFBB with 5 per cent fat and CG (0.5%), TS (1.5%), PSC (2%) and their blends as FR are developed economically with very acceptable overall acceptability, CY, nutritional quality, reduced PL and oxidative rancidity and shelf life up to 10 days at $0-4^{\circ}\text{C}$ and 30 days at -20°C under AP and VP. The best LFBB with overall acceptability was CG-TS-PSC followed by CG-TS, CG-PSC, TS-PSC, PSC, CG and TS. Blends of FR are better than single FR, particularly CG-TS-PSC, as they increased CY, FRP, MRP, WHC, sensory attributes and decreased pH, CL, DS, WBSF, PL and TBARS. Further investigations with production of large quantities are required for calculation of cost of production at commercial scale.

