EFFECTS OF PROCESSING PARAMETERS ON SOME QUALITY ATTRIBUTES OF YOGHURT FROM MILK OF WEST AFRICAN DWARF GOAT

BY

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A dissertation submitted to the Food Processing and Value Addition Programme, Center of Excellence in Agricultural Development and Sustainable Environment (CEADESE), Federal University of Agriculture, Abeokuta, Nigeria, in partial fulfillment of the requirement for the degree of Master in Agricultural Development and Sustainable Environment.

FEDERAL UNIVERSITY OF AGRICULTURE, ABEOKUTA

JUNE, 2018

DECLARATION

I hereby declare that this Thesis was written by me and is a correct record of my own research work. It has not been presented in any previous application for any degree of this or any other University. All citations and sources of information are clearly acknowledged by means of references.

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CERTIFICATION

We certify that this Thesis entitled "Effect of Processing Parameters on Some Quality Attributes of Yoghurt from Milk of West African Dwarf Goat" is the outcome of the research carried out by I.I. Popoola in the Department of Food Processing and Value Addition, World Bank Africa Centre of Excellence in Agricultural Development and Sustainable Environment, Federal University of Agriculture, Abeokuta.

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ABSTRACT

Pasteurization temperature, incubation temperature and time are some of the important processing parameters for the development of fermented dairy products with desired qualities. This study investigated the effects of processing parameters on physico-chemical and microbiological qualities of West African Dwarf (WAD) goat milk yoghurt and also compared some sensory qualities and acceptability of the optimized milk yoghurt from goat and cow milk yoghurt as control. Response surface methodology (RSM) based on Box Behnken design was used to optimize the processing parameters; pasteurization temperature (PT; 80 - 85 °C), incubation temperature (IT; 40 - 45 °C) and incubation time (ITm; 2.5 - 4.5 h), while pH, titrable acidity, total solids, fat, protein, viscosity, total plate, fungal, lactic acid bacteria, and coliform count were determined using standard laboratory procedures. Aroma, taste and mouth-feel were monitored for the sensory qualities. The acceptability of the products were evaluated using thirty untrained panelists. Data were analyzed using quadratic polynomial models and analysis of variance. Numerical optimization technique was used to obtain the optimum processing parameters for WAD goat milk yoghurt. The values for pH, titrable acidity, total solid, viscosity, fat, and protein content of goat milk yoghurt were in the range 4.35 - 5.97, 0.57 - 3.70%, 13.54 - 32. 64 mg/L, 130158 - 272712 mm^2/s, 4.00 - 10.33% and 3.24 - 28.44% respectively. The total plate, fungal and lactic acid bacteria counts ranged from 5.0×10^4 to 3.5×10^5 cfu/ml, 1.0×10^4 to 2.4×10^5 cfu/ml and 2.0×10^4 to 5.50×10^6 cfu/ml respectively with no growth detected for coliform counts. Sensory assessment for the yoghurts showed that optimized WAD goat yoghurt had a sensory rating of 6.07 to 6.37 while cow yoghurt had a rating of 7.73 - 8.20. Significant ($p<0.05$) differences were observed among the optimized goat and cow milk yoghurts. The coefficient of determination (R^2) of the quadratic models ranged between 0.60 and 0.97 while F-value was from 1.15 to 24.89. Also, pH was significantly

($p<0.05$) affected by IT and ITm while titrable acidity was significantly ($p<0.05$) affected by PT and ITm. PT significantly ($p<0.05$) affected the total solids. Viscosity and protein were significantly ($p<0.05$) affected by PT, IT and ITm (quadratic term), while fat was significantly ($p<0.05$) affected by IT and ITm (quadratic term) as well as PT and IT (interaction term). Total plate and fungal counts were significantly ($p<0.05$) affected by interaction of PT, IT and ITm in addition to the quadratic term of PT and IT. In conclusion, the optimum processing parameters for WAD goat yoghurt was found to be PT of 84.24 ^{o}C, IT of 44.22 ^{o}C and ITm of 3.8 h with optimized WAD goat milk yoghurt had a higher mean value for its protein content than cow milk yoghurt.

DEDICATION

This dissertation is dedicated to Almighty God, my tower of strength, my source of inspiration, wisdom, knowledge and understanding. I also dedicate this work to my son; Enoch (my love-bug) who has been affected in every way possible by this quest. To my wonderful parents and siblings they have encouraged me all the way and their encouragement has motivated me to give it all it takes to finish that which I have started. Lastly, I dedicate this work to my husband; Oluwaseyi Afuwape. My love for you all can never be quantified. God bless you.

ACKNOWLEDGEMENTS

My gratitude goes to God for provision to complete this project and the program, throughout this entire study he took control of everything that would have stopped me in my tracks and strengthened me even through my most difficult times.

I am highly indebted to the World Bank of Africa, for providing this platform for career development and also for their support in funds. My sincere appreciation to the Prof. O.M. Onagbesan (Director of the programme) for his efforts and advice to achieve success, also to my major supervisor Prof. (Mrs) J.M. Babajide whose contribution and constructive criticism has assisted me to achieve the best in this work. I am grateful to her for making me experience true research and my knowledge on the subject matter has been broadened. I am grateful to my co-supervisors, Dr. O.P. Sobukola and Prof. B.O. Oluwatosin they have impacted me during this work and their contributions have assisted in the success of the work.

I also appreciate the efforts of Dr Dike, Engr. Omotade and Mrs O.O Idowu of Federal Institute of Industrial Research, Oshodi who assisted me with the required information, ideas, knowledge, apparatus and contact links for my work.

My extreme regards goes to my lovely parents, Engr. Olakunle O.O. and Mrs Ibitola I.S. who painstakingly laid the foundation for my education giving it all it takes, and also for their support morally and financially. I am grateful to my mother- in- love, Elder Mrs Cecilia Afuwape for her prayers and words of encouragement.

I am and will forever be grateful to my loving husband Mr Oluwaseyi Afuwape who has given everything possible to ensure I achieve this feat. I also from the depth of my heart appreciate my son (Le Boo-boo) who even at such tender age has had to endure so much stress and discomfort just for me. This page can’t tell it all. I appreciate my siblings Abiola

(My prayer partner and laundry assistant), Tolani (My chef, nanny and assistant all rolled into one), Esther (baby sitter), you are all wonderful and am glad you are part of me.

I appreciate all my colleagues, friends and well-wishers. Opeyemi Faloye special thanks for your support all through the data analysis and interpretation of this work. Many thanks to Adesina Gbemiro who truly inspired me.

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CHAPTER ONE

1 INTRODUCTION

The process required for the production of yoghurt is an ancient art that has been in existence some thousands of years ago this could be as result of the domestication of cow, sheep, or goat, but before the nineteenth century the process involved in yoghurt making can accurately be presumed to be little understood. In spite of the current principle of industrial technology, the process of yoghurt making is still a complex process which combines both art and science together. The yoghurt process survived through the ages as a result of its production in small scale and the art was handed down from parents to children (Tamime and Robinson, 1999).

An important part of human diet in many regions of the world in ancient times is fermented dairy foods which have been consumed ever since the domestication of animals. Yoghurt is a product made from heat treated milk that may be homogenized prior to the addition of lactic acid bacteria (LAB) cultures containing *Lactobacillus bulgaricus and Streptococcus thermophilus* (Code of Federal Regulations Section 131.203, 2011). Yoghurt can also be defined as a product of the lactic acid fermentation of milk by addition of a starter culture, which results in a decrease of milk pH to less than or equal to 4.6 (Tamime, 2002). The conversion of lactose to lactic acid has preservative effect on milk; moreover, the low pH of cultured milk inhibits the growth of putrefactive bacteria and other determined organisms, thereby, prolonging the shelf life of the products (Elagamy *et al.,* 1992). An advantage of fermentation of milk of various domesticated animals is the production of products in which their essential nutrients are conserved that otherwise would deteriorate rapidly

under the high ambient temperatures. Thus, the process permitted consumption of milk constituents over a period significantly longer than was possible for milk itself.

Goats are widely populated in all types of ecology with more concentrated in the tropics and dry zones of most developing countries. Small ruminants provide a source of profitable income in small farm system and agriculture and an area of market specialization (Devendra, 2001).

Goat milk is a distinctive dairy resource, which is well known as “the king of milk” it is easily digested and has a rich nutrition (Tamime and Robinson, 2000; Agnihotri and Prasad, 1993). Goat milk is more completely and easily absorbed than cow's milk, leaving less undigested residue behind in the colon to quite literally ferment and cause the uncomfortable symptoms of lactose intolerance (Haenlein, 1992).

There are several process parameters that influence flavour, body, and texture of yoghurt such as the starter culture, incubation temperature, processing conditions (e.g., heat treatment, homogenization) and compositional properties of the milk base (Labropoulos *et al*., 1984; Tamime and Robinson, 1999; Shaker *et al*., 2001; Hassan *et al*., 2003). One of the most important processing parameters that affects the texture and consistency of yoghurt is pasteurization of milk (Mulvihill and Grufferty, 1995).

1.1 Justification

Haenlein (2004) reported that the use of goat milk as an excellent food source is undeniable. It has beneficial effects for health maintenance, physiological functions, in the nutrition of children and elderly people, can be consumed without negative effects by people suffering cow milk allergy.

Goat milk and its product e.g. yoghurt has three-fold significance in human nutrition: (1) feeding more starving and malnourished people in the developing world than from cow milk (2) treating people afflicted with cow milk allergies and (3) filling the gastronomic requirements of connoisseur consumers which correspond to a growing marker in many developed countries. (Haenlein, 2004).

Monounsaturated (MUFA), polyunsaturated fatty acids (PUFA), and medium chain triglycerides (MCT), which all are known to be beneficial for human health, especially for cardiovascular conditions, in goat milk is greater than that of cow milk. This biomedical superiority has not been promoted much in marketing goat milk, and goat yoghurt, but has great potential in justifying the uniqueness of goat milk in human nutrition and medicine (Babayan, 1981; Haenlein, 1992).

Yoghurt derived from the milk of species other than bovine tends to vary in several sensory and physico-chemical characteristics, due to differences per milk composition. For instance, yoghurt derived from milk with high fat content (e.g. sheep, goat, and buffalo) has a more creamy texture compared to that derived from milk with lower fat content (e.g. bovine, mare, and ass). Therefore, the species of the milk-producing mammal significantly influence the characteristics of the produced yoghurt (Tamime and Robinson, 2007).

Urban consumers believe that goat dairy products have a good ecological image, and goat milk and dairy products are not rich in fat, are more digestible, are healthy for many gastrointestinal illnesses, and are less allergenic than cow milk. Consequently, goat milk and goat dairy products have real future economic potentials (Morand-Fehr *et al*., 2004; FAOSTAT, 2009; Orman *et al*., 2011).

In the last decade, there has been an increased interest for goat milk production and its conversion to value added products as well as a renewed interest in goat milk as an alternative milk source for people with cow milk intolerance (Tziboula-Clarke, 2003; Albenzo *et al*., 2006).

1.2 Objective

The aim of the study is to evaluate the effect of processing parameters on some quality attributes (sensory, chemical and microbiological attributes) of yoghurt from milk of West African Dwarf Goat in order to obtain optimum process parameters.

1.2.1 Specific objectives

The specific objectives are to:

1. Determine the effect of pasteurization, incubation temperature and time on some chemical and microbiological attributes of yoghurt made from West African Dwarf (WAD) goat milk;
2. Optimize the processing parameters of WAD goat milk yoghurt; and
3. Compare some sensory quality and acceptability of the optimized WAD goat milk yoghurt with the control (cow milk yoghurt).

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Goat Milk

Goat milk and its products have played an important role in the economic viability in many parts of the world, especially in developing countries. A variety of manufactured products can be produced from goat milk, including fluid products (low fat, fortified, or flavored), fermented products such as cheese, yoghurt or buttermilk, frozen products such as ice cream or frozen yoghurt, butter, and condensed and powdered products (Park, 2011). According to Haenlein and Abdellatif (2004), the world production of goat milk has been relatively minor compared to that of bovine milk (2.1% versus 84.6% of the total milk production, respectively), the worldwide goat population has reached 758 million heads with 55% increase during the last 20 years, and goat milk production has reached 12.2 million tones with 58% increase during the same period.

Producing high quality raw milk is of utmost importance for successful production and marketing of dairy goat products. The products must be safe to consume and free of pathogenic bacteria, antibiotics, insecticides, and herbicides. They should have a good taste with no objectionable flavor or odor, be free of spoilage from bacteria, and contain legal limits of all nutrients (Park, 2011).

Goat milk exhibits beneficial virtues for individuals with certain dietetic problems, thus it is recommended traditional by physicians for infant and others allergic to cow milk. Similarly it has been used in treatment of ulcers (Mereado, 1982, Kumar *et al*., 2012).

Milks of different species of mammals have been used for the production of yoghurt, as a result, variations in the quality of yoghurt do occur, depending on the type of milk used, for example, milk containing a high percentage of fat (sheep, buffalo and reindeer) produces a rich and creamy yoghurt with an excellent “mouthfeel” compared with yoghurt manufactured from milk containing a low level of fat, or milk deprived of its fat content, for example skimmed milk. The lactose in milk provides the energy source for the yoghurt starter organisms, but the protein plays an important role in the formation of the coagulum and hence the consistency/viscosity of the product is directly proportional to the level of protein present; yoghurt produced from unfortified mare’s and ass’s milk would be less viscous than yoghurt made from sheep’s or reindeer’s milk. Although the flavour of yoghurt is mainly the result of complex biochemical reactions initiated by microbial activity, the flavour of the milk base varies from species to species and this characteristic is reflected in the end product (Tamime *et al*., 2000).

2.2 Production of Quality Goat Milk

Fresh goat milk is a white, opaque liquid with a slightly sweet taste and no odour. Milk drawn from the lacteal glands is highly perishable. It is adversely affected by improper practices of feeding, handling of animals and milk before, during and after milking; and by its cooling, transportation, pasteurization, processing method, packaging, and processing equipment (Park, 2011).

High-quality, pasteurized goat milk must contain no pathogens or foreign substances, such as antibiotics, antiseptics, or pesticide residues. It is similar in taste and odour to quality cow's milk. Pasteurization and protection from sunlight or UV light control oxidized and “goaty” flavors. Goaty flavour is attributable to caproic, caprylic, and

capric acids, which are present at high levels in goat milk fat and subject to release from fat globule membranes by lipases if improper milking and processing are practiced (Park, 2011).

2.3 Composition of Goats' Milk

Milk of various domesticated animals differs in composition and produces fermented milk with a characteristic texture and flavour (Table 1). Goat milk differs from cow or human milk in having better digestibility, alkalinity, buffering capacity and certain therapeutic values in medicine and human nutrition (Park, 2007; Haenlein, 1984).

In relation to other types of milk, goat milk presents advantages such as smaller size fat globules, low allergenic properties (Martín-Diana *et al*., 2003), a balance of essential amino acids, high levels of calcium, selenium, phosphate and rich in vitamins A and B. Goat's milk has a similarity to human milk that is unmatched in cow milk and also has several medicinal values.

Therefore awareness about advantage of consumption of goats milk should be popularized so that production and utilization of goat's milk could be enhanced (Kumar *et al*., 2012). Goat milk is superior to milk of other mammals due to better fat and protein digestibility and assimilation; to its significantly higher minerals and vitamins composition and to incidence of allergy is lower (Bielak 1993; and Dostalova, 1994, Belewu and Aiyegbusi, 2002). However, goat milk is deficient in folic acid and vitamin D. Goat's milk fat contains more vitamin A than cow's milk. The fatty acid composition of goat's milk is also different, being richer in volatile fatty acids (caproic, caprylic, and capric) that are responsible for the specific taste and odour of the respective dairy products. The higher content of medium-chain fatty acids accounts also for the more prolonged bacteriostatic stage (Boycheva *et al*., 2011).

Table 1: Proximate Composition of Milk of Mammals used for Fermented Milks

	%Total Solids	%Fat	%Total Protein	%Casein	% Whey Protein	% Lactose	%Ash
Cow	12.2	3.4	3.4	2.8	0.6	4.7	0.7
Cow, Zebu	13.8	4.6	3.3	2.6	0.7	4.4	0.7
Buffalo	16.3	6.7	4.5	3.6	0.9	4.5	0.8
Goat	13.2	4.5	2.9	2.5	0.4	4.1	0.8
Sheep	19.3	7.3	5.5	4.6	0.9	4.8	1.0
Camel	13.6	4.5	3.6	2.7	0.9	5.0	0.7
Mare	11.2	1.9	2.5	1.3	1.2	6.2	0.5
Donkey	8.5	0.6	1.4	0.7	0.7	6.1	0.4
Yak	17.3	6.5	5.8	-	-	4.6	0.9

Chandan and Shahani (1993), Chandan (2002)

2.3.1 Some factors affecting the composition of goat's milk

Composition of goat's milk widely differs according to many various factors. The following effective factor, breeds (indigenous or selected breeds), stage of lactation, feeding or rations components affect the composition of goat milk.

a) Breed Influence

Milk yield and composition is affected by Origin and type of breed. There are two types of goat milk, the first (which is the more common) is produced from indigenous breeds which have a low average milk yield but have a high total solid. The second type is produced by highly selected breeds with high yield but with a lower total solid (Akinsoyinu *et al*., 1977, El Zayat *et al*., 1984, Kalantzopoulos, 1993).

b) Stage of Lactation

Stage of goat lactation is markedly affected the resultant milk either yield or composition. Brown *et al.* (1995) stated that relative amount of αs2-CN decreased with stage of lactation, also relative amount of K-CN increased by 50% after peak lactation and its concentration almost doubled near the end of lactation. Kracmar *et al.* (1998) studied the change in amino acids composition of goat's milk during lactation period from 5^{th} to 33^{rd} days in White Short Wooled goats, and concluded that:

(a) Decrease in non-essential amino acids was ranged from 0.39 to 10.05

(b) Decrease in essential amino acids was ranged from 0.79 to 41.6%

(c) Threonine and Iso-leucine was decreased sharply

(d) All other amino acids widely decreased.

Bhosale *et al*. (2009) indicated that lactation had significant increasing effect on fat, protein, ash, total solid, solid not fat, titrable acidity and viscosity. All milk components are gradually increased from I to IV lactation with exception of lactose and pH.

c) Feeding Ration

Feed ration is one of the main factors that affects milk composition as it is the source of milk constituents, and controls the fermentation process in rumen. Kholif and Abou-El-Nor (1998) studied the effect of replacing corn with powder date seeds in diets of Baladi lactating goat's on their productive performance during the 1st week of lactation. Kholif and Abou-El-Nor (1998) reported that fat, total solid, total protein as well as total saturated, short and medium chain fatty acids contents tended to be higher, while lactose content and C15, C16 total unsaturated fatty acids were decreased. Morsy *et al*. (2012) concluded that supplementing Anise oil, Clove oil or Juniper oil for lactating goats improve rumen fermentation as propionate production and reduce acetate proportion and improved milk protein of lactating goats. Juniper oil supplementation improved conjugated linoleic and omega 3 fatty acids in milk fat. Juniper oil supplementation to dairy animals can contribute to improve the health properties of milk.

2.4 General Information on Yoghurt

2.4.1 Fermentation process

Fermentation is one of the oldest methods practiced by human beings for the transformation of milk into products with an extended shelf life. The exact origin(s) of the making of fermented milks is difficult to establish, but it could date from some 10 – 15000 years ago as the way of life of human beings changed from being food gatherer to food producer (Pederson, 1979),

Concomitantly, conversion of milk to fermented milks resulted in the generation of a distinctive viscous consistency, smooth texture, and unmistakable flavour. Furthermore, fermentation provided food safety, portability, and novelty for the

consumer. Accordingly, fermented dairy foods evolved into the cultural and dietary ethos of the people residing in the regions of the world to which they owe their origin.

2.4.2 Definition and classification

Yoghurt is a semisolid fermented milk product made by the symbiotic activity of a blend of *Streptococcus salivarius subsp. Thermophilus* and *Lactobacillus delbrueckii subsp. Bulgaricus* and can include other lactic acid bacteria. According to the International Dairy Federation definition for fermented milk, it is a milk product fermented by the action of specific microorganisms and resulting in reduction of pH and coagulation. These specific micro-organisms shall be viable, active and abundant (at least 10^7 cfu/g) in the product to the date of minimum durability" (Ouwenhand and Salminen, 1999).

Yoghurt is made from a mix standardized from whole, partially defatted milk, condensed skim milk, cream, and nonfat dry milk. Supplementation of milk solids non -fat (SNF) of the mix with non-fat dry milk is frequently practiced in the industry. The FDA specification calls for a minimum of 8.25% non - fat milk solids. However, the industry uses up to 12% SNF or non-fat milk solids in the yoghurt mix to generate a thick, custard-like consistency in the product.

The milk fat levels are standardized to 3.25% for full fat yoghurt. Reduced fat yoghurt is made from mix containing 2.08% milk fat. Low fat yoghurt is manufactured from mix containing 1.11% milk fat. Non-fat yoghurt mix has milk fat level not exceeding 0.5%. These fat levels correspond to the Food and Drug Administration requirement for nutritional labeling of non-fat, reduced fat, and low fat yoghurt (Chandan, 1997).

All dairy raw materials should be selected for high bacteriological quality. Ingredients containing mastitis milk and rancid milk should be avoided. Also, milk partially

fermented by contaminating organisms and milk containing antibiotic and sanitizing chemical residues cannot be used for yogurt production.

Yoghurts can be classified industrially into two types. A set-style yoghurt which is made in retail containers giving a continuous undisturbed gel structure in the final product (Tamime and Robinson, 1999). On the other hand, stirred yogurt has a delicate protein gel structure that develops during fermentation (Benezech and Maingonnat, 1994). In stirred yoghurt manufacture, the gel is disrupted by stirring before mixing with fruit and then it is packaged. Stirred yoghurts should have a smooth and viscous texture (Tamime and Robinson, 1999). In terms of rheology, stirred yoghurt is a viscoelastic and pseudo plastic product (De Lorenzi *et al*., 1995).

Yoghurt come in a variety of textures (e.g. liquid, set, and smooth), fat contents (e.g. luxury, low-liquid, virtually fat-free) and flavours (e.g. natural, fruit, cereal), can be consumed as a snack or part of a meal, as a sweet or savory food, and are available all year round. This versatility, together with their acceptance as a healthy and nutritious food, has led to their widespread popularity across all population sub-groups (McKinley, 2005).

A number of changes can be noticed in the casein micelle, with the increased acidity. As the pH falls amorphous calcium is released but the as1-casein skeleton is retained. This disaggregation is followed by a subsequent aggregation, initiated by the b-casein once the pH has fallen sufficiently for the two main casein species to carry opposite charges (Varnam and Sutherland, 1994, Banon and Hardy, 1991). Generally, the overall qualities of yoghurt,which includes acidity level,free fatty acid production,production of aroma compounds (di acetyle,acetaldehyde and acetoin) as

well as sensory qualities and nutritional values are important attributes of the product (Lee and Lucey, 2010).

2.4.3 Manufacture of yoghurt

The main ingredient in the manufacture of yoghurt is milk. The type of milk used depends on the type of yoghurt – whole milk for full fat yoghurt, low-fat milk for low-fat yoghurt, and skim milk for non-fat yoghurt. To ensure a high quality end-product, the milk should have a low bacterial count (i.e. maximum of 1.0×10^5 colony-forming-units (cfu g^{-1}). Furthermore, the milk and other dairy ingredients should be free from taints, antibiotic compounds, sanitizing agents and bacteriophages; somatic count should be $< 4.0 \times 10^5$ cells mL^{-1} (optimum $\leq 2.5 \times 10^5$ cells mL^{-1}) (Tamime and Robinson, 1999; Oliveira *et. al*., 2002).

Other dairy ingredients are allowed in yoghurt to adjust the composition, such as cream to adjust the fat content, and nonfat dry milk to adjust the solids content. The solids content of yoghurt is often adjusted above the 8.25% minimum to provide a better body and texture to the finished yoghurt. Stabilizers may also be used in yoghurt to improve the body and texture by increasing firmness, preventing separation of the whey (syneresis), and helping to keep the fruit uniformly mixed in the yogurt. Stabilizers used in yoghurt are alginates (carrageenan), gelatins, gums (locust bean, guar), pectins, and starch. Sweeteners, flavours and fruit preparations are used in yoghurt to provide variety to the consumer. Codex regulations for yoghurt indicate that the minimum milk protein content is 2.7% (except for concentrated yoghurt where the minimum protein content is 5.6% after concentration) and the maximum fat content is 15% (Codex Standard for Fermented Milk, 2008). The flow chart for the production of yoghurt is shown in Figure 1.

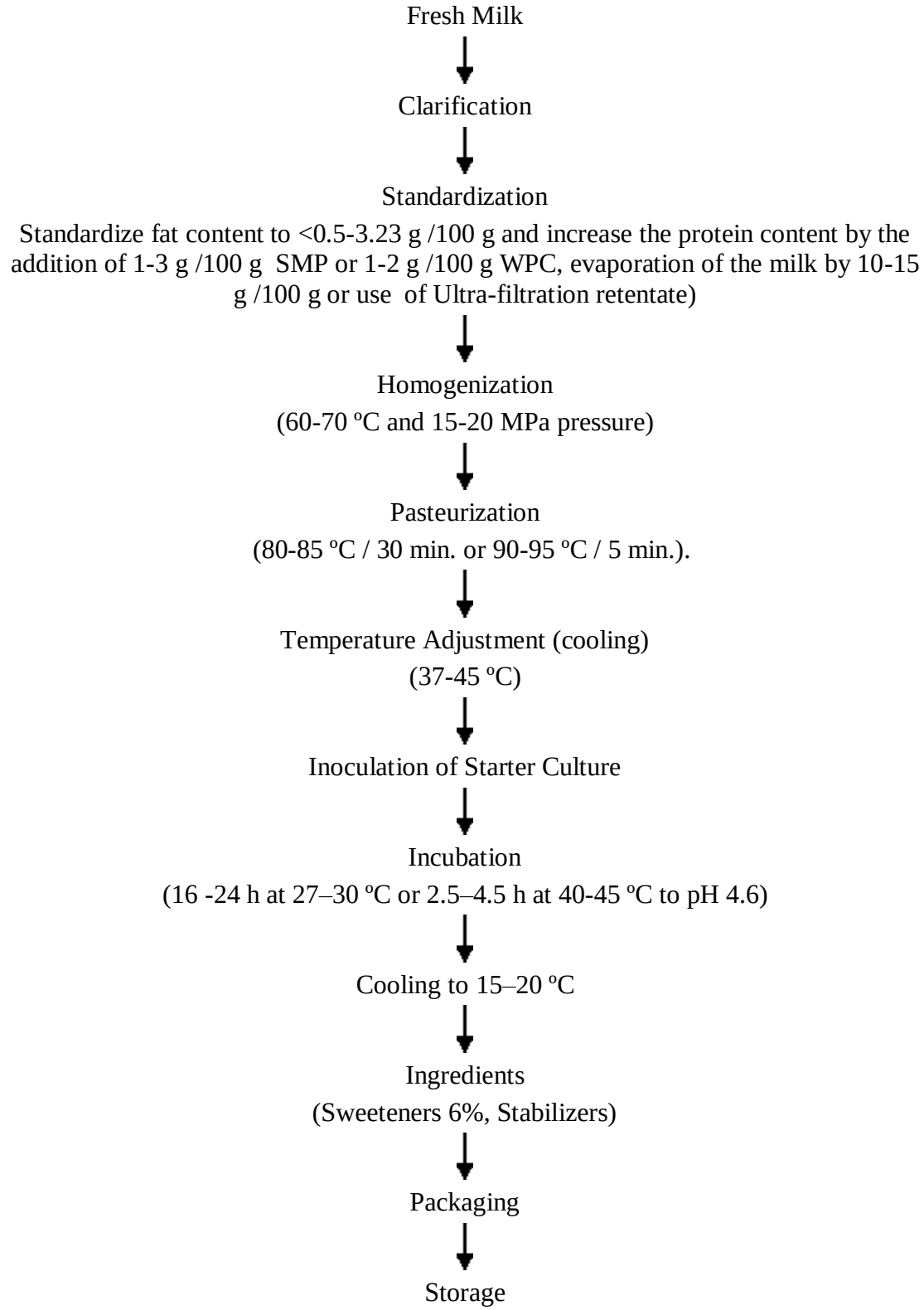


Figure 1: Flow Chart for Yoghurt Production Modified Method of Tamime and Robinson (1999).

2.4.4 Yoghurt starter culture

Spontaneous souring of milk yields uncontrollable flavor and texture characteristics with food safety concerns. Modern industrial processes utilize defined lactic acid bacteria as a starter for yogurt production. A starter consists of food grade microorganism(s) that on culturing in milk predictably produce the attributes that characterize yogurt (Chandan, 2004).

The main (starter) cultures in yoghurt are *Lactobacillus delbrueckii* subsp. *bulgaricus* (LB) and *Streptococcus thermophilus* (ST). Both *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* are fairly compatible and grow symbiotically in milk medium (Chandan, 2004).

Lactobacillus acidophilus is commonly added as additional culture to commercial yogurt. Other cultures added belong to various *Lactobacillus* and *Bifidobacterium* species. However, the optional organisms do not necessarily exhibit compatibility with LB and ST. Judicious selection of strains of LB, ST, and the optional organisms is necessary to ensure the survival and growth of all the component organisms of the starter. Nevertheless, product characteristics, especially flavour, may be slightly altered when yoghurt culture is supplemented with optional bacteria (Chandan, 2004).

The supplementation of fermented products with probiotic bacteria becomes beneficial by providing better use of the lactose, anti-carcinogenic activity and intestinal infection control. Probiotics are referred to as “live microorganisms, which when administered in adequate amounts confer a health benefit on the host” (FAO/WHO, 2001; Allgeyer *et al*., 2010). Strains of *L. acidophilus* and of *Bifidobacterium lactis* predominate in commercial probiotic products (Tabasco *et al*., 2007).

The function of the starter cultures is to ferment lactose (milk sugar) to produce lactic acid. The increase in lactic acid decreases pH and causes the milk to clot, or form the soft gel that is characteristic of yoghurt. The fermentation of lactose also produces the flavour compounds that are characteristic of yoghurt.

Commercial production of yoghurt relies heavily on the fermentation ability of and the characteristics imparted by the starter. Satisfactory starter performance requires rapid acid development; development of typical yoghurt flavour, body, and texture; exopolysaccharide secreting strains to enhance the viscosity of the yoghurt; scale-up possibilities in various production conditions, including compatibility with the variety and levels of ingredients used and with fermentation times and temperatures; survival of culture viability during the shelf life of the yoghurt; probiotic properties and survival in the human gastrointestinal tract for certain health attributes; and minimum acid production during distribution and storage at 4 – 10 °C until yoghurt is consumed (Chandan, 2004).

The activity of a starter culture is determined by direct microscopic counts of culture slides stained with methylene blue. This exercise also indicates physiological state of the culture cells. Cells of *Streptococcus thermophilus* grown fresh in milk or broth display pairs or long chains of spherical, coccal shape. Under stress conditions of nutrition and age (old cells, cells exposed to excessive acid, colonies on solid media, milk containing inhibitor), the cells appear oblong in straight chains that resemble rods (Chandan, 2004).

Acid-producing ability is measured by pH drop and titrable acidity rise in 12% reconstituted nonfat dry milk medium (sterilized at 116 °C for 18 min.) incubated at 40 °C for 8 hours. A ratio of *Streptococcus thermophilus* to *Lactobacillus delbrueckii*

subsp. *bulgaricus* of 3:1 gives a pH of 4.20 and titrable acidity of 1.05% under the above conditions (Chandan, 2004).

The influence of temperatures of incubation on the growth of yoghurt bacteria is shown in Table 2. Acid production is normally used as a measure of growth of a yoghurt culture. However, growth of the organisms is not necessarily synonymous with their acid-producing ability. Differences in acid liberated per unit cell mass, which are related to both environmental effects and genetic origin, have been recorded (Chandan, 2004).

Yoghurt fermentation constitutes the most important step in its manufacture. To optimize parameters for yoghurt production and to maintain both a uniformity of product quality and cost effectiveness in the manufacturing operation, an understanding of the factors involved in the growth of yoghurt bacteria is important (Chandan, 2004).

Table 2: Growth Temperature P rofile of Yoghurt Bacteria

Growth Temperature	*Streptococcus thermophilus*	*Lactobacillus delbrueckii Subsp. Bulgaricus*
	^{o}C	^{o}C
Minimum	20	>15
Maximum	50	50-52
Optimum	39-46	40-47

Chandan and Shahani (1993)

2.5 Methods of Production and Classification

The methods of production of yoghurt have in essence changed little over the years and although there have been some refinements, especially in relation to lactic acid bacteria, that bring about fermentation, the essential steps in the process are still the same, namely:

• Raising the level of total solids in the process milk to around 14 – 16g 100 g^{-1}.

• Heating the milk, ideally by some method that allows the milk to be held at high temperature for a period of 5 – 30 mins; the precise time will depend on the temperature selected.

• Inoculating the milk with a bacterial culture in which *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* are the dominant organisms.

• Incubating the inoculated milk, in bulk or retail units, under conditions that promote the formation of a smooth viscous coagulum and the desired aromatic flavour /aroma.

• Cooling and, if desired, further processing, e.g. the addition of mixture of fruit and other ingredients, pasteurization or concentration.

• Packaging for distribution to the consumer under chilled conditions.

Variations in milk composition, irregular behavior of the starter organisms, faulty regulation of the incubation temperature, along with a number of other process variables, can all give rise to an end product that is deficient in respect of overall quality, and only a thorough understanding of the fermentation can provide an operative with the foresight to reduce the risk of product failure.

2.6 Factors Affecting the Physical and Sensory Properties of Yoghurts

It is well established that the way the milk is handled or prepared; including the processing conditions used in yoghurt manufacture, greatly influence the gel texture, strength and stability (Lucey and Singh, 1997; Walstra, 1998; Tamime and Robinson, 1999; Jaros and Rohm, 2003a, b), and could be briefly summarized as:

• Fortification level and material(s) used in the mix;

• Stabilizer type and usage levels;

• Fat content and homogenization conditions;

• Milk heat treatment conditions;

• Starter culture (type, rate of acid development and production of exopolysaccharides – EPS);

• Incubation temperature (influences growth of starter cultures, gel aggregation, bond strength);

• pH at breaking of the gel (stirred) and/or start of cooling (set);

• Cooling conditions;

• Post-manufacture handling of the product, e.g. physical abuse (vibration) and temperature fluctuations (i.e. if the product is not maintained at ≤ 5 °C).

2.6.1 Dry matter fortification

The physical and sensory properties of yoghurt gels are greatly influenced by the total solids content of the yoghurt milk, especially the protein content. The G′ values of yoghurt increases with an increase in the total solids content obtained by the addition of

skim milk powder or by ultra-filtration (Biliaderis *et al*., 1992). Increased yoghurt viscosity is observed when the total solids content of milk is increased (Guirguis *et al*., 1984; Becker and Puhan, 1989; Wacher-Rodarte *et al*., 1993). The oral viscosity of yoghurt or perceived thickness also increases with an increase in total solids content of milk (Skriver *et al*., 1999; Sodini *et al*., 2004). The increased solids content in yoghurt milk as a result of fortification also creates increased buffering that requires additional acid development by the starter cultures to achieve a similar pH target (Lee and Lucey, 2010). Most yoghurt products are sweetened (not plain). The use of sucrose increases the total solids of the mix and strengthens the gel network. A range of sweeteners are used commercially, especially for low calorie products. Another option is to use β-galactosidase to hydrolyse lactose as the products are glucose and galactose, which are much sweeter than lactose (Lee and Lucey, 2010).

2.6.2 Heat treatment

Native whey proteins from unheated milk are inert fillers in yoghurt (Lucey *et al*., 1999). When milk is heated at >70 °C, the major whey proteins, such as, β-lacto globulin, are denatured. During denaturation β-lacto globulin interacts with the κ-casein on the casein micelle surface (and any soluble κ-casein molecules, i.e. κ-casein that dissociates from the micelle at high temperatures) by disulfide bridging, which results in increased gel firmness and viscosity of yoghurt (Dannenberg and Kessler, 1988; Lucey *et al*., 1997). Denatured whey proteins that have become attached to the surface of casein micelles are a critical factor involved in the increased stiffness of yoghurt gels made from heated milk (Lucey *et al*., 1998).

Heat treatment of milk for 15 min at ≥80 °C results in significantly increased denaturation of β-lacto globulin compared with milk heated at 75 °C for a similar time

(Lucey *et al*., 1997). The extent of denaturation of whey proteins during the heat treatment of milk affects the firmness and viscosity of acid milk gels (Dannenberg and Kessler, 1988). High heat treatment of milk causes a shift in gelation pH towards higher pH values, Lucey *et al*. (1998) suggested that this shift was due to the higher isoelectric pH (~5.3) of β-lacto globulin, which is the main whey protein.

Yoghurt mix is pasteurized (80 to 85 ℃ for 30 min. or 90 to 95 ℃ for 10 min.) to destroy pathogens but as temperature/time exceeds pasteurization minimums (63 ℃ for 30 min. or 72 ℃ for 15 s) (CFR 1240.61), other desirable outcomes occur–for instance, inactivation of some non-pathogenic microorganisms, production of stimulatory/inhibitory factors for starter cultures, inactivation of enzymes and alterations to the physicochemical properties of milk constituents (Tamime and Robison, 1999).

The heat treatment of milk prior to package for liquid consumption, or manufactured into milk based product, is an important critical control point to ensure that pathogenic organisms are killed. It also ensures spoilage organisms are eliminated, or at least reduced in a number, for optimum keeping quality (IDF, 1994).

2.6.3 Fermentation

After the heat treatment stage, the milk will be cooled to 42 – 43 ℃ ready for the addition of the starter culture consisting of an equal mixture of *Lb. delbrueckii* subsp. *bulgaricus a*nd *S. thermophilus*. How the culture is added to the milk will depend on its physical form, for a liquid culture prepared in the dairy, the bulk culture will be held in tanks, and then pumped into the process milk at an addition rate of 2.0 mL 100 mL^{-1}; the addition rate for concentrated freeze-dried or frozen cultures purchased for direct inoculation into the process vat is set by the culture supplier. However, the need

to avoid contamination of the milk with undesirable bacteria, yeasts and moulds during inoculation is universal, and a number of systems have been developed to achieve this aim (Tamime, 2002). Once the milk has been inoculated, it will be filled into cartons for incubation as set yoghurt or it will be fermented in a bulk tank (stirred yoghurt). Although 42 °C is the typical fermentation temperature for yoghurt, using slightly lower incubation temperatures (e.g. 40 °C rather than 45 °C) will lead to slightly longer gelation times, but firmer and more viscous gels are formed that are less prone to whey syneresis or lumpy/grainy defects on stirring (Robinson, 1981; Lucey, 2002; Lee and Lucey, 2003). At a lower incubation temperature, there is an increase in the size of the casein particles due to a reduction in hydrophobic interactions, which, in turn, leads to an increased contact area between the casein particles (Lee and Lucey, 2003); a similar trend occurs when gels are cooled. A high incubation temperature also makes the gel network more prone to rearrangements, and these changes can lead to greater whey separation (Lucey, 2001; Mellema *et al*., 2002).

The result of the microbial activity of the starter culture is that the acidity of the milk will have risen to around 1.0 – 1.2 g 100 mL^{-1} lactic acid (around pH 4.2 – 4.3) after 3 – 4 h. At this acidity the milk proteins will have coagulated to form a firm gel (Lucey and Singh, 1997, 2003).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Source of Materials

Fresh West African Dwarf Goat milk was collected from Federal University of Agriculture Abeokuta, research farm, while the fresh cow milk was purchased from local famers in Abeokuta. The West African Dwarf Goat were managed semi intensively, they were fed in the morning and allowed to scavenge for the rest of the day. Milk samples were then kept in an ice box immediately after collection. A commercial starter culture *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* (Yogourmet; Freeze- dried yoghurt starter) was used in the production of the experimental yoghurt runs.

3.2 Experimental Design

Response Surface Method- Box Behnken Design - Expert version 6.0.8 was used to generate the experimental design, as shown in Table 3. The fresh goat and cow milk obtained was clarified using a clean muslin cloth to remove dirt, debris, and udder tissues, the raw milks were then subjected to physical, chemical, and microbiological analyses. The West African Dwarf goat milk was processed based on experimental runs shown in Table 4 to produce yoghurt, 250 ml of the raw milk was used for each treatments. The effect of pasteurization, incubation temperature and time on quality attributes of yoghurt from West African Dwarf goat milk was then determined; some of the sensory, chemical and microbiological quality parameters of the yoghurt produced were evaluated. A total of 17 experiments / treatments were carried on the goat milk.

Table 3: Process Variables for West African Dwarf Goat Milk Yoghurt

Process Variables	Units	-1	0	+1
Pasteurization Temp.	°C	80	82.5	85
Incubation Temp.	°C	40	42.5	45
Incubation Time	h	2.5	3.5	4.5

Source: Response Surface Method- Box Behnken Design - Expert version 6.0.8

Table 4: Experimental Design

Runs	Pasteurization Temp. (ºC)	Incubation Temp. (ºC)	Time (h)
1	80	40	3.5
2	80	42.5	2.5
3	80	42.5	4.5
4	80	45	3.5
5	82.5	40	2.5
6	82.5	40	4.5
7	82.5	42.5	3.5
8	82.5	42.5	3.5
9	82.5	42.5	3.5
10	82.5	42.5	3.5
11	82.5	42.5	3.5
12	82.5	45	2.5
13	82.5	45	4.5
14	85	40	3.5
15	85	42.5	2.5
16	85	42.5	4.5
17	85	45	3.5

Source: Response Surface Method- Box Behnken Design - Expert version 6.0.8

3.3 Yoghurt Production

3.3.1 Yoghurt production for physico-chemical and microbiological analysis

Yoghurt was manufactured using the method outlined by Tamime and Robinson (1999) with some modifications (Fig 2). The goat milk obtained from West African Dwarf Goat was filtered with a clean muslin cloth to remove dirt, debris, and udder tissues. The clarified goat milk was then pasteurized in 3 batches; $WADGP_1$ - 80 ºC, $WADGP_2$ - 82.5 ºC, $WADGP_3$ - 85 ºC, respectively for 30 min. After which the pasteurized milk samples were cooled to inoculation temperature of 42 ºC ± 1 ºC and then inoculated with yoghurt starter culture (freeze-dried yoghurt starter) consisting of *Lactobacillus bulgaricus, Streptococcus thermophilus* and *Lactobacillus acidophilus*, the yoghurt was fermented as outlined in the experimental runs in Table 4. The plain yoghurt was then packaged in polyethylene terephthalate bottles, chilled in a refrigerator and presented for chemical, microbiological evaluation.

where,

$WADGP_1$ - West African Dwarf Goat Pasteurization 1st batch

$WADGP_2$ - West African Dwarf Goat Pasteurization 2nd batch

$WADGP_3$ - West African Dwarf Goat Pasteurization 3rd batch

3.3.2 Yoghurt production for sensory acceptability test

WAD goat and cow milk were processed to yoghurt (Fig. 3 and 4) for sensory evaluation based on the optimization solution for the process parameters. Cow milk yoghurt was used as the control for the evaluation. The optimized solution for the

process parameters are pasteurization temperature (84.24 ^{o}C), incubation temperature (44.22 ^{o}C), and incubation time (3.8 h).

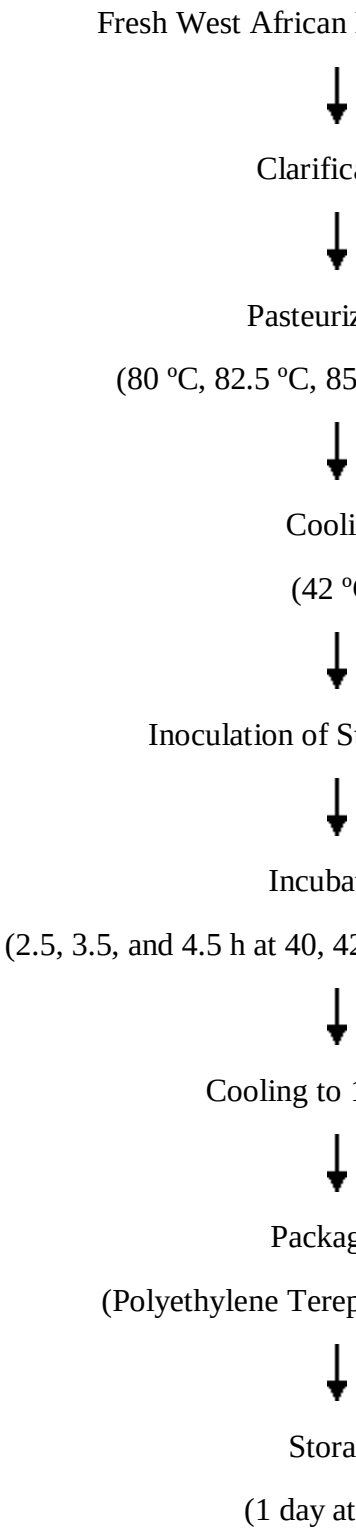


Figure 2: Flow Chart for WAD Goat Milk Plain Yoghurt Production (Tamime and Robinson, 1999)

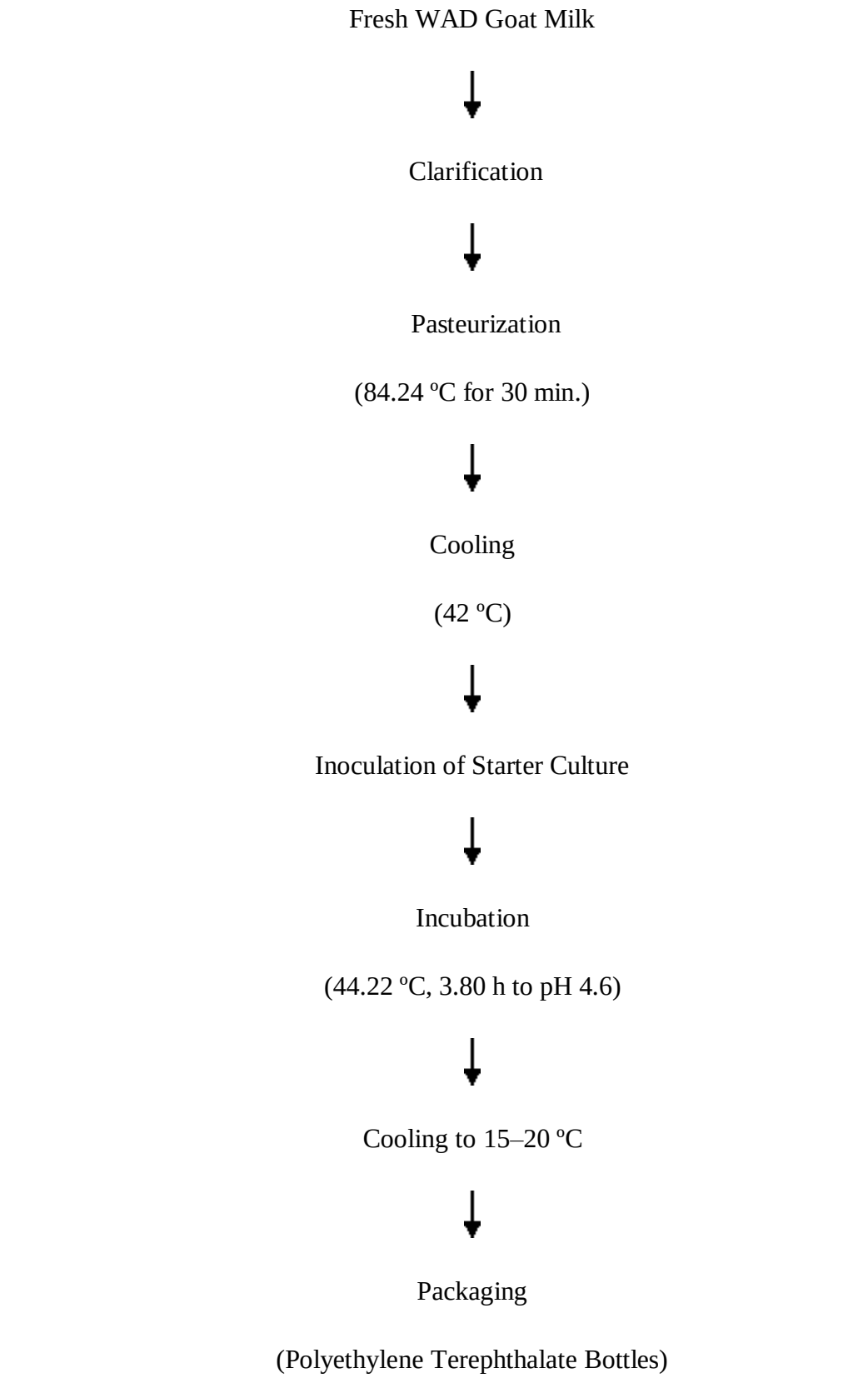


Figure 3: Flow Chart for WAD Goat Milk Plain Yoghurt Production for Sensory Evaluation

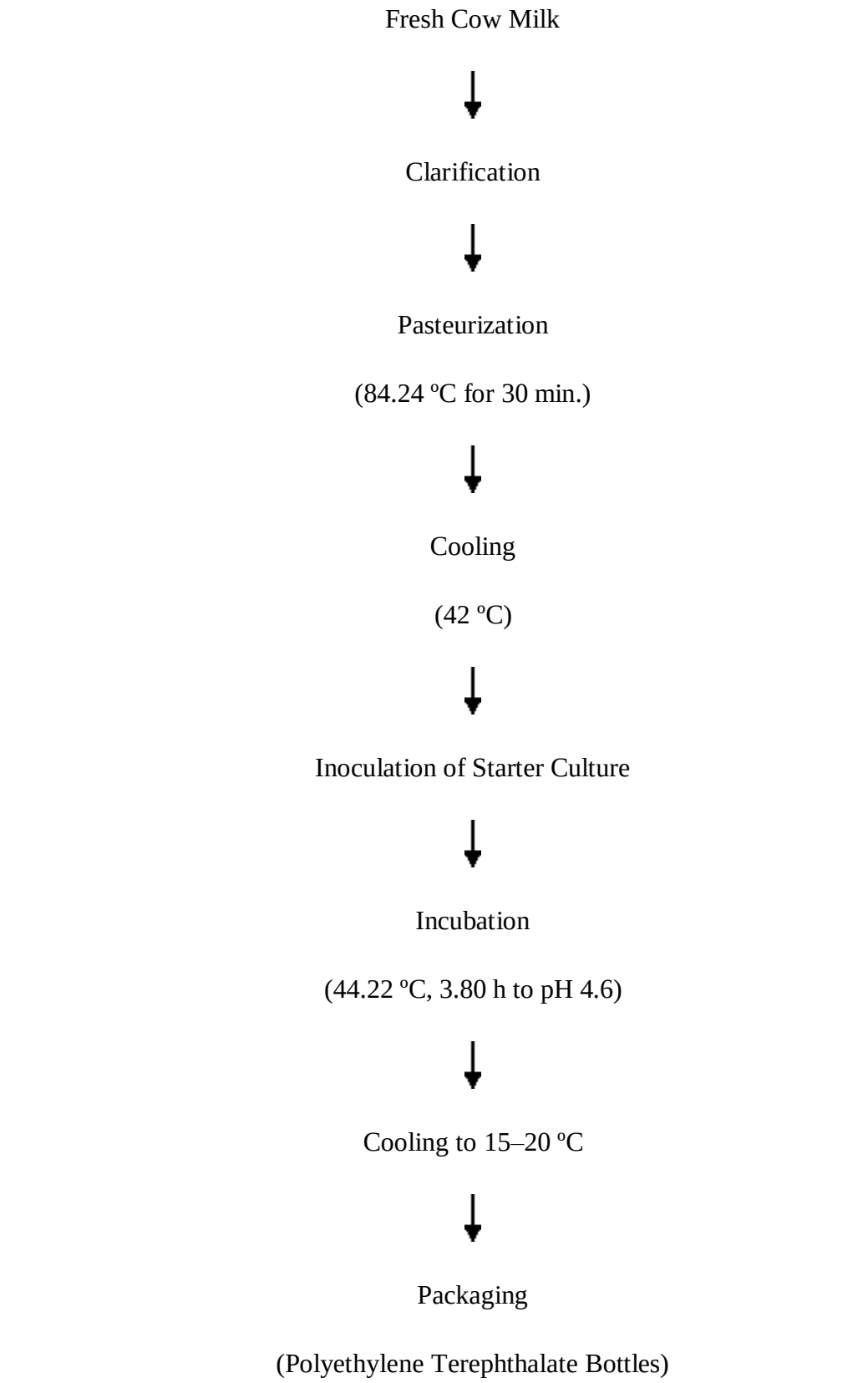


Figure 4: Flow Chart for Cow Milk Plain Yoghurt Production for Sensory Evaluation

3.4 Raw Goat Milk Analysis

The basic ingredient for the production of yoghurt is milk and hence the quality of the incoming milk is an important consideration. The raw milk was subjected to the following physical and chemical analyses which were carried out in triplicates.

3.4.1 Chemical analysis of raw goat milk

3.4.1.1 Fat content determination

The fat content of goat milk was determined using Acid Digestion Method of Fat determination in Milk (Werner Schmidt Method) as described by Bradley *et al.* (1992) as follows; in a clean dry Gerber tube, 10 ml of sulphuric acid (density 1.815 gm/ml at 20 °C) was poured, and then 10.94 ml of goat milk sample was added in the butyrometer. Amyl alcohol (1-2 ml) was added to the tube. The content was thoroughly mixed till no white particles could be seen. The Gerber tube was centrifuged at 1100 revolutions per minute (rpm) for 4-5 min at 65 °C. The fat column was then read immediately.

Calculation;

$$\text{Fat \%} \left({}^{W}/_{W}\right) = \frac{100(W_1 - W_2)}{W3} \quad \text{.....Equation 1}$$

where,

W_1= Weight in grams of contents in flask before removal of fat.

W_2= Weight in grams of contents in flask after removal of fat and

W_3= Weight in grams of material taken for the test. (10 g)

3.4.1.2 Protein content determination

Total protein in the goat milk was determined as described by the International Dairy Federation Method, IDF 20-1 (2001). Three grams of the goat milk was weighed and poured in digestion tube along with a digestion tablet and 20 mL of concentrated H_2SO_4. Digestion was done initially by slow heating for 45 min. to avoid frothing and then at 80 °C until appearance of clear or pale green colour. The digested sample was allowed to cool for half an hour. Then 100 ml distilled water was added and mixed gradually and transferred to 250 ml volumetric flask, and the digestion flask was rinsed 2-3 times with distilled water and the volume made up to 250 ml by adding distilled water.

Ten milliliters of the digested sample and 10 ml of NaOH was distilled in micro Kjeldahl apparatus. The ammonia produced was trapped in 4% boric acid solution containing few drops of methyl red indicator. With the addition of ammonia, boric acid color changed from red to yellow. The distillation was continued for 2 - 3 min. after first appearance of yellow color to catch maximum ammonia. The content was then titrated against 0.1 N H_2SO_4 solutions till pink colour end point appeared. The volume of H_2SO_4 used was noted.

Total nitrogen (%) was calculated with the following formula and the value obtained was multiplied with the factor in the equation below to get total protein:

$$\%\ Nittrogen = \frac{\text{Vol. of sulphuric acid used(ml)} \times 250 \times 0.0014}{\text{Vol. used for digestion} \times \text{Vol. of digested sample}} \times 100$$

$$\%\ Total\ Protein = \%\ Nitrogen \times 6.38 \quad \text{.....Equation 2}$$

3.4.1.3 Total solids determination

The Total Solids was determined as described by AOAC (2005). Five milliliter of the sample was weighed into a dry petri dish of a known weight. The total portion was pre-dried for 25 min. on steam bath and then dried for 3 h at 100 ºC in forced draft air oven. The Total Solid sample is the weight of the dried sample residue and was calculated as:

$$\%\ Total\ Solids\ =\ \frac{W2-W1}{W1-W}\times\ 100$$ .....Equation 3

where, W = Weight of the dish

W_1 = Weight of dish and sample test portion

W_2 = Weight of dish and dry sample

3.4.1.4 Determination of total titrable acidity (TTA)

This was determined using the titrimetric method as described by AOAC (2005). One (1) ml of phenolphthalein indicator was introduced into 10 ml of the mixed solution. It was then titrated against standard 0.1 N sodium hydroxide solution until pink colour persisted for about 10-15 seconds for complete neutralization. The titration figure is divided by 10 to get the percentage of lactic acid.

3.4.1.5 pH measurement

The pH of the raw milk was measured with a digital pH meter. pH buffers 4 and 7 was used for the calibration of the pH meter. After calibration, 20 ml of raw milk was taken

in a beaker and then electrode is immersed in the milk until constant reading attained (Ong *et al*., 2007).

3.4.1.6 Determination of viscosity

The viscosity of the sample was determined using the Ostwald viscometer, the sample was allowed to flow through its capillary tube between two etched marks and the time of flow of the liquid was measured (Abbas *et al*., 2010).

Then the viscosity was calculated as follows:

$$\eta = KPt$$ .....Equation 4

where ŋ = viscosity (mm^2/s)

K = constant

T = time (Sec)

P = hydrostatic pressure (mm^2)

3.5 Microbial Analysis of Raw Goat Milk

3.5.1 Preparation of serial dilutions

One millimeter of the raw milk was weighted using a micro pipette aseptically into a test tube containing 9 ml sterile distilled water (autoclaved at 121 °C for 15 min) Further serial dilutions were made by mixing one ml of the initial dilution with 9 ml sterile distill water until 1/10 dilution.

3.5.2 Total plate count

The total plate count of raw milk was determined as described by (Harrigan and MacCance, 1976). The colony count method to determine the total spores was followed. One millimeter from the dilution was aseptically transferred into sterile petri-dishes. Then to each plate nutrient milk agar was added. The inocula was mixed with the medium and allowed to solidify. The plates were then incubated at 37 ℃ for 24 – 48 h.

3.5.3 Fungal count

From suitable dilutions of sample, 1ml was aseptically transferred into Sabouraud Dextrose Agar (SDA) containing 0.1 g chloramphenicol per one liter to inhibit bacterial growth. The sample was spread all over the plates using sterile bent glass rod and then the plates was incubated at 28 ℃ for 48 h (Harrigan and MacCance, 1976).

3.5.4 Coliform count

Coliform bacteria was carried out on violet red bile agar medium and incubated for 24 hours at 37 ℃ for total coliforms and 44 ℃ for fecal coliforms according to the standard (ISO 4832); *E. coli* was streaked onto eosine methylene blue (EMB) agar and then incubated overnight at 37 ℃.

3.5.5 Methylene blue reduction time test

In the methylene blue reduction (MBRT) test 1 ml of methylene blue was added to 10 ml of raw milk. The tube was is sealed with rubber stopper and slowly inverted three times to mix. It was placed incubated at 37 ℃ and examined at intervals up to 6 h. The time taken for the methylene blue to become colorless is the methylene blue reduction time (MBRT) (Benson, 2002).

3.5.6 Alcohol test

Alcohol test was performed by mixing equal amounts of fresh milk and 75% alcohol, followed by detecting precipitation according to the method used by Widodo *et al.* (2013).

3.6 Chemical Analysis of Yoghurt

3.6.1 Fat content determination

The fat content of the yoghurt sample was determined using Acid Digestion Method of Fat determination in Milk (Werner Schmidt Method) method as described by Bradley *et al.* (1992) as follows: In a clean dry Gerber tube, 10 ml of sulphuric acid (density 1.815 gm/ml at 20 °C) was poured, and then 10.94 ml of sample was added in the butyrometer. Amyl alcohol (1-2 ml) was added to the tube. The content is thoroughly mixed till no white particles could be seen. The Gerber tube was centrifuged at 1100 revolutions per minute (rpm) for 4-5 min at 65 °C. The fat column was then read immediately.

Calculation;

$$\text{Fat } \% \left({}^{W}/_{W}\right) = \frac{100(W_1 - W_2)}{W3} \quad \text{.....Equation 5}$$

where,

W_1= Weight in grams of contents in flask before removal of fat.

W_2= Weight in grams of contents in flask after removal of fat and

W_3= Weight in grams of material taken for the test (10 g)

3.6.2 Total solids determination

The Total Solids was determined as described by AOAC (2005). Ten milliliter of the yoghurt sample was weighed into a dry petri dish of a known weight. The total portion was pre-dried for 25 min. on steam bath and then dried for 3 h at 100 ºC in forced draft air oven. The Total Solid sample is the weight of the dried sample residue and was calculated as:

$$\%\ Total\ Solids\ = \frac{W2 - W1}{W1 - W} \times 100$$ .....Equation 6

where, W = Weight of the dish

W_1 = Weight of dish and sample test portion

W_2 = Weight of dish and dry sample

3.6.3 Total titrable acidity (TTA) determination

This was determined using the titrimetric method as described by AOAC (2005). One (1) ml of phenolphthalein indicator was introduced into 10ml of the mixed solution. It was then titrated against standard 0.1N sodium hydroxide solution until pink colour persisted for about 10 - 15 seconds for complete neutralization. The titration figure was divided by 10 to get the percentage of lactic acid.

3.6.4 pH measurement

The pH of yoghurt was measured with digital pH meter. pH buffers 4 and 7 was used for the calibration of the pH meter. After calibration, 20 ml of yoghurt was taken in a beaker and then electrode is immersed in the milk until constant reading attained (Ong *et al*., 2007).

3.6.5 Protein content determination

Total protein in the yoghurt was determined as described by the international dairy federation method, IDF 20-1 (2001). Three grams of the sample was weighed and poured in digestion tube along with a digestion tablet and 20 ml of concentrated H_2SO_4. Digestion was done initially by slow heating for 45 min. to avoid frothing and then at 80 °C until appearance of clear or pale green colour. The digested sample was allowed to cool for half an hour. Then 100 ml distilled water was added and mixed gradually and transferred to 250 ml volumetric flask, and the digestion flask was rinsed 2 - 3 times with distilled water and the volume made up to 250 ml by adding distilled water.

Ten milliliters of the digested sample and 10 ml of NaOH were distilled in micro Kjeldahl apparatus. The ammonia produced was trapped in 4% boric acid solution containing few drops of methyl red indicator. With the addition of ammonia, boric acid color changed from red to yellow. The distillation was continued for 2 - 3 min. after first appearance of yellow color to catch maximum ammonia. The content was then titrated against 0.1 N H_2SO_4 solutions till pink colour end point appeared. The volume of H_2SO_4 used was noted.

Total nitrogen % was calculated with the following formula and the value obtained was multiplied with the factor in the equation to get total protein:

$$\%\ Nittrogen = \frac{\text{Vol. of sulphuric acid used(ml)} \times 250 \times 0.0014}{\text{Vol. used for digestion} \times \text{Vol. of digested sample}} \times 100$$

$$\%\ Total\ Protein = \%\ Nitrogen \times 6.38 \quad \text{.....Equation 7}$$

3.6.6 Determination of viscosity

The viscosity of the sample was determined using the Ostwald viscometer, the sample was allowed to flow through its capillary tube between two etched marks and the time of flow of the liquid was measured (Abbas *et al*., 2010).

Then the viscosity was calculated as follows:

$$\eta = KPt$$

……..Equation 8

Where ŋ = viscosity (mm^2/s)

K = constant

T = time (Secs)

P = hydrostatic pressure (mm^2)

3.7 Microbial Analyses of Yoghurt

3.7.1 Preparation of serial dilutions

One millimeter of the yoghurt sample was weighted using a micro pipette aseptically into a test tube containing 9 ml sterile distilled water (autoclaved at 121 ℃ for 15 min). Further serial dilutions were made by mixing one ml of the initial dilution with 9 ml sterile distill water until 1/10 dilution.

3.7.2 Total plate count

The total plate count of raw milk was determined as described by (Harrigan and Mac Cance, 1976). The colony count method to determine the total spores was followed. One millimeter from the dilution was aseptically transferred into sterile petri-dishes.

Then to each plate nutrient milk agar was added. The inocula was mixed with the medium and allowed to solidify. The plates were then incubated at 37 °C for 24 – 48 h.

3.7.3 Fungal count

From suitable dilutions of sample, 0.1 ml was aseptically transferred into Sabouraud Dextrose Agar (SDA) containing 0.1g chloramphenicol per one liter to inhibit bacterial growth. The sample was spread all over the plates using sterile bent glass rod and then the plates is incubated at 28 °C for 48 hours (Harrigan and Mc Cance, 1976).

3.7.4 Coliform count

Coliform bacteria will be carried out on violet red bile agar medium and incubated for 24 hours at 37 °C for total coliforms and 44 °C for fecal coliforms according to the standard (ISO 4832); *E. coli* will be streaked onto eosine methylene blue (EMB) agar and then incubated overnight at 37 °C.

3.7.5 Enumeration of lactic acid bacteria

Viable bacteria count in the yoghurt sample was enumerated using the pour plate technique. The counts were enumerated on De Man Rogosa Sharpe agar (Oxoid, Australia) and anaerobic incubation at 43 °C for 72 h was used for the differential enumeration of the lactic acid Bacteria (Dave and Shah, 1996).

3.8 Sensory Quality Evaluation and Acceptability Test

Acceptance testing method described by Ihekoronye and Ngoddy (1985) was used to investigate the acceptability of the goat milk yoghurt compared with cow milk yoghurt (control) using the optimized processing conditions. Determination of acceptability was done using 30 untrained panelists who were familiar with yoghurt and were

willing to participate, the panelist were recruited at Federal University of Agriculture, Abeokuta. Briefing regarding the evaluation was given at the beginning of the session. Each panelist was assigned a number for identification purposes and he/she was responsible to evaluate two different samples. Samples were coded using a 3-digit random number and served successively. Panelists were asked to fill out a score sheet for each yoghurt sample they evaluated in term of taste, mouthfeel, aroma and overall acceptability. Each sample attribute was rated using a nine-point Hedonic Scale. The nine points on the Hedonic Scale were: dislike extremely = 1, dislike very much = 2, dislike moderately = 3, dislike slightly = 4, neither like nor dislike = 5, like slightly = 6, like moderately = 7, like very much = 8 and like extremely = 9. The average and mean values of scores for each of attributes was computed and analyzed statistically.

3.9 Statistical Analysis

The physico-chemical and microbiological data of yoghurt samples were evaluated using design expert version 8.0 while the sensory analysis of the yoghurt samples was statistically evaluated using paired t-test.

CHAPTER FOUR

4. RESULTS

4.1 Physico- Chemical and Microbiological Quality of Fresh West African Dwarf (WAD) Goat Milk and Cow Milk

The physical, chemical and microbiological quality of the fresh West African Dwarf goat milk (WAD) and cow milk were analyzed and presented in Table 5. WAD goat milk had total solid (TS) of 20.88 mg/l and cow milk had a TS of 13.32 mg/l. Table 5 also shows the mean values for fat, protein, viscosity, pH and titrable acidity for WAD goat milk to be 8.93%, 6.16%, 163627 mm^2/s, 6.48, 0.2%, respectively while cow milk had mean values for fat, protein, viscosity, pH and titrable acidity 3.86%, 9.84%, 282009 mm^2/s , 6.06, 0.63% respectively. The mean values for the microbial quality of WAD goat milk are 1.28×10^8 cfu/ml for total plate count (TPC), 6.0×10^6 cfu/ml for fungal count (FC), and 1.68×10^8 cfu/ml for lactic acid bacteria count (LABC) while for cow milk the mean values for TPC is 1.11×10^8 cfu/ml, FC is 3.2×10^7 cfu/ml and LABC is 1.56×10^8 cfu/ml. There was no growth for the coliform count of the fresh WAD goat milk and cow milk. WAD goat milk and cow milk had a negative result to the 75% alcohol test, and the methylene blue reduction time (Hr.) results for WAD goat milk and cow milk are > 6.0 and > 4.5 respectively.

4.2 Effect of Processing Parameters on Physico-Chemical and Microbiological Quality of WAD Goat Yoghurt

Table 6 shows the mean values of the responses at different experimental runs. The pH, titrable acidity, viscosity, fat, and protein content of the West African Dwarf goat yoghurt ranges from 4.35 to 5.97, 0.57% to 3.70%, 130158 mm^2/s to 272712 mm^2/s, 4.00% to 10.33% and 3.24% to 28.44%, respectively.

Table 5: Mean Values for Physico-Chemical and Microbiological Quality of Fresh Goat Milk and Cow Milk

Parameters	Goat milk	Cow milk	t –Stat	(T<=t)2-tail
Total Solids (mg/L)	20.88 ± 0.27	13.32 ± 0.07	1513	0.0004*
Fat (%w/w)	8.93 ± 0.60	3.86 ± 0.5	506	0.001*
Protein (%w/w)	6.16 ± 0.04	9.84	-1462	0.004*
Viscosity (mm^2/s)	163627± 0.5	282009± 0.5	-118383	5.38×10^{-06}
pH	6.26 ± 4.58	6.06 ± 4.29	41	0.016*
Titrable Acidity (% Lactic Acid)	0.2 ± 0.14	0.63 ± 0.45	-42	0.015*
Methylene Blue Reduction Time (h)	> 6.0	> 4.5	16	0.039*
Alcohol Test	Negative	Negative		-
Total Plate Count (cfu/ml)	1.28×10^8	1.11×10^8	33	0.019*
Fungal Count (cfu/ml)	6.0×10^6	3.2×10^7	29	0.022*
Lactic Acid Bacteria Count(cfu/ml)	1.68×10^8	1.56×10^8	11	0.022*
Coliform Count (cfu/ml)	Nil	Nil	Nil	Nil

*Values are means of duplicate determination
*significant ($p \leq 0.05$)

Table 6: Mean Values of the Responses at Different Experimental Runs for WAD Goat Yoghurt

Experimental Runs	Pasteurization Temp (ºC).	Incubation Temp. (ºC).	Time (h)	pH	Titrable Acidity (%)	Viscosity (mm^2/s)	Fat (%w/w)	Protein (%w/w)
1	80	40	3.5	4.52	3.60	201435	4.00	6.21
2	80	42.5	2.5	4.63	0.91	272712	5.07	28.44
3	80	42.5	4.5	4.58	2.30	139445	6.00	4.86
4	80	45	3.5	4.63	0.91	272712	5.07	28.44
5	82.5	40	2.5	5.52	0.72	148752	7.00	4.59
6	82.5	40	4.5	4.68	1.14	179742	10.33	28.44
7	82.5	42.5	3.5	4.63	0.91	272712	5.07	28.44
8	82.5	42.5	3.5	5.48	2.10	170445	7.00	5.43
9	82.5	42.5	3.5	4.98	0.76	130158	6.57	17.50
10	82.5	42.5	3.5	4.58	1.90	247920	6.00	5.43
11	82.5	42.5	3.5	4.57	2.20	192138	8.00	3.24
12	82.5	45	2.5	5.97	0.57	210732	8.36	8.75
13	82.5	45	4.5	4.82	3.30	210732	8.00	4.11
14	85	40	3.5	4.63	0.91	272712	5.07	28.44
15	85	42.5	2.5	4.63	0.91	272712	5.07	28.44
16	85	42.5	4.5	4.35	3.70	185940	7.00	4.77
17	85	45	3.5	4.70	0.67	136356	9.76	15.31

4.2.1 Effect of processing parameters on the pH of WAD goat yoghurt

From the regression coefficient table (Table 7), the quadratic model developed for the pH as a function of the independent variables has the coefficient of determination (R^2) of 0.96 and F-value of 18.69. The response surface and contour plots for pH of yoghurt at different experimental conditions are presented in Figure 5 and 6. From the figures, it can be observed that as pasteurization temperature and incubation temperature increase at constant incubation time, the pH value decreases, and also an increase in pasteurization temperature and incubation time at a constant incubation temperature shows a decrease in the pH value. Furthermore, when pasteurization temperature was held constant, lower pH value was also obtained as incubation temperature and time increases. The main effect of incubation temperature and time significantly ($p<0.05$) affects the pH parameter negatively, also the quadratic effects of incubation time, and the interaction effects of incubation temperature and time significantly ($p<0.05$) affects the pH of the goat milk yoghurt positively.

4.2.2 Effect of processing parameters on the titrable acidity of WAD goat yoghurt

The quadratic model developed for the titrable acidity as a function of the independent variables has the coefficient of determination (R^2) of 0.96 and F-value of 18.04. The main effects of pasteurization temperature and incubation time significantly ($p<0.05$) affected the titrable acidity of the yoghurt, the quadratic effects of pasteurization temperature significantly ($p<0.05$) affected the titrable acidity of the yoghurt, also quadratic effect of incubation time significantly ($p<0.05$) affected the titrable acidity value negatively. The response surface and contour plots for titrable acidity of yoghurt at different experimental conditions are presented in Figure 7 and 8.

Table 7: Regression Coefficients of the Responses as a Function of the Independent Variables

Parameters	pH	Titrable Acidity (%)	Viscosity (mm^2/s)	Fat (% w/w)	Protein (% w/w)
β_0	4.63	0.91	24.00	5.07	28.44
A	-1×10^{-2}	0.70*	-2710.38	-0.13	0.30
B	-0.17*	0.11	8523.50	-0.40	2.80
C	-0.46*	0.41*	11233.88	0.52	2.21
A^2	-0.054	1.63*	-39126.12*	-0.29	-18.14*
B^2	0.046	0.31	-63143.37*	1.72*	-5.70*
C^2	0.41*	-0.43*	-45321.63*	1.97*	-5.24*
AB	-0.073	0.100	-2.50	-1.50*	0.93
AC	-0.048	0.11	-20918.25	0.25	-0.38
BC	0.24*	0.070	30990.00	0.59	1.09
R^2	0.96	0.96	0.89	0.88	0.95
F-value	18.69	18.04	6.14	5.83	15.24
PRESS	1.90	12.22	8.648×10^{10}	95.11	1524.21

*Values are significant at 5% level *A- Pasteurization Temperature *B- Incubation Temperature *C- Incubation Time

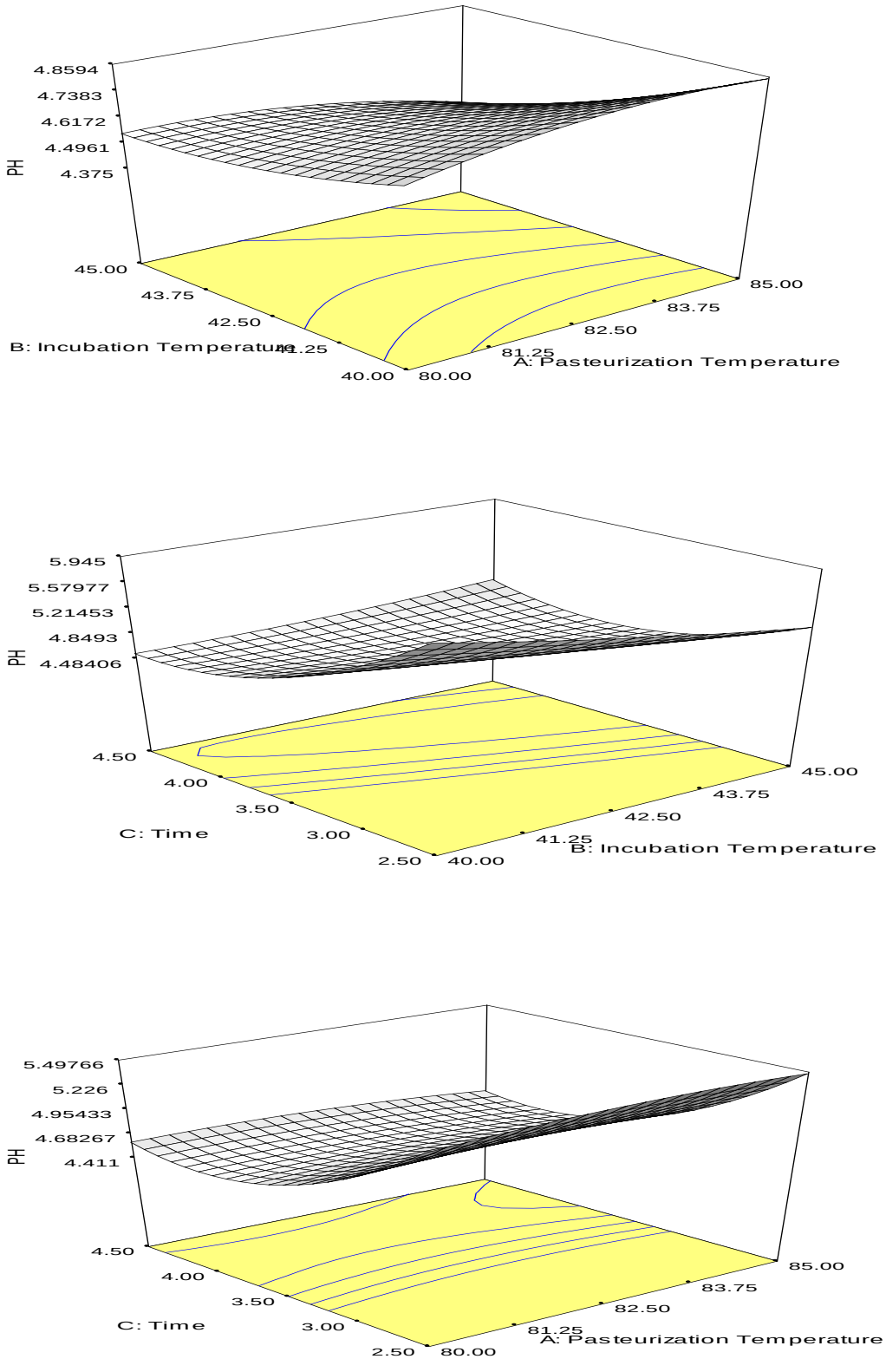


Figure 5: Response Surface Plots for pH parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions

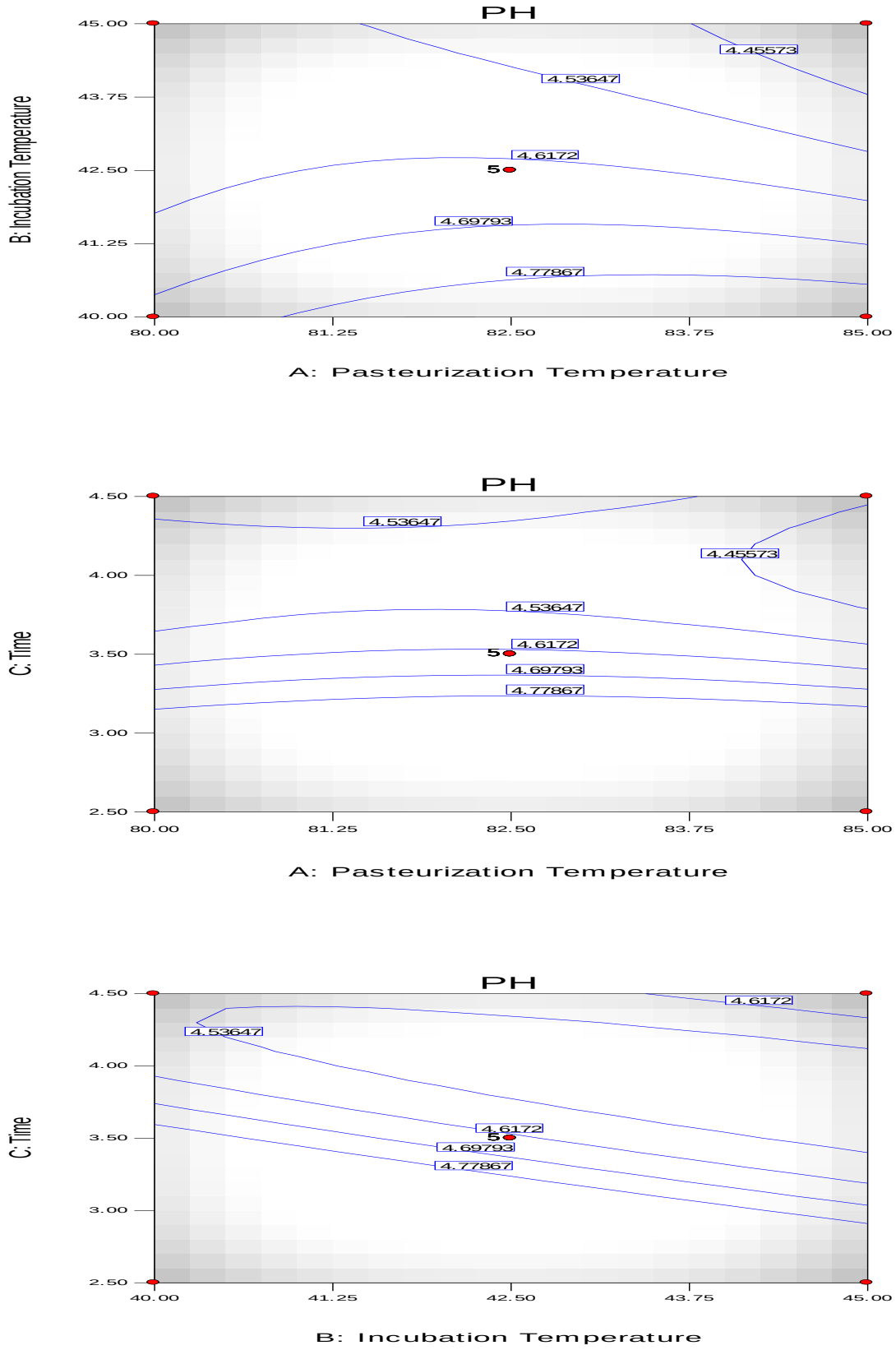


Figure 6: Contour Plots for pH parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions

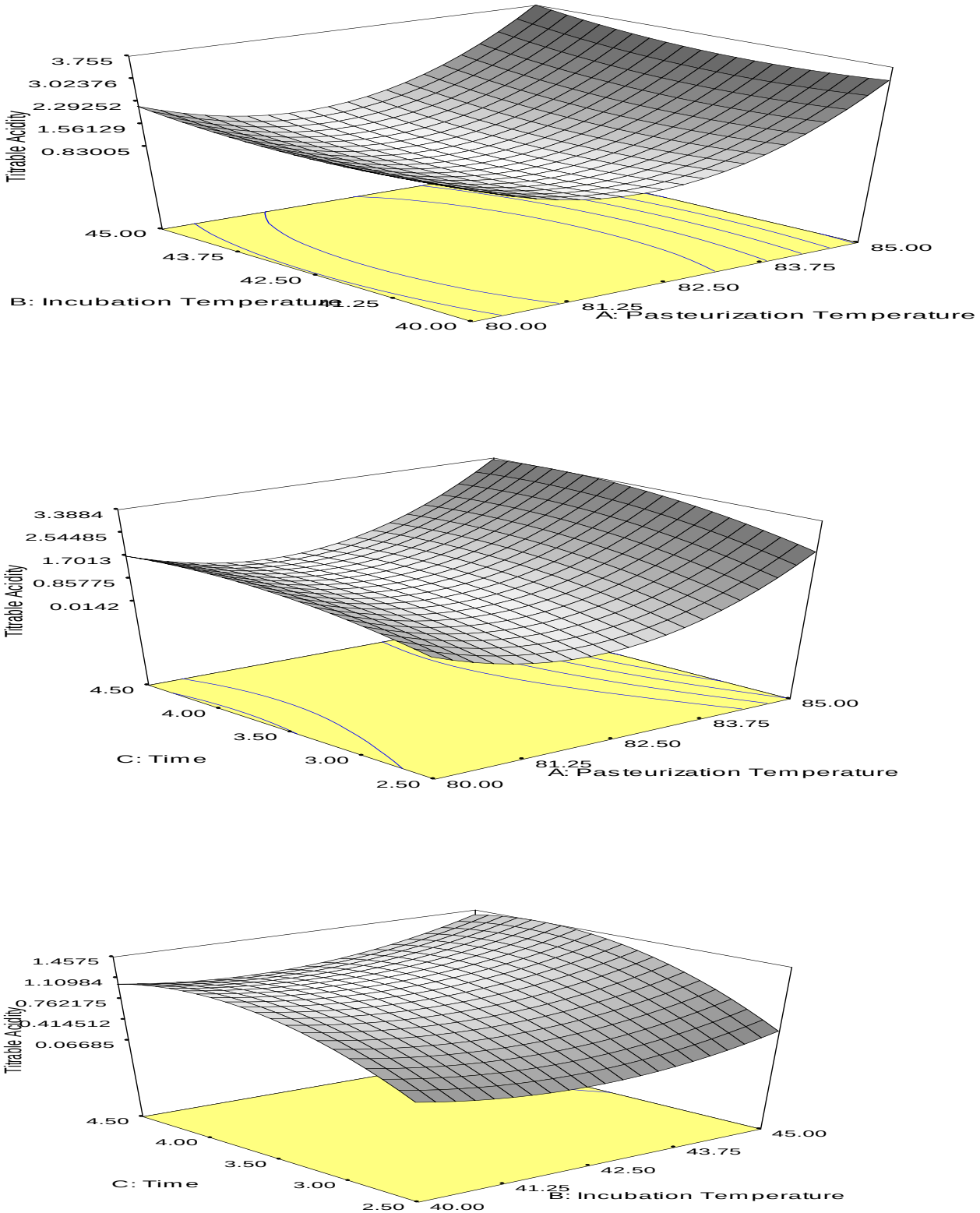


Figure 7: Response Surface Plots for Titrable Acidity Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

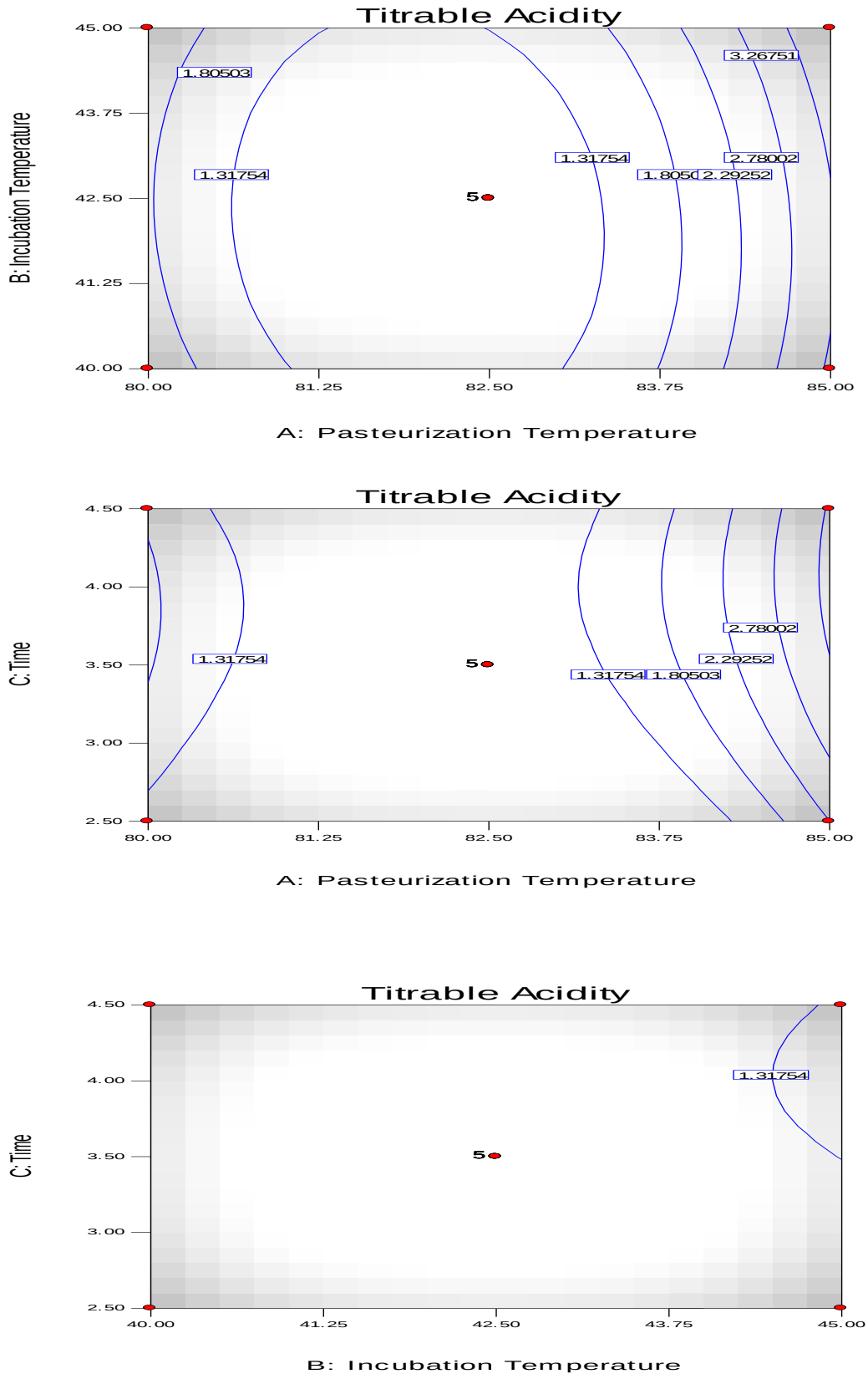


Figure 8: Contour Plots for Titrable Acidity Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

From the figures, it can be observed that as pasteurization temperature and incubation temperature increase at constant incubation time, the titrable acidity value shows an increase, also an increase in pasteurization temperature and incubation time at a constant incubation temperature, shows an increase in the titrable acidity value and at constant pasteurization temperature, higher titrable acidity value was also obtained as incubation temperature and incubation time increases.

4.2.3 Effect of processing parameters on the viscosity of WAD goat yoghurt

The coefficient of determination of the regression model (Table 7) for viscosity value of yoghurt was 0.89 while the F -value was 6.14. Quadratic effects of pasteurization temperature, incubation temperature and time significantly ($p<0.05$) affected the viscosity parameter of the goat milk yoghurt negatively. From the response surface and contour plots (Figure 9 and 10), increasing pasteurization and incubation temperature (constant incubation time) increases the viscosity values while increasing incubation time and pasteurization temperature at constant incubation temperature also shows an increase respectively. In a similar manner, increase in incubation temperature and time at constant pasteurization temperature increases respectively.

4.2.4 Effect of processing parameters on the fat content of WAD goat yoghurt

From the regression coefficient table (Table 7), the quadratic model developed for fat has the coefficient of determination (R^2) of 0.88 and F-value of 5.83. The response surface and contour plots for fat parameter at different experimental conditions are presented in Figure 11 and 12. From the figures, as incubation temperature and pasteurization temperature increases, the fat values increase when incubation time was held constant, and when pasteurization temperature and incubation time increases, fat value increased and also show reduction when incubation temperature was held constant. At constant pasteurization temperature increase in incubation temperature and time resulted in low fat value.

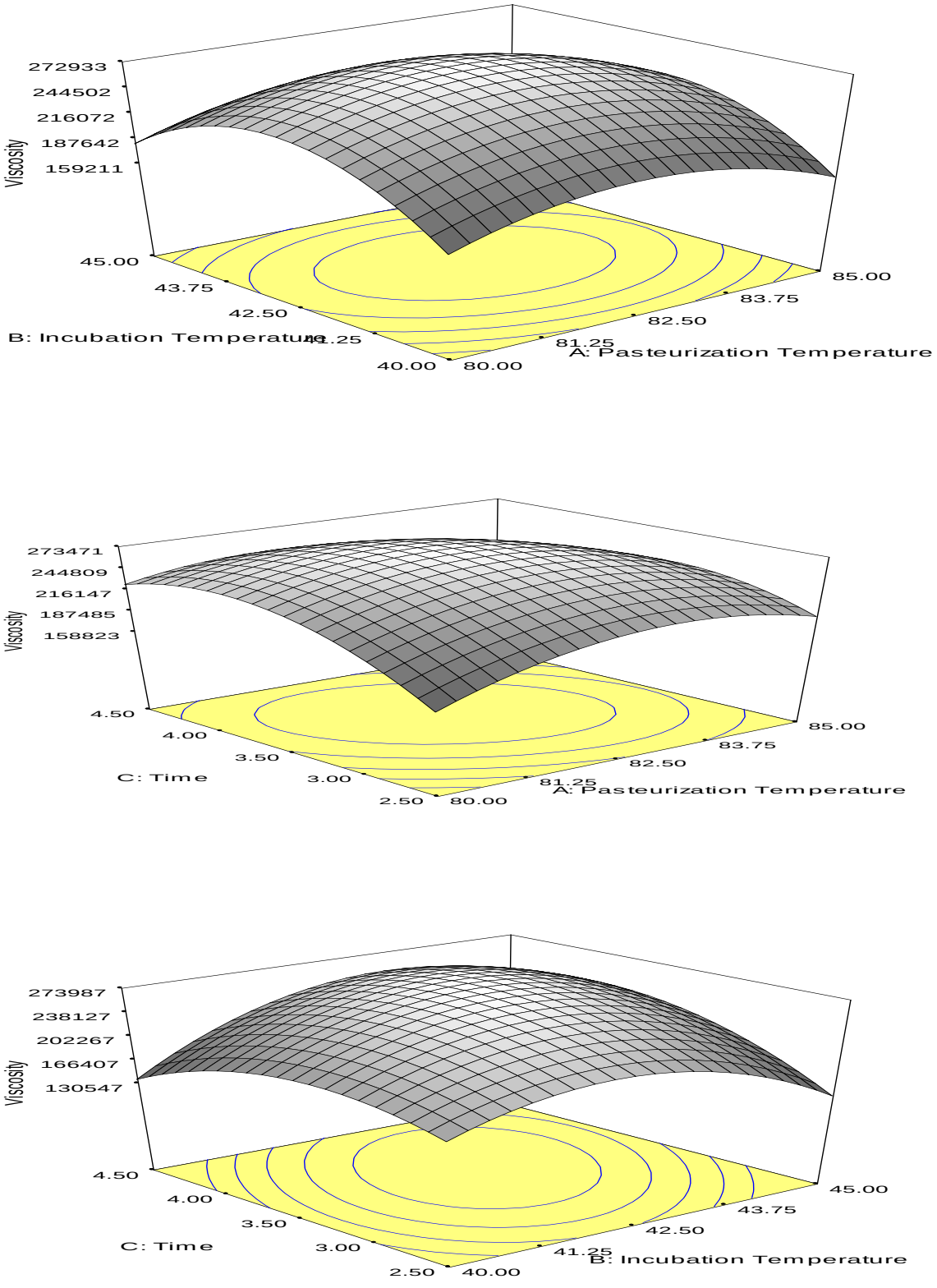


Figure 9: Response Surface Plots for Viscosity Parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions

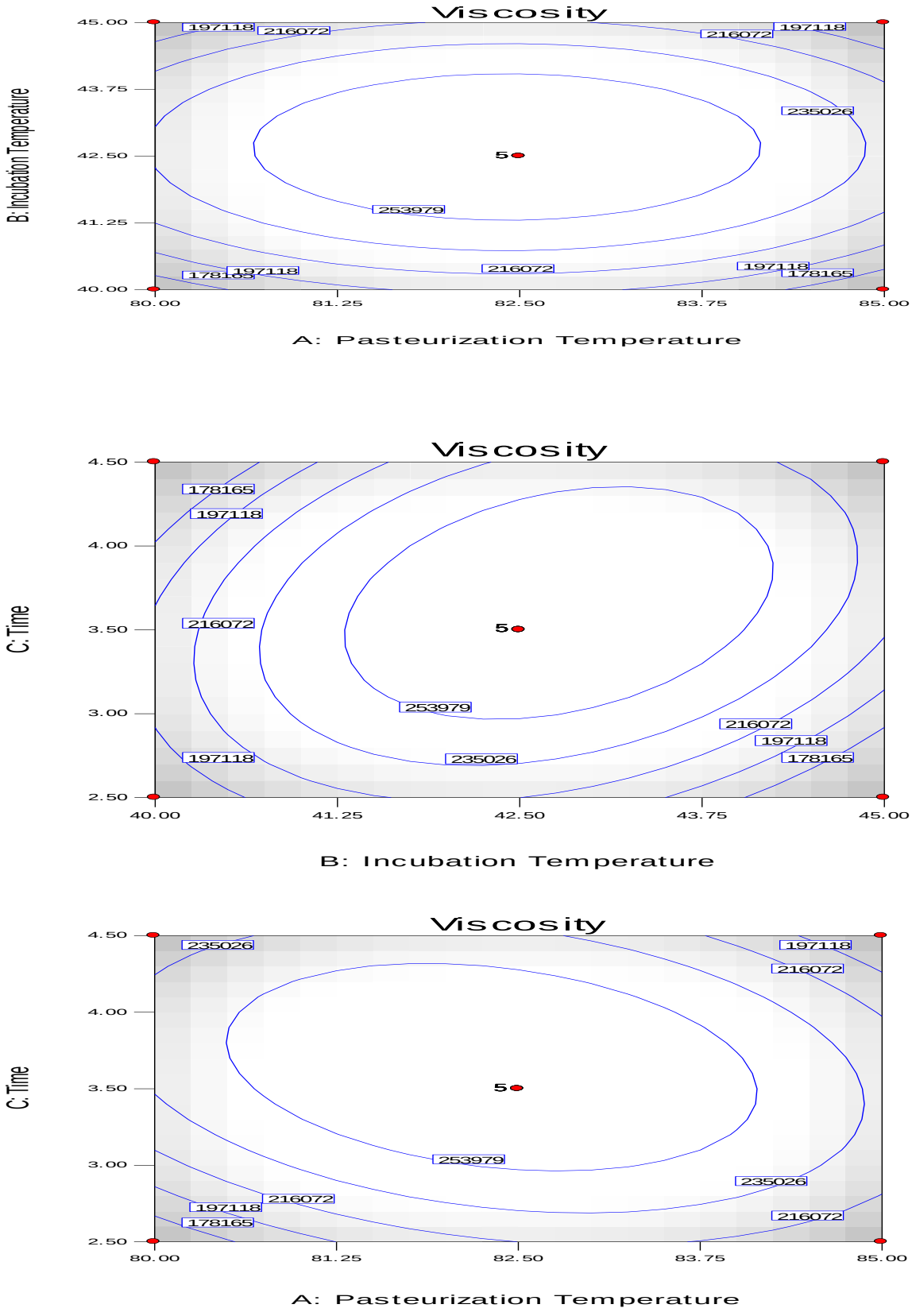


Figure 10: Contour Plots for Viscosity Parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions

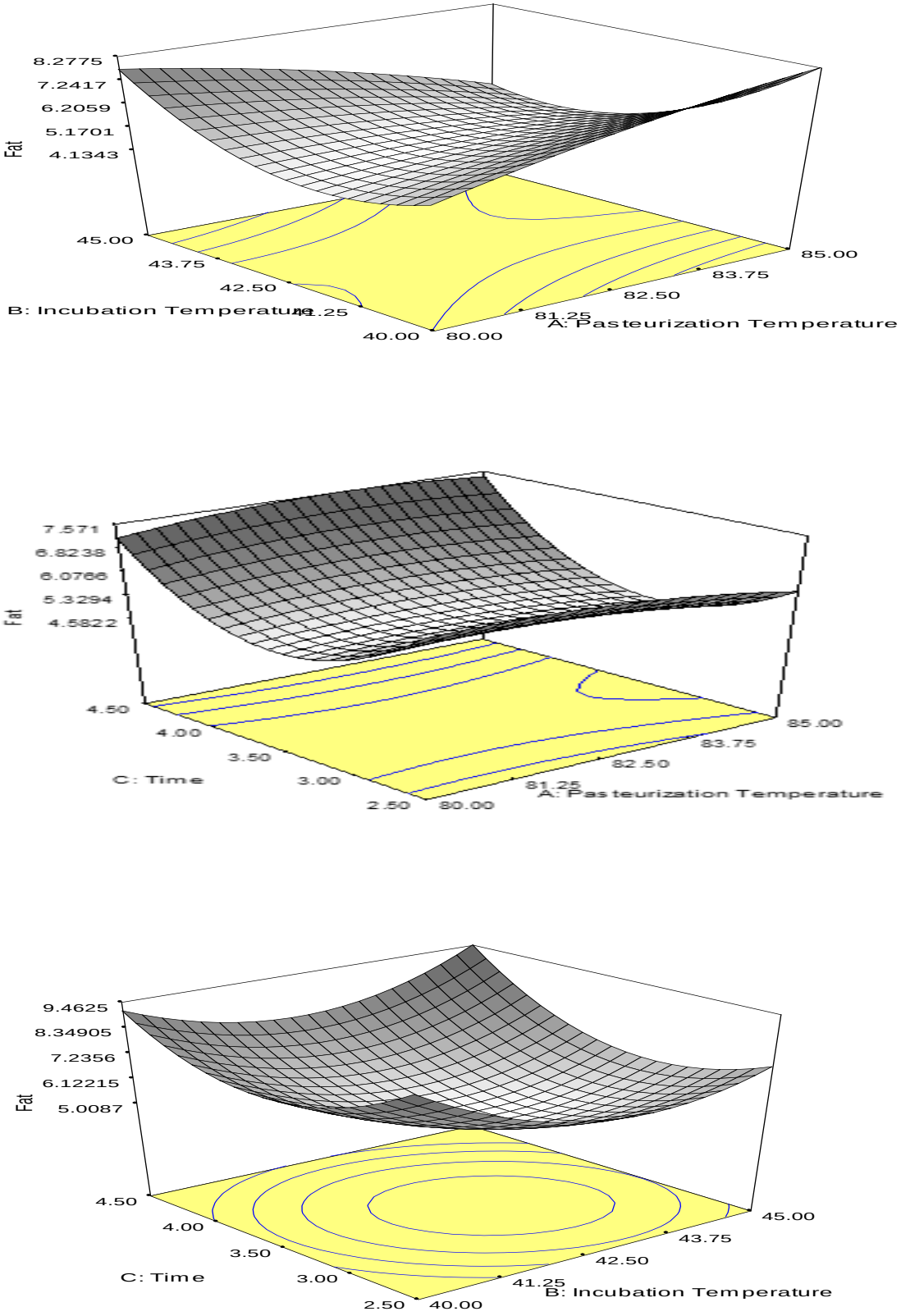


Figure 11: Response Surface Plots for Fat Parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions

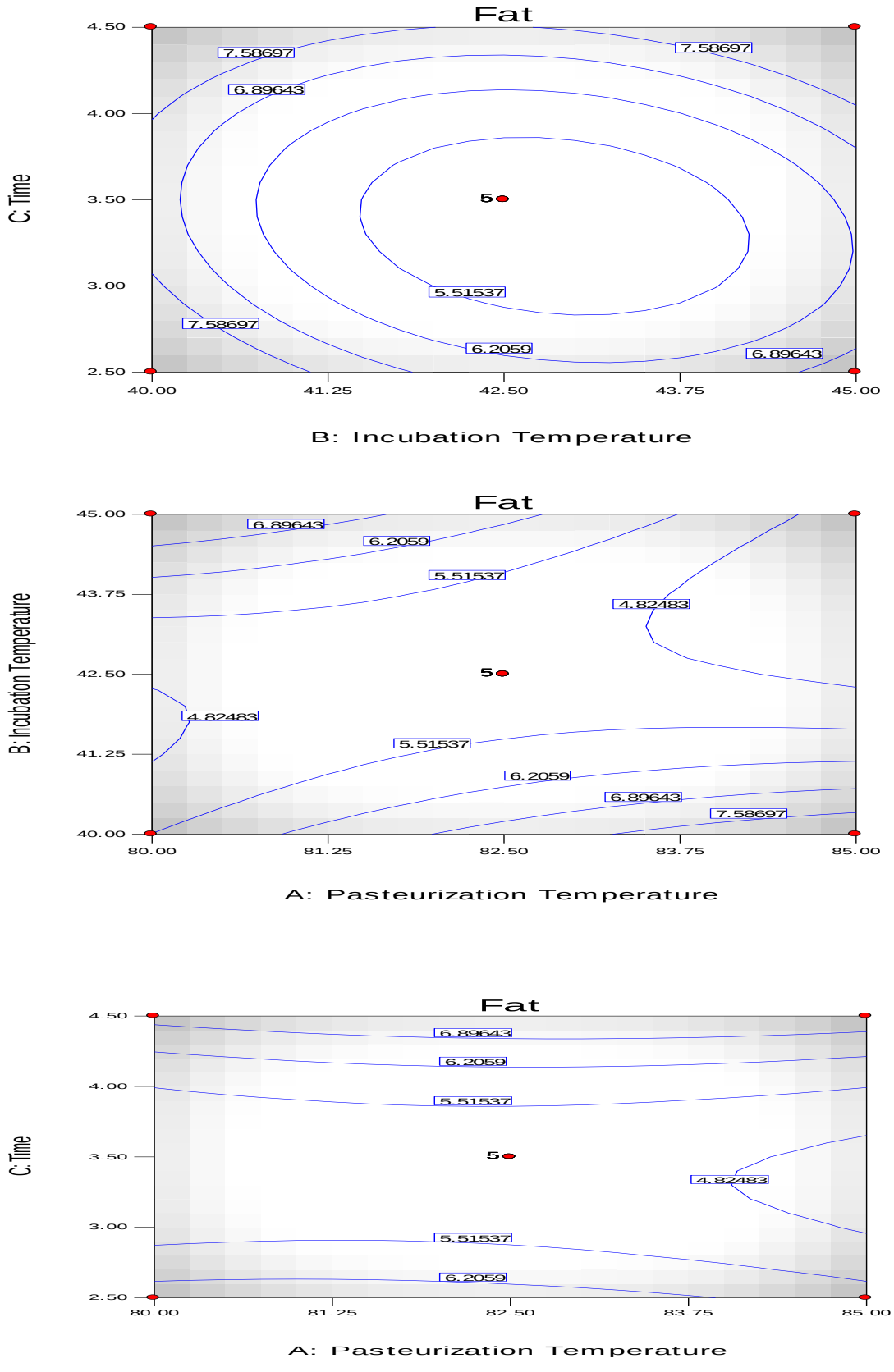


Figure 12: Contour Plots for Fat Parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions

The quadratic effect of incubation temperature and time significantly ($p<0.05$) affected the fat value. Also, the interaction effects of pasteurization temperature and incubation temperature significantly ($p<0.05$) affected the fat value negatively.

4.2.5 Effect of processing parameters on the protein content of WAD goat yoghurt

The quadratic model developed for protein parameter of the goat milk yoghurt has the coefficient of determination (0.95) and f-value of 15.24. From the contour and response surface plot for protein parameter (Figure 13 and 14) at constant incubation time, as incubation temperature and pasteurization temperature is increasing, protein value shows an increase in the value. Also, it was observed that when the pasteurization temperature and incubation time increased protein value increased when incubation temperature was fixed. Similarly, at constant pasteurization temperature as incubation temperature and time is increasing there was an increase in value of the protein content of the yoghurt. The quadratic effect of pasteurization temperature, incubation temperature and time significantly ($p<0.05$) affected the protein value negatively.

4.2.6 Effect of processing parameters on the total solid content of WAD goat yoghurt

Table 8 shows the total solid parameter of the WAD goat yoghurt varying between 13.54 and 32.64. From the regression coefficient table (Table 9), the quadratic model developed for total solid has the coefficient of determination (R^2) of 0.88 and F-value of 5.89. The response surface and contour plots for total solid parameter at different experimental conditions are presented in Figure 15 and 16.

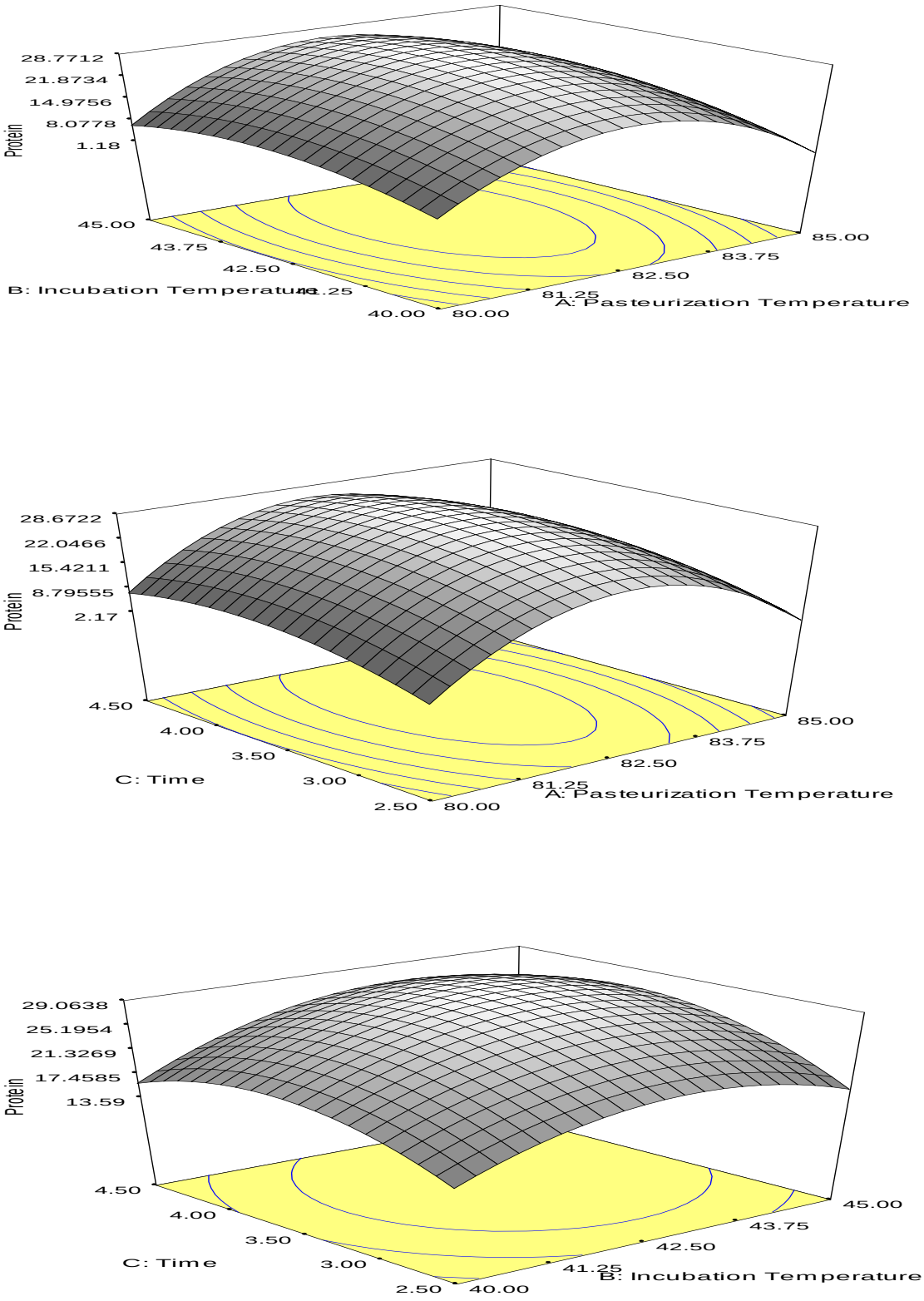


Figure 13: Response Surface Plots for Protein Parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions.

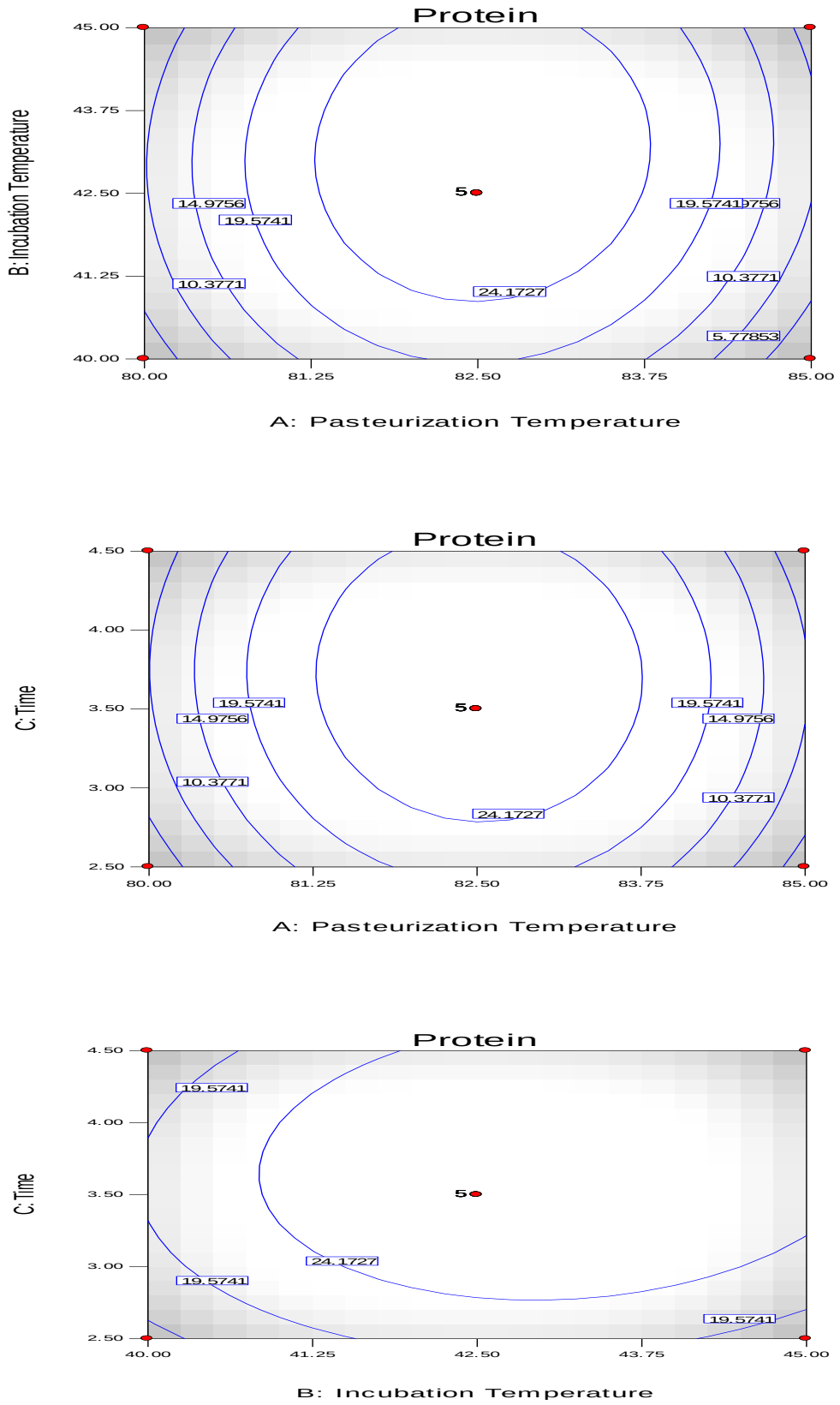


Figure 14: Contour Plots for Protein Parameters of West African Dwarf Goat Yoghurt at Different Experimental Conditions.

Table 8: Mean Values of the Responses at Different Experimental Runs

Experimental Runs	Total Solid (mg/L)	Total Plate Count (cfu/ml) (10^4)	Fungal Count (cfu/ml) (10^4)	Lactic Acid Bacteria Count (cfu/ml) (10^4)
1	16.82	9	0	26
2	24.93	27	0	114
3	16.79	29	21	340
4	24.93	24	0	205
5	17.44	20	24	0
6	26.57	30	10	550
7	24.93	23	1	289
8	14.35	11	2	82
9	32.61	24	7	280
10	21.18	32	23	11
11	18.94	35	20	468
12	32.64	21	2	70
13	13.54	5	0	2
14	24.93	20	4	216
15	24.93	24	0	156
16	14.12	0	2	100
17	19.66	14	16	43

Table 9: Regression Coefficients of the Responses as a Function of the Independent Variables

Parameters	Total Solid (mg/L)	Total Plate Count (cfu/ml) (10^4)	Fungal Count (cfu/ml) (10^4)	Lactic Acid Bacteria Count (cfu/ml) (10^4)
β_0	24.93	23.60	1.00	196.00
A	-1.94	-11.37*	-10.50*	-76.13
B	1.54	3.63*	-0.25	108.63
C	-1.94	0.00	2.00	34.00
A^2	-9.75*	-5.30*	6.62*	-87.25
B^2	1.35	1.20	2.62	100.25
C^2	1.60	-2.55	5.13*	-60.50
AB	0.28	-0.50	0.25	-26.00
AC	-0.99	-5.75*	0.25	1.75
BC	1.74	3.25*	-2.75	74.25
R^2	0.88	0.97	0.96	0.60
F-value	5.89	24.89	19.11	1.15
PRESS	1065.10	371.38	674.75	2.566×10^6

*Values are significant at 5% level *A- Pasteurization Temperature *B- Incubation Temperature *C- Incubation Time

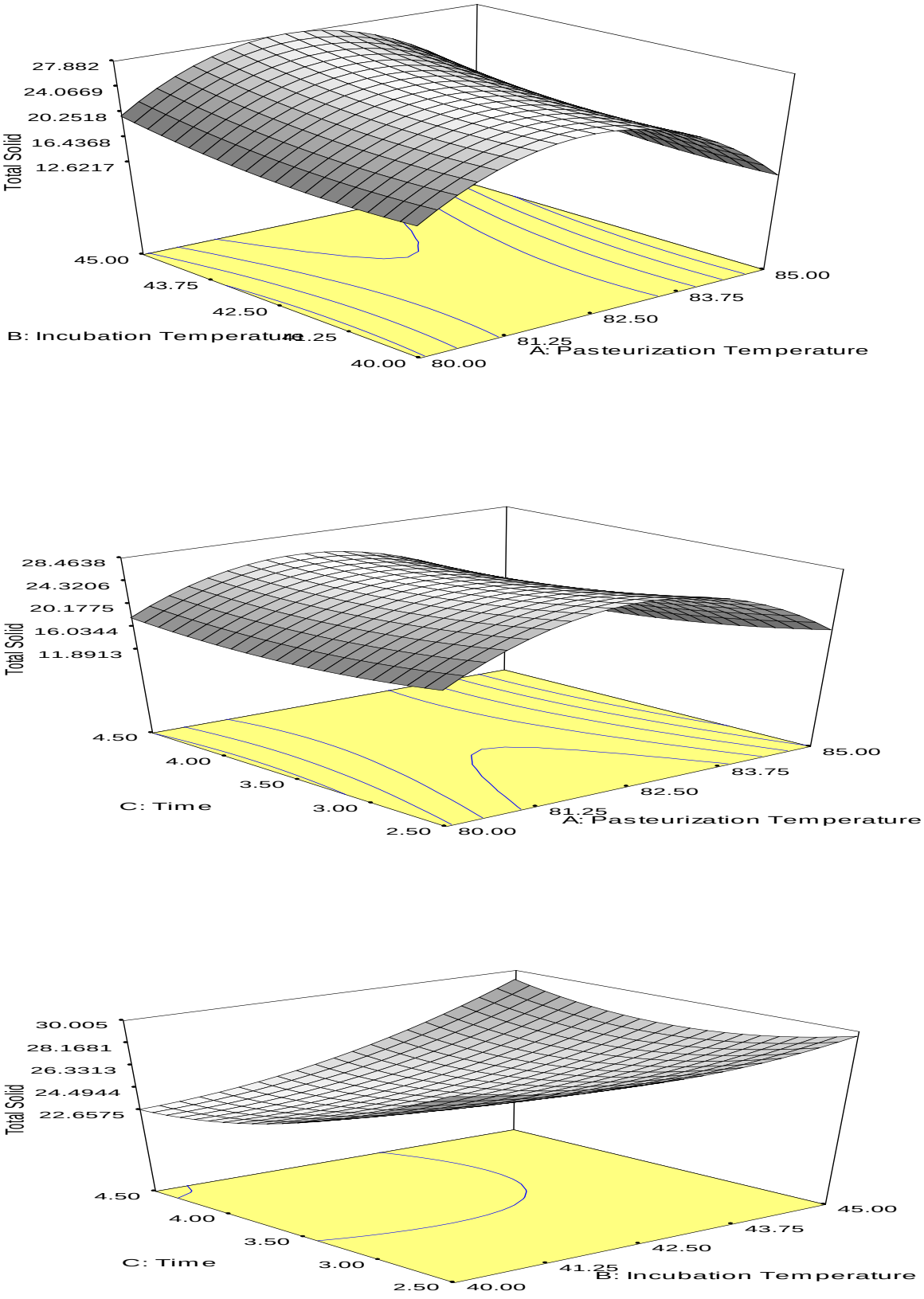


Figure 15: Response Surface Plots for Total Solid Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

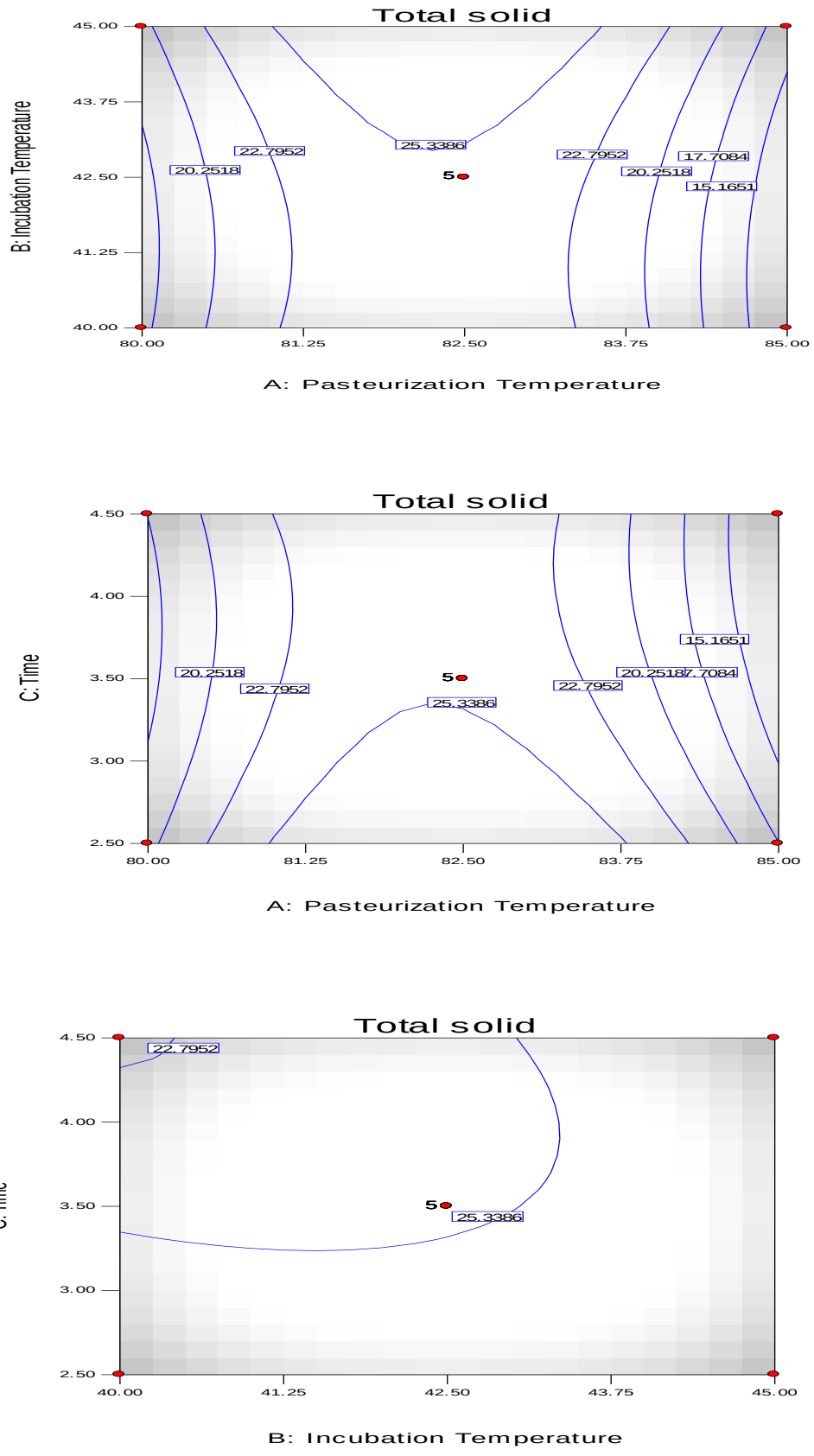


Figure 16: Contour Plots for Total Solid Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

From the figures, it can be observed that as incubation temperature and pasteurization temperature increases, the total solid values shows an increase and decrease when incubation time was held constant, and when pasteurization temperature and incubation time increases, total solid value increased and also show reduction when incubation temperature was held constant. At constant pasteurization temperature, increase in incubation temperature and time resulted in high total solid value. The quadratic effect of pasteurization temperature significantly ($p<0.05$) affected the total solid value negatively.

4.2.7 Effect of processing parameters on the total plate count of WAD goat yoghurt

Mean values for total plate count of the yoghurt varied between 5.0×10^4 and 3.5×10^5. The developed quadratic model can predict more than 90% of the experimental with F-value of 24.09. From the contour and response surface graphs shown in Figure 17 and 18, at fixed incubation time, when incubation temperature and pasteurization temperature is increasing total plate count increased and decreased respectively, also at constant incubation temperature total plate count value shows a similar trend as incubation time and pasteurization temperature increases. It was also observed that at constant pasteurization temperature when incubation time and incubation temperature is increasing total plate count value decreased and increased. Main effect of pasteurization temperature and incubation temperature significantly ($p<0.05$) affected total plate count, also the quadratic effect of pasteurization temperature significantly ($p<0.05$) affected the total plate count, the interaction effects of pasteurization temperature and incubation time, and incubation temperature and time respectively show a significant ($p<0.05$) effect on total plate count.

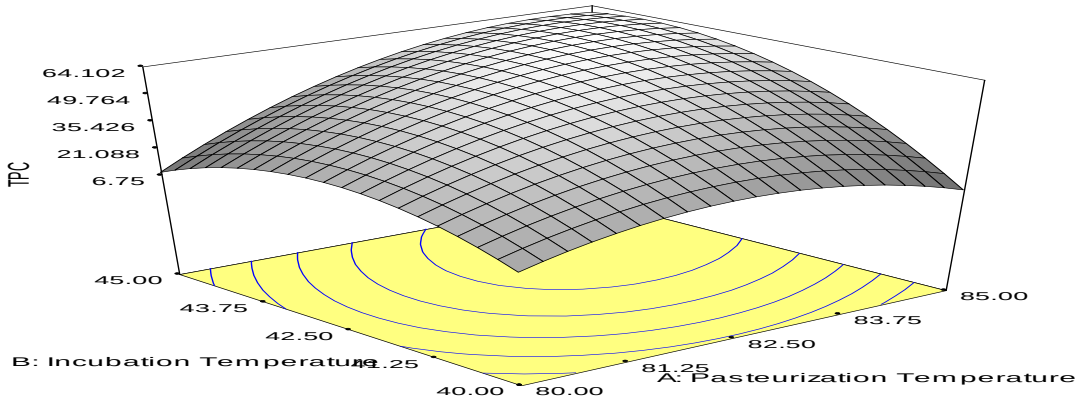
4.2.8 Effect of processing parameters on the fungal count of WAD goat yoghurt

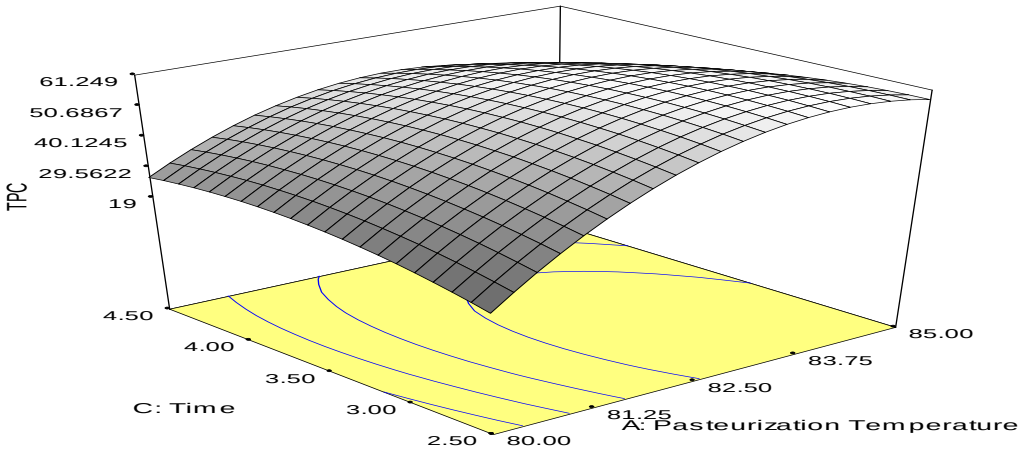
Fungal count value of the goat milk yoghurt varied between 1.0×10^4 and 2.4×10^5. The quadratic model developed for the fungal count of yoghurt has the highest coefficient of determination (0.96) and f-value of 19.11. Figure 19 and 20 shows the response surface and contour plots for the fungal count. From the figures, it can be observed that increasing incubation temperature and pasteurization temperature at constant incubation time decreases fungal count value. However, increasing incubation temperature and incubation time at constant pasteurization temperature resulted in decrease and increase of fungal count value. At constant incubation temperature, increase in incubation time and pasteurization temperature resulted in increase and decrease in fungal count value. The main effect of incubation temperature significantly ($p<0.05$) affected the fungal count of the goat milk yoghurt, also the quadratic effect of pasteurization temperature, incubation temperature, and time significantly ($p<0.05$) affected the fungal count, furthermore interaction effects of incubation temperature and time significantly ($p<0.05$) affected the fungal count of the goat milk yoghurt.

4.2.9 Effect of processing parameters on the lactic acid bacteria (LAB) count of WAD goat yoghurt

Lactic acid bacteria count of WAD goat yoghurt varied between 2.0×10^4 and 5.50×10^6. The quadratic model developed for the LAB count of the yoghurt has the coefficient of determination (0.60) and f-value of 1.15. Figure 21 and 22 shows the response surface and contour plots for Lactic acid bacteria count. From the figures, (at constant incubation time) it can be observed that increasing incubation temperature results in an increase and increasing pasteurization temperature decreases the lactic acid bacteria count. However, increasing pasteurization temperature and incubation time (at constant incubation temperature) resulted in a decrease. At constant pasteurization temperature, an increase occurs in lactic acid bacteria count of the

yoghurt at increase in incubation time and incubation temperature, also no significant difference was observed.





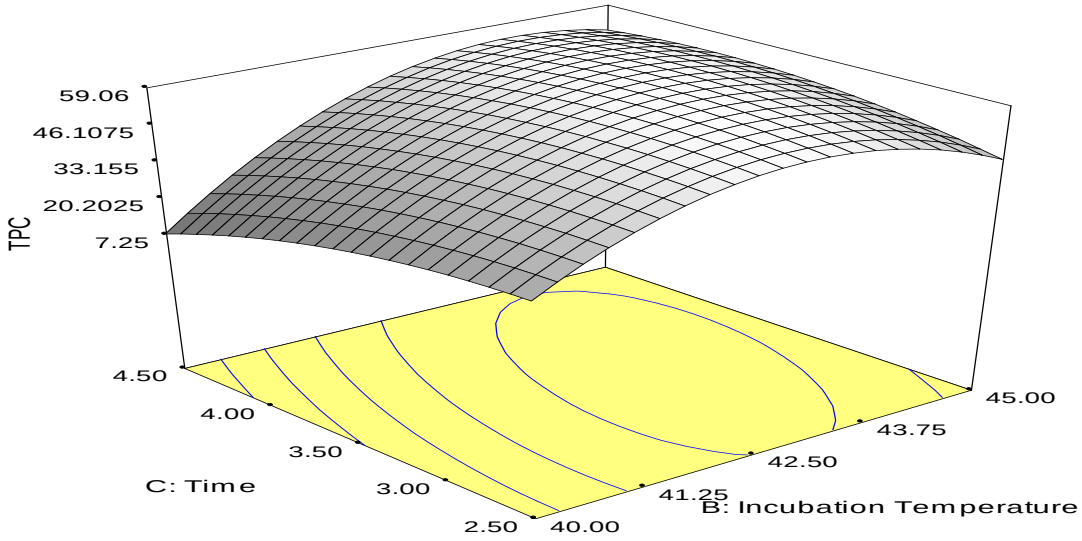
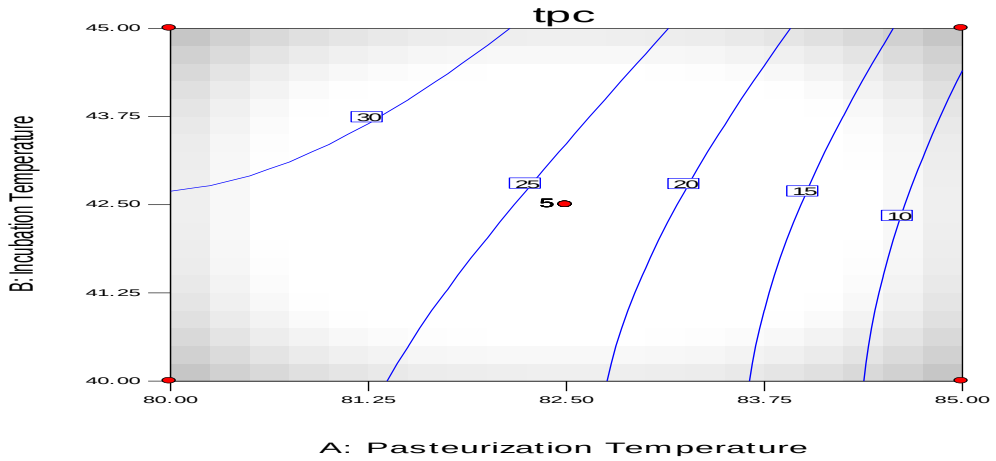
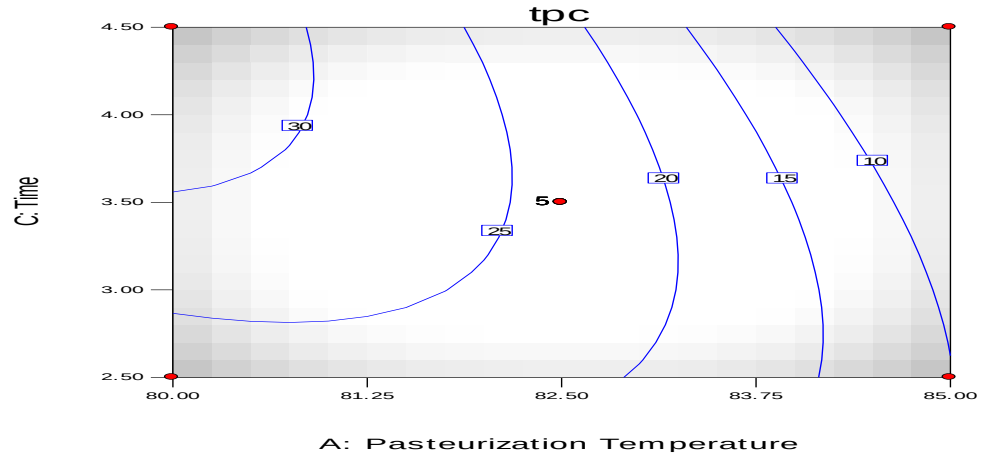


Figure 17: Response Surface Plots for Total Plate Count (TPC) Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions





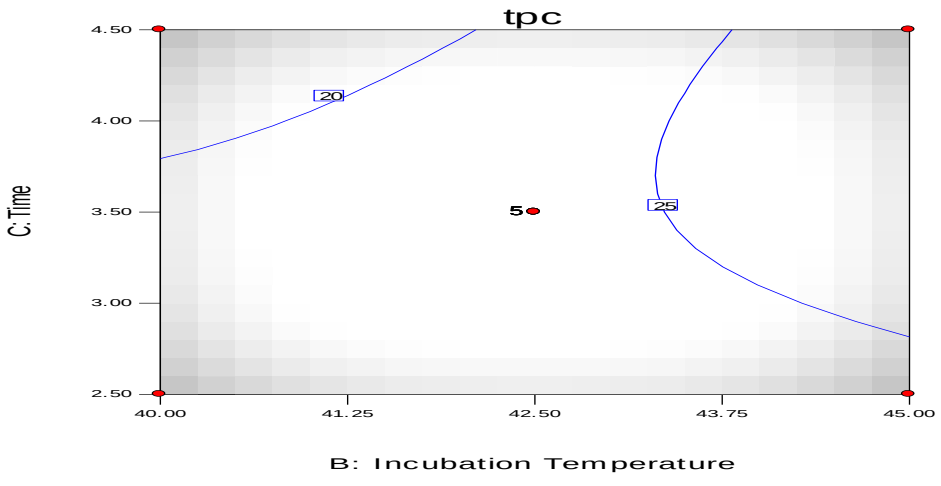


Figure 18: Contour Plots for Total Plate Count (TPC) Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

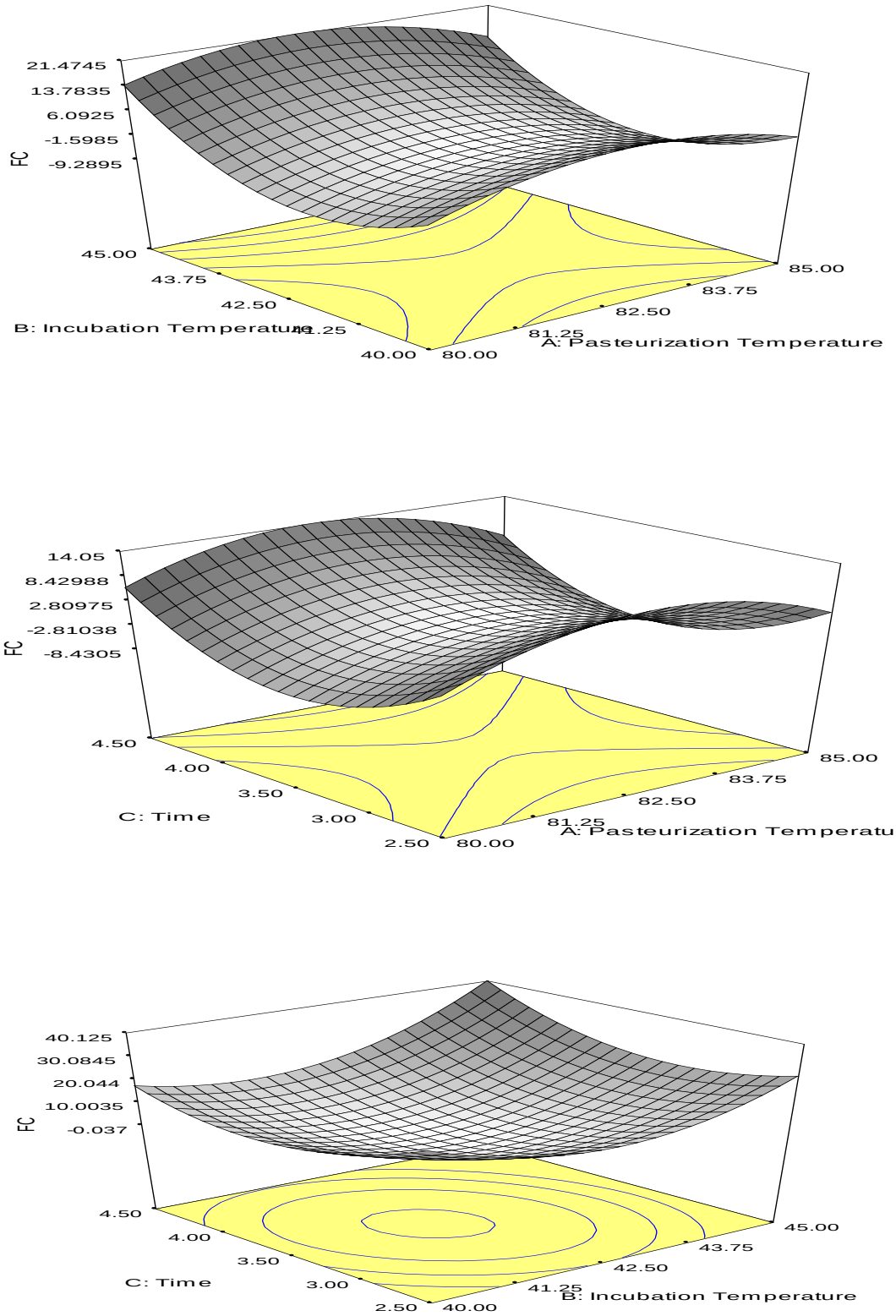


Figure 19: Response Surface Plots for Fungal Count (FC) Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

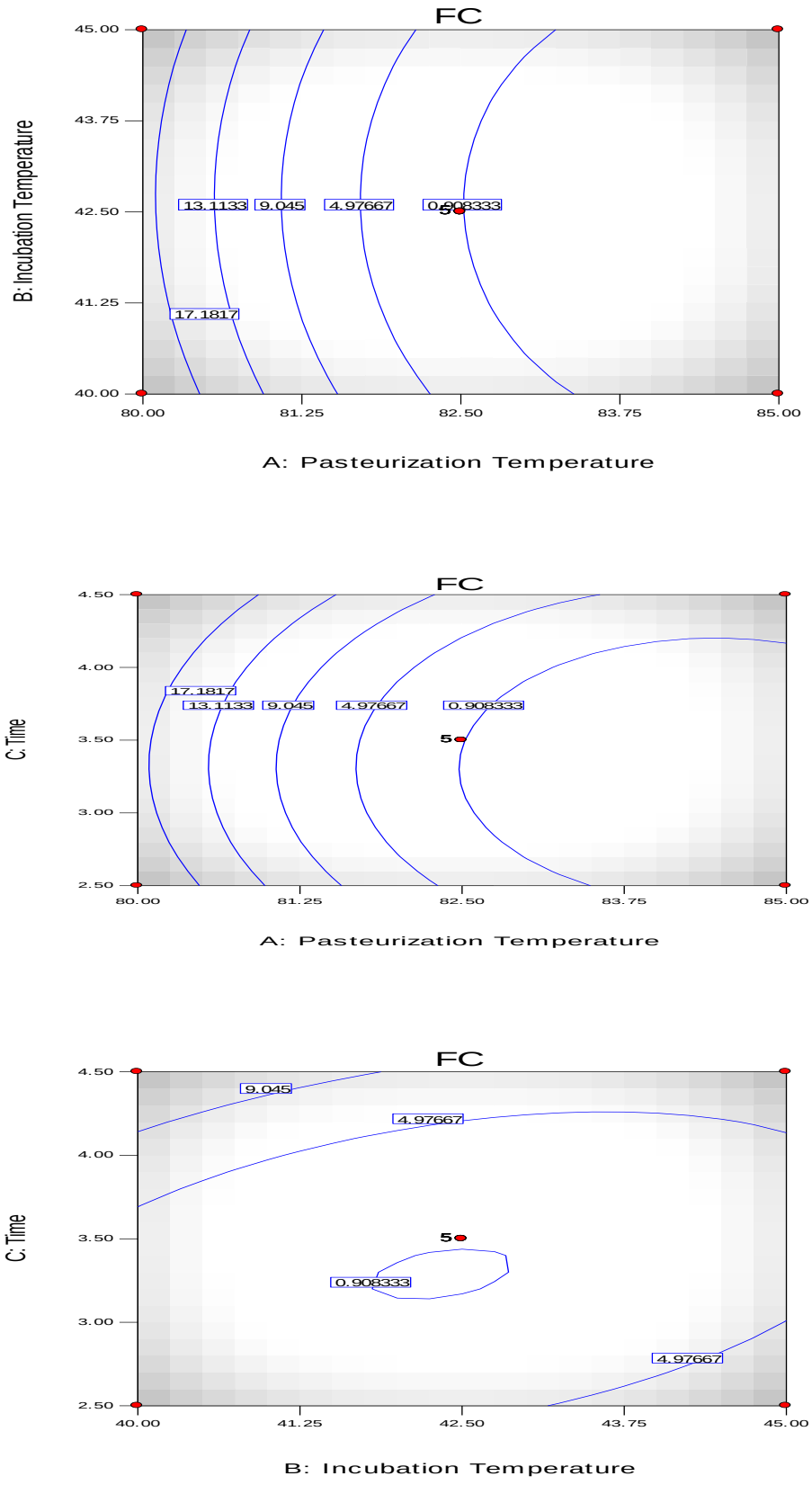


Figure 20: Contour Plots for Fungal Count (FC) Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions

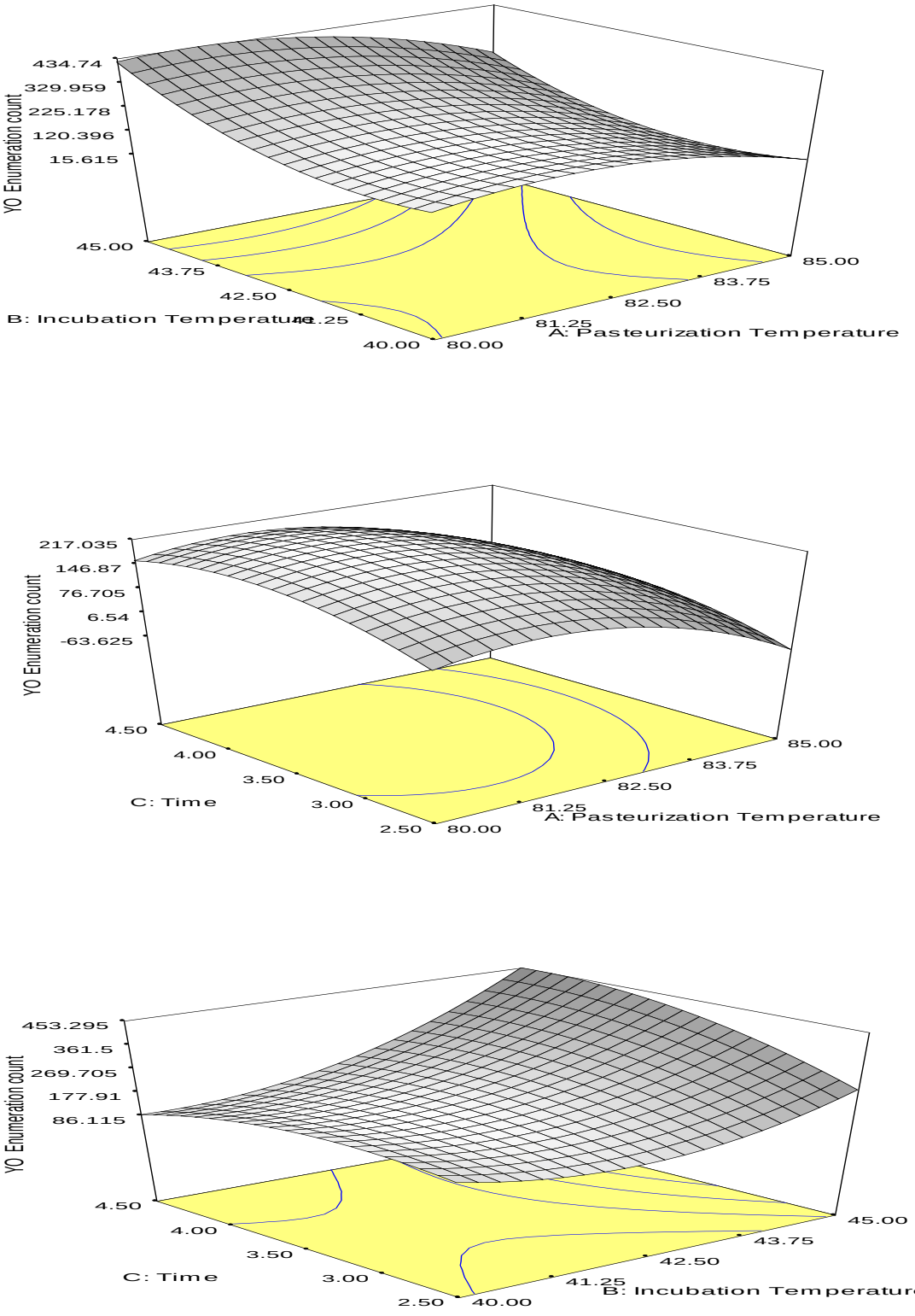
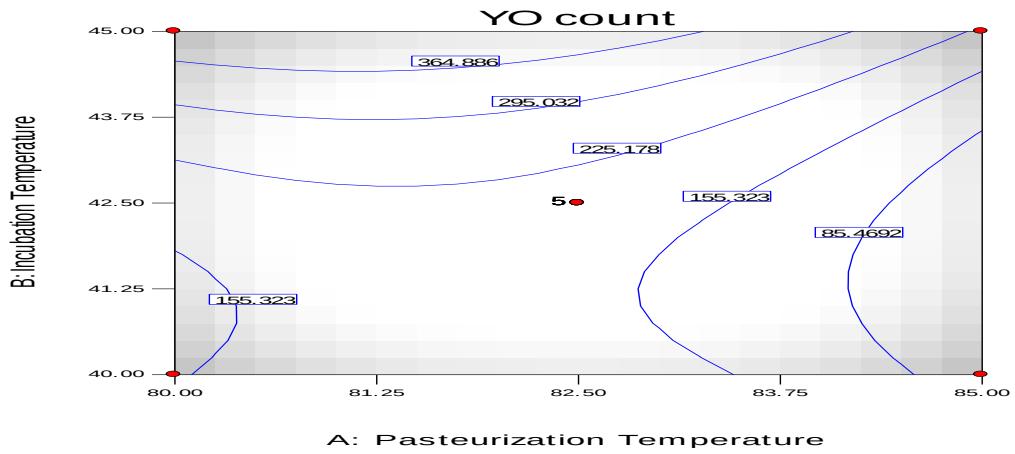
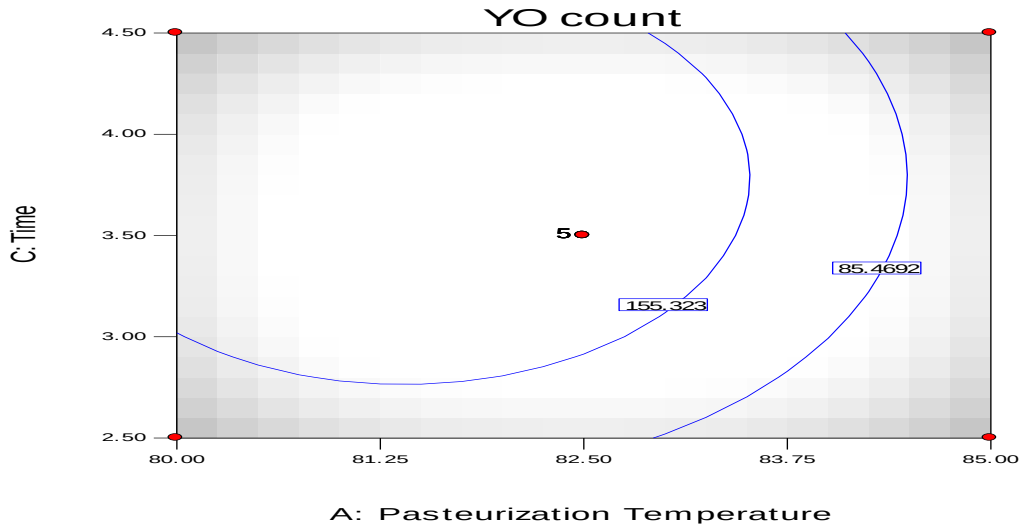


Figure 21: Response Surface Plots for Lactic Acid Bacteria Count Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions.





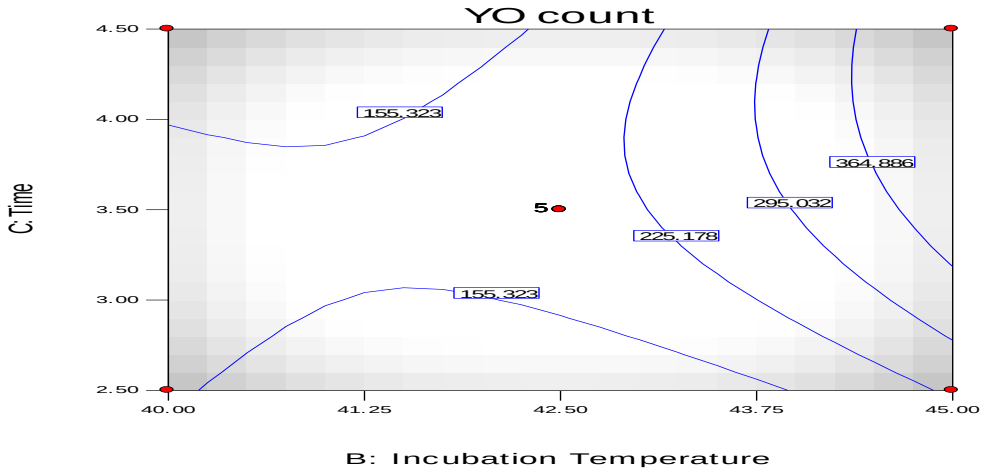


Figure 22: Contour Plots for Lactic Acid Bacteria Count (YO Count) Parameter of West African Dwarf Goat Yoghurt at Different Experimental Conditions.

4.3 Optimization of Process parameters for WAD Goat Yoghurt

In order to optimize the pasteurization temperature, incubation temperature and incubation time during the yoghurt process, while retaining high quality of the WAD goat yoghurt, response surface methodology (RSM) was used and selecting the significant ranges for the variables is the most important step in response surface methodology. pH, titrable acidity, viscosity, fat, protein, total solid, total plate count, fungal count and lactic acid bacteria count were the main quality parameters of the WAD goat yog3hurt in this research which were also the criteria based on desirability concept with pasteurization temperature (A), incubation temperature (B) and incubation time (C) as well as these main quality parameters serving as the constraints to process optimization. The solution to the optimized WAD goat milk yoghurt has a pasteurization temperature of (84.24 ^{o}C), incubation temperature of (44.22 ^{o}C) and incubation time of (3.80 h).

In order to compare the optimized goat milk yoghurt with those from the cow milk yoghurt, the optimized processing parameters obtained were used (pasteurization temperature, incubation temperature and incubation time of 84.24 ^{0}C, 44.22 ^{0}C, and 3.8 h respectively). Table 10 shows the mean values ± SD of the determined parameters. The total solid, fat and protein content, viscosity, titrable acidity and pH values for the optimized goat and cow milk yoghurt were 20.53% and 16.95%, 5.18% and 1.37%; 20.09 % and 8.75%; 229780 mm^2/s and 309900 mm^2/s, 2.55% and 1.17%; 4.41 and 4.62, respectively. With respect to the microbiological parameters, total plate count, fungal count, lactic acid bacteria count and coliform count mean values for the goat and cow milk yoghurt were 1.5×10^5 and 5.0×10^4; -1.35×10^6 and 1.0×10^4; 2.33×10^6 and 3.60×10^6 for goat and cow milk yoghurt respectively, no count was detected for their coliform count. Significance ($p<0.05$) differences were observed in all the parameters analyzed for the optimized WAD goat and cow milk yoghurt.

Table 10: Mean Values for Physico-Chemical and Microbiological Quality of Optimized WAD Goat Milk Yoghurt and Control (Cow Milk Yoghurt)

Parameters	Quality Average			
	Goat Milk Yoghurt	Cow Milk Yoghurt	t Stat	P (T<=t) 2-tail
Total Solids (mg/L)	20.53	16.95	357	0.002 *
Fat (% w/w)	5.18	1.37	380	0.002 *
Protein (% w/w)	20.09	8.75	1133	0.0006 *
Viscosity (mm^2/s)	229780	309900	626030.4	1.02×10^{-6} *
pH	4.41	4.62	- 20	0.032 *
TTA (%)	2.55	1.17	137	0.005 *
TPC (cfu/ml)	1.5×10^5	5.0×10^4	231	0.003 *
FC (cfu/ml) 890P;[O/=		-1.35×10^6	1.0×10^4	-155.67 0.004 *
LABC (cfu/ml)	2.33×10^6	3.60×10^6	-126	0.005 *
Coliform Count (cfu/ml)	Nil	Nil	Nil	Nil

*Significance at (P<0.05) *TTA- Titrable Acidity * TPC- Total Plate Count * FC- Fungal Count * LABC-Lactic Acid Bacteria Count

4.4 Sensory Acceptability of WAD Goat Milk Yoghurt

Based on nine-point hedonic scale (1 = dislike extremely, 9 = like extremely), sensory scores (Table 11), showed the acceptability of the yoghurt for the control (cow milk yoghurt) and the optimized goat milk yoghurt. It was observed that sample 103 (goat milk yoghurt) was moderately accepted having the sensory rating of (6.07 to 6.37) while sample 268 (cow milk yoghurt) rating ranged between (7.73 to 8.20) was highly accepted, respectively. Significant ($p<0.05$) differences were observed in terms of all the attributes evaluated.

Table 11: Sensory Acceptability of Goat Milk Yoghurt Optimized Process Parameter

Sample	Taste	Mouth feel	Aroma	Overall Acceptance
West African Dwarf Goat Yoghurt	6.07±1.30	6.27±2.20	6.27±3.72	6.27±3.72
Cow Milk Yoghurt	8.20±0.71	8.20±0.71	8.20±0.71	8.20±0.71
t Stat	-9.33	-4.91	-3.89	-5.57
P(T<=t)2-tail	3.09×10^{-10} *	3.21×10^{-5} *	0.00 *	5.14×10^{-6} *

*Significance at (P<0.05)

CHAPTER FIVE

5. DISCUSSION

The basic ingredient for the production of yoghurt is milk and hence the quality of the incoming milk is an important consideration. pH of milk samples collected from the two species (WAD goat milk and cow milk) was determined at the time of sampling. pH is a measure of the hydrogen ion concentration, the mean value for pH of WAD goat milk was 6.26 and cow milk had a mean value of 6.06 Zahraddeen *et al.* (2007) reported a similar value for the pH of WAD goat milk to be 6.21. The mean value for the pH of cow milk (6.06) in this study was lower than the mean value for cow milk (6.65) reported by Rashida *et al.* (2004). The total solids, fat content for WAD goat milk in this study was similar to those reported by Akinyosinu *et al.* (1977), while the protein content was a little higher. Eissa (2008) reported the mean values for total solid and fat content of cow milk to be 12.60 and 3.75 respectively which is similar to the values for cow milk obtained in this study. According to Haenlein (1996), the composition of goat milk has a higher value of total solids, protein and fat than cow milk. The variation in the physico-chemical qualities of goat milk can be greatly influenced by several factors such as seasons, stages of lactation, breeds, diet, individual animal and human management conditions (Haenlein and Abdellatif, 2004). Titrable acidity is an important quality indicator, because it indicates lactose fermentation by lactic acid bacteria (Borsato-Moysés *et al.*, 2009). Asif and Sumaira (2010) investigated the physico-chemical parameters of bovine and non-bovine animals, their result for titrable acidity of goat milk is similar to values obtained in this study, while their values for cow milk is lower than that obtained in this study. The viscosity of fresh goat milk was found lower than values reported by Amor *et. al.* (2013).

This research shows that both WAD goat milk and cow milk used in the processing of yoghurt had a good microbiological quality as seen by Methylene Blue Reduction Time MBRT which was more than 2 h and having negative result at alcohol 75% test. The time taken for the methylene blue to become colourless is the methylene blue reduction time (MBRT), the quicker the time (less than 2h) required to neutralize methylene blue, the worse microbiological quality of the fresh milk (Anderson *et al.*, 2011). Both fresh milk samples (WAD goat milk and cow milk) had no growth for the coliform count but had a higher count for total plate, fungal count and lactic acid bacteria count before the fresh milk samples were pasteurized.

West African Dwarf goat milk (WAD) was processed to WAD goat milk yoghurt as outlined in the experimental runs and further analyzed for the effect of pasteurization temperature, incubation temperature and incubation time on its physico-chemical, microbiological and sensory quality. Pasteurization temperature is an important process parameter, it causes; a partial breakdown of the whey proteins to amino acids that stimulate the activity of the starter culture, expulsion of oxygen from the milk which aids the growth of the lactic acid bacteria and a reduction in the indigenous microflora in the milk that might otherwise compete against the added lactic acid bacteria (Tamime and Robinson, 1999). Incubation process is the next step after pasteurization; the pasteurized milk was cooled to 42–43 °C, the starter culture which consists of *Lb. delbrueckii subsp. Bulgaricus* and *S. Thermophilus* were then inoculated. Bacterial fermentation converts lactose into lactic acid, which reduces the pH of milk. Several changes were observed in the physico- chemical and microbiological characteristics of the raw goat milk when

processed to yoghurt. Eissa *et al.* (2011) showed that the gross composition of fresh goat milk changes after yoghurt processing.

There was an increase in the titrable acidity and decrease in pH of the processed yoghurt this indicates that temperature is one of the most important process parameters in the yoghurt making process. A pH of less than or equal to 4.6 is an indication of end point of fermentation in yoghurt making according to Chandan and O'Rell (2006). El Zubeir *et al*. (2012) reported a pH range 4.3 – 6.0 for goat milk yoghurt; this is in agreement with the pH of goat yoghurt in this study. Titrable acidity is a measure of the number of acid molecules present; acidity of yoghurt is as a result of lactic acid bacteria fermentation which converts lactose to lactic acid (Lee and Lucey, 2010). An increase in the titrable acidity and decrease in pH of goat milk yoghurt was reported by Bozanic *et al*. (1998), Viscosity of yoghurt is influenced by the composition of the raw milk, incubation temperature and the activity of the lactic acid bacteria during fermentation which contributes to the higher consistency of the yoghurt (Chandan, 2004; Lucey and Singh, 1997; Walstra, 1998; Tamime and Robinson, 1999). Viscosity of yoghurt is also affected by the level of heat treatment; an increase in milk heating temperature resulted in an increase in apparent viscosity of stirred yoghurts (Lee and Lucey, 2006). Pasteurization temperature, incubation temperature and time have a significant effect on the viscosity of the yoghurt. The resultant effect of the variation of process parameters for WAD goat yoghurt showed an increase in the viscosity of WAD goat yoghurt, this is in agreement with other findings that reported a higher viscosity in stirred yoghurts incubated at lower temperatures (<40 °C) compared to yoghurts incubated at high temperature (>40 °C) (Beal *et al*., 1999; Martin *et al*., 1999; Sodini *et al*., 2004; Lee and Lucey, 2006).

The change in the rheological properties of the yoghurt is determined by the amount of protein and lipids of the yoghurt. Higher protein or lipid significantly improved the rheological properties of the yoghurt (Rodriguez *et al.,* 2008). At constant pasteurization temperature, an increase in incubation temperature and time resulted in low fat content value and an increase in the protein content of the goat milk yoghurt. This is in agreement with the findings of Koestanti and Romziah (2008), they reported a decrease in the fat content of fresh goat milk and an increase in the protein content when processed to yoghurt, the decrease in fat content could be as a result of lipid breakage during fermentation while the increase in protein content in yoghurt could be as a result of the proteolytic activity of lactic acid bacteria, which hydrolyses proteins (caseins) into peptides and amino acids (Thomas and Mills, 1981). Ehirim and Onyeneke (2013) also reported a higher value for protein content of goat milk yoghurt.

Total solids content is an important quality parameter for yoghurt, in an attempt to prevent syneresis most yoghurt producers increase the total solids contents to (14 to 16%) or by adding stabilizers like pectin and gelatin (Lucey *et al.,* 1998, Amatayakul *et al.,* 2006).

The effect of processing parameters on the totals solid content of WAD goat milk yoghurt shows an increase in the total solid content of the yoghurt, the increase in total solid contents could be due to loss of moisture. Damunupola *et al*. (2014) reported higher total solids (23.56%) in goat yoghurt. Weaver (1993) in their study reported that a low percentage of total solids in yoghurt can lead to malfunctions of the starter culture.

The main and quadratic effect of pasteurization temperature, and the main and interaction effect of incubation temperature and time significantly affected the total plate count, while

the main, interaction effect of incubation temperature and time, and quadratic effect of pasteurization temperature, incubation temperature and time significantly affected the fungal count for WAD goat milk yoghurt when compared to the mean values obtained in the goat milk before pasteurization. This could be due to the combined effect of high heat treatment of milk and the suppressive effect of the used LAB culture during the manufacture of yoghurt which associated with their ability to produce some of acidity and antimicrobial compounds (Abd El-Aty *et al.,* 1998). There was no coliform growth detected in all the samples analyzed. Mac Graw (1997) reported that processed milk should contain no trace of coliform. The absence of coliform is a good indication of the Good Manufacturing Practices employed during the process.

The lactic acid bacteria count plays an essential role in the production of yoghurt; an increase in the lactic acid bacteria count was observed in this research. The increase in available nutrients from caseinate or whey proteins may partially influence the growth of yoghurt bacteria (Amatayakul *et al*., 2006). Tamime and Robinson (1999) reported that yoghurt should contain 10^7 viable cells of lactic acid bacteria per milliliter.

The sensory acceptability result indicates that consumer acceptability of yoghurt was significantly affected by the source of milk used. It was observed that there were significant differences in the degree of likeness of taste, aroma, mouthfeel and overall acceptability of WAD goat milk yoghurt when compared with cow milk yoghurt. WAD Goat milk yogurt was the least accepted while cow milk yoghurt was highly accepted. Goat milk has a "goaty smell" this might attribute to the low acceptable scores. Cow milk yoghurt had a higher sensory acceptability score than WAD goat yoghurt and this finding

is in agreement with what was reported by Eissa *et al.* (2010) who found out that cow yoghurt had better sensory scores compared to goat yoghurt.

5.1 Conclusions and Recommendation

This research work have revealed that the physico-chemical and microbiological qualities of WAD goat milk yoghurt was significantly affected by the pasteurization temperature, incubation temperature and time.

The optimized solution for process parameters for WAD goat milk yoghurt are pasteurization temperature (84.24 ^{o}C), incubation temperature (44.22 ^{o}C) and incubation time (3.80 h). The results obtained for the physico-chemical and microbiological quality of the optimized WAD goat milk yoghurt and the control (cow milk yoghurt) show that WAD goat milk yoghurt was significantly different from cow milk yoghurt and has higher mean values in terms of its total solid content, titrable acidity, fat content, protein content, total plate count and fungal count, while the control had a higher value in its viscosity, lactic acid bacteria count and pH.

The sensory evaluation result also shows that Cow milk yoghurt which was taken as the reference in this study had a higher sensory acceptability score than WAD goat milk yoghurt. The overall liking of WAD goat milk yoghurt by sensory panelists indicates that yoghurt processed from it may be accepted if the goat like aroma is removed.

From the economic point of view there is a possibility for the use of WAD goat milk for processing yoghurt, it is therefore recommended that the milk yield of WAD goat milk is increased by improving its feed ration in order to have an ample supply of milk for commercial scale yoghurt production.

Addition of flavouring compounds to goat's milk products is highly recommended for the unacceptable aroma of goats' milk.

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