

Sudan University of Science and Technology
College of Graduate Studies



**The Effect of Acetic Acid Treatment on some Quality
Properties of Chicken Breast during Refrigeration**

**تأثير المعاملة بحمض الخل في بعض صفات جودة صدر الدجاج الطازج
خلال التخزين المبرد**

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الآية

قال تعالى:

(وَلَحْمِ طَيْرٍ مِّمَّا يَشْتَهُونَ* وَحُورٌ عِينٌ)

صدق الله العظيم

سورة الواقعة الآية (21-22)

Dedication

I would like to dedicate this work to:

My Father

My Mother

My brother and my sisters

and to all my friends.

Layla

Acknowledgements

First of all I would like to express my thanks to my God to give me health and help me complete this study and supervisor **Prof. Mukhtar Ahmed Mukhtar** for his help and advice for helpful assistance ,patience and supervision during this work.

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Abstract

This study was conducted at Sudan University of Science and Technology, College of Agricultural Studies, Department of Animal Production and National Food Research Center (NFC) at Shabbat, in order to study the Possibility of prolonging the shelf life of chicken meat by Packaging and treating with acetic acid concentration 1%, 2% and 3% for 30 Seconds.

Samples were preserved at 4 ± 1 °C for 21 days and examined after 0, 7, 14 and 21 days of refrigeration for microbiological, chemical and sensory Properties.

Microbiological analyses included determination of total count of Bacteria, (*Salmonella*, *E. coli*, *Staphylococcus*), (anaerobic bacteria), yeasts and molds. Chemical analysis included determination of pH value and non-protein nitrogen. However Sensory properties were included for color, smell and texture. Results revealed that samples immersed into a solution with 1%, 2% and 3% of acetic acid concentrations had affectively improved quality and sensory properties of chicken meat for more than 14 days compared with control samples and the samples treated with 3% of acetic acid recorded the best quality results.

ملخص البحث

أجريت هذه الدراسة في جامعه السودان للعلوم والتكنولوجيا -كلية الدراسات الزراعية- قسم الإنتاج الحيواني و المعمل القومي لبحوث الأغذية لدراسة إمكانية إطالة مدة التخزين للحوم الدواجن المحفوظة في درجة حرارة 4⁰م لمدة 21 يوم والمعاملة بحمض الخليك بتركيز (1%، 2%، 3%) لمدة 30 ثانية تم اختبار العينات في مدة تتراوح بين 0، 7، 14، 21 يوم في التبريد وأجريت الاختبارات الميكروبية والكيميائية والحسية. تضمن التحليل الميكروبي تقييم العد الكلي للبكتيريا (السالمونيلا -ايكولاي-ستفايلوكوكس-البكتيريا غير الهوائية-خمائر والاعفان).

والتحليل الكيميائي تضمن تقييم الـ pH الازوت الغير بروتيني بينما تضمن التحليل الحسي تقييم اللون -القوام-النكهة. أظهرت النتائج أن كل العينات التي تم تغطيسها في المحلول بتركيز (1%، 2%، 3%) من حمض الخليك المركز كانت فعالة في تحسين الجودة وخصائص التخزين لمدة أكثر من 14 يوم مقارنة مع العينات القياسية. بينما العينات المعاملة بتركيز 3% من حمض الخليك سجلت أفضل صفات الجودة.

CHAPTER ONE

INTRODUCTION

Poultry production increased in the last five years and it's principality located in Khartoum state (it is the capital of Sudan), due to an increase in demanded for poultry products.

Poultry meat is very popular food commodity around the world due to its low cost of production (Barbut, 2002, Patsias *et al*, 2008).Poultry meat and eggs are highly nutrition's .the meat is rich in proteins and is a good source of phosphorus and other minerals, and of b-complex vitamins. Poultry meat contains less fat than most cuts of beef. It has a higher proportion of unsaturated fatty acids than saturated fatty acids (Bourre, 2005; FAO,2009).

Poultry meat being a nutrient dense food product is therefore highly susceptible to spoilage from microbes which may occur in two ways during refrigeratin .microbial growth and oxidative rancidity (Sebranek *et al*.2005,Jenny,2011), causing quality defects such as off-lover, off-odor etc (Jayasenaet al.2013).the extending the shelf life of perishable chicken products is a major concern for the poultry industry (Wang *et al*,2004). So a variety of physical preservation techniques (Zhou, Xuand liu,2010) as well as chemical preservation on techniques are used to preserve poultry meat (James and Jay, 2000).

Chlorine, organic acids, inorganic phosphates, organic preservatives, bacteriocins and oxidizers are the most frequently used for decontamination of animal carcasses (Bolder, 1997, Aculf, 2005).

Acetic acid is amonocarboxylic acid with a pungent odour and taste known as vinegar which has antimicrobial capabilities due to its ability to lower the

phand cause instability of bacterial cell membrane (Ransom *et al.*,2003) reduced salmonella population or incidence (Tomblynant Conner 1997 a,b).

The acetic acid is generally recognized as safe substance with no upper limit of daily intake for humans (FAO, 1965).

This study was carried out to evaluate the effectiveness of acetic acid on the physiochemical, microbial sensorial properties and the effects of storage periods on the quality characteristics of chicken meat enriched with acetic acid.

The specific objectives of the current work are to study:

1. To produce healthier fabricated chicken meat products.
2. To study the physiochemical, microbial and sensorial properties of chicken meat.
3. To evaluate the effects of storage periods on the quality characteristics of chicken meat enriched with acetic acid consistent basis to achieve optimum performance .

CHAPTER TWO

LITERATURE REVIEW

2.1 Meat definition

FSANZ (2002) defined meat as the whole or part of any buffalo, cattle, deer, pig, poultry, rabbit or hare slaughtered other than in a wild state. This definition does not include eggs or fetuses. The term, meat refers only to meat flesh (skeletal muscle plus any attached muscle connective tissue or fat), but the FSANZ definition also includes offal's (i.e. meat other than meat flesh, including brain, heart, kidney, liver, pancreas, spleen thymus, tongue and tripe), and excludes bone marrow. White meat Chicken meat.

Consumption

Poultry meat production worldwide approached 94.7 million metric tons (MT) in 2009 (FAO, 2009). Currently the US, China, Brazil and European Union (EU) with 19.4, 12.1, 11.3, and 8.5 million MT respectively, are the primary broiler producers (USDA, 2009a). In 2008 the amount of poultry production in Canada reached 1.2 million MT (Statistics Canada, 2009). Based on their solubility function, proteins in skeletal muscle have been categorized into sarcoplasmic, myofibrillar, and stromal proteins (Strasburg *et al.*, 2008). Sarcoplasmic proteins include proteins located in the sarcoplasm (cellular fluid) of the myofiber including myoglobin, hemoglobin, cytochromes, glycolytic enzymes and creatine kinase. These proteins are also called 'water soluble' proteins. This fraction constitutes about 30% of the total muscle protein content (Scopes, 1970).

Myofibrillar proteins include 50-60% of muscle proteins. These proteins are salt soluble and thus they are called 'salt soluble' proteins. Myosin and actin which are categorized in this group are thick and thin filaments, respectively (Strasburg *et al.*, 2008). Stromal proteins, which comprise 10-20% of total

muscle protein content, provide strength and protection for muscle tissue. The composition and abundance of these kinds of proteins greatly affect the quality of meat products. The major protein of this group is collagen (Strasburg *et al.*, 2008).

2.2 Structure of poultry meat muscle

The skeletal muscle is a complex structure composed of individual muscle fibers. A single skeletal muscle is surrounded by the epimysium, which is a thin layer of connective tissue extending. Each muscle is composed of muscle fiber bundles, which is covered by the perimysium, another thin layer of connective tissue. In turn, each muscle fiber bundle is composed of individual muscle fibers, which is covered by another membrane of connective tissue, the endomysium. Each muscle fiber consists of myofibrils, which are made up of my filaments, actins (thin filament) and myosin (thick filament) (Cassens, 1994). The overlapping arrangement of my filaments results in dark (A) and light (I) bands. The A band is the area in which actins and myosin overlap. The area in the A band which contains no thin filaments is the H zone while I band is the area which contains no thick filaments (Feiner, 2006). I bands are bisected which results in dark lines known as Z-lines, while bisected A bands are known as M-lines (Toldrá, 2002). The contractile unit of a muscle fiber is the sarcomere, which is located between two Z lines and is approximately 2.5 μm long. Actins and myosin are connected to the Z line and M line, respectively. Muscle fibers have a striated appearance due to the special arrangement of actins and myosin. My filaments are attached to the cell membrane called the sarcolemma, which has a net-like structure. Muscle fibers are filled with intracellular substance, sarcoplasm (cellular fluid), which is a liquid composed of approximately 80% water as well as proteins, enzymes, lipids, carbohydrates, and inorganic constituents (Aberle *et al.*, 2001)

2.3 Nutritive value of chicken meat

Chicken meat is an excellent source of protein and can be produced on most small and backyard farms. FAO 2009, reported that poultry meat accounts for 30% of global meat consumption. Poultry meat and eggs are highly nutritious being a rich source of proteins, phosphorus and other minerals, and of B-vitamins.

Protein

Based on their solubility function, proteins in skeletal muscle have been categorized into sarcoplasmic, myofibrillar, and stromal proteins (Strasburg *et al.*, 2008). Sarcoplasmic proteins include proteins located in the sarcoplasm (cellular fluid) of the myofiber including myoglobin, hemoglobin, cytochromes, glycolytic enzymes and creatine kinase. These proteins are also called 'water soluble' proteins. This fraction constitutes about 30% of the total muscle protein content (Scopes, 1970). Myofibrillar proteins include 50-60% of muscle proteins. These proteins are salt soluble and thus they are called 'salt soluble' proteins. Myosin and actin which are categorized in this group, are thick and thin filaments, respectively (Strasburg *et al.*, 2008). Stromal proteins, which comprise 10-20% of total muscle protein content, provide strength and protection for muscle tissue. The composition and abundance of these kinds of proteins greatly affect the quality of meat products. The major protein of this group is collagen (Strasburg *et al.*, 2008).

2.3.2 Fat and fatty acid

Poultry meat contains less fat than most cuts of beef and pork. Poultry liver is especially rich in vitamin A. It has a higher proportion of unsaturated fatty acids than saturated fatty acids. This fatty acid ratio suggests that poultry may be a more healthful alternative to red meat (FAO, 2009).

2.3.3 Vitamins and minerals

Thirteen vitamins and thirteen minerals are required by poultry to maintain bodily functions and to prevent growth deficiencies (Austic and Nesheim, 1990). These vitamins and minerals influence the final poultry product by aiding in important metabolic processes. Minerals aid in skeletal formation, hormone functionality, enzyme activation, and in regulation of osmotic pressure in the body of birds (Scanes *et al.*, 2004). Vitamins are only required in minute amounts to support normal growth, reproduction and health. A common practice among poultry producers is to supply various vitamins and minerals in excess of the minimum recommended amounts established by the NRC to account for degradation during storage (Austic and Nesheim, 1990). Calcium (Ca) and phosphorus (P) are integral minerals involved in bone strength. Zinc (Zn) redistribution occurs during times of immunological stress and is critical for maintenance of cells involved in the immune response (Bartlett and Smith, 2003). Supplementation of poultry feeds with vitamin E as a preventative measure towards the oxidation of unsaturated fatty acids resulted in a significant increase of α -tocopherol levels in breast and thigh meat ($P < 0.05$) (Nam *et al.*, 1997). Furthermore, increased thiamin levels in feeds significantly ($P < 0.05$) increased 28-d live weights of birds and improved feed conversion rates (Hulan *et al.*, 1980).

2.3. 5 Carbohydrate content

Starch is a major component in poultry feedstuffs and supplies over 50% of the apparent ME in the diet (Aar *et al.*, 2003). Energy, or kilocalories, not supplied by fat or protein in poultry feed is provided by carbohydrates.

Carbohydrates are quickly metabolized by broilers as energy to maintain body temperature and all basal processes (Scanes *et al.*, 2004).

2.3.6 Pigments

The characteristic chicken color is contributed by different meat pigments. Those pigments includes meat hemoglobin, myoglobin and cytochrome (Aberle *et al.*, 2001).

2.3.7 Enzymes

Enzymes as special proteins that catalyse or accelerat the rate of specific chemical reactions in which the enzyme activity may be dependent on the substrate in a random manner or it may be through very specific sites on substrates such as fat, protein, or carbohydrates (Ferket, 1993). In non-ruminantsdiets, exogenous enzymes are used to improve digestibility of a wide range of feed components such as fibre, phytate, protein, etc. Fibre-degrading enzymes are used to break down specially non-starch polysaccharides (NSP), which are large polymers, tosmaller polymers to alleviate their anti-nutritive activities (Choct and Annison, 1992). This is reflected in better flock performance, better litter quality and improved bird health, which in turn, has a positive influence on total production costs (Saleh *et al.*, 2005).

2.4 Quality characteristic of chicken meat

Meat quality is a term used to describeth the overall meat characteristics includingits physical, chemical, morphological, biochemical, microbial, sensory, technological,hygienic, nutritional and culinary properties (Le, 2006). Appearance, texture, juiciness,wateriness, firmness, tenderness, odor and flavor are among the most important andperceptible meat features that influence the initial and final quality judgment by consumers before and after purchasing a meatproduct (Jaczynski and Park, 2006).Furthermore, quantifiable properties of meat such as water holding capacity, shear force, drip loss,cook loss, pH, shelf life, collagen content, protein solubility, cohesiveness, and fat binding capacity are indispensable for processors

involved in the manufacture of value-added meat products (Le, 2006). Raw meat used in further processed products is required to have excellent functional properties that will ensure a final product of exceptional quality and profitability. However, despite their importance, the poultry grading system used worldwide continues to be based on aesthetic attributes such as conformation, presence or absence of carcass defects, bruises, missing parts, and skin tears without taking into account the functional properties of meat (Jaczynski and Park, 2006). Consequently, this grading system has not been beneficial for the further processing industry that is for the most part interested in the functional properties of meat (Le, 2006).

2.4.1 Color

Color is an important meat quality which greatly affects consumers' preference (Froning, 1995). Total heme pigments including myoglobin and hemoglobin are responsible for the color of meat. The meat pigment is mainly myoglobin because hemoglobin, which is the blood pigment, will be mostly removed after the slaughter. Therefore, myoglobin is the determining factor for the meat color, and variations in meat color indicate the differences in myoglobin content. For example, poultry breast and thigh which are known as white and dark meat respectively easily can be differentiated from each other (Schwartz *et al.*, 2009).

2.4.2 Flavor and Taste

Flavor is a complex sensation. It involves odor and taste. Of these, odor is the most important. Without it, one of the four primary taste sensations, bitter, sweet, sour or saline-predominates (Lawrie, 1991).

2.4.3 Tenderness

According to the International Organization for Standardization, texture of a food is defined as the rheological and structural attributes of a food product which is perceived by human senses (ISO, 1992). Texture of meat is an

attribute that is determined by several factors such as hardness, springiness, chewiness, and cohesiveness. Differences in meat texture are related to the composition and structure of the meat including different kinds of proteins as well as fat and connective tissue. Some other factors such as cooking also affect meat texture (Solomon *et al.*, 2009).

2.4.5 Water Holding Capacity (WHC)

The water holding capacity (WHC) is the ability of meat to retain its water or added water during application of external forces such as cutting, heating, grinding or pressing (Lawrie, 1991). NPPC (2002) defined water holding capacity (WHC) the ability of muscle to retain naturally occurring moisture, and generally expressed as drip loss or purge. Water holding capacity is important in meat processing because it affects many of the physical properties of meat products, such as color, texture, juiciness and tenderness. This ultimately will affect the overall product palatability (Brewer, 2004).

2.5 Acetic acid

The word vinegar comes from the French word - vinaigre which means -sour wine. It was probably discovered by accident thousands of years ago - a cask of wine had gone bad. When the wine was first made, natural sugars were fermented into alcohol (Morales, 2003). Over time; bacteria in the air transformed the alcohol into acetic acid, which gave the - sour wine its bite (Ebner, 1982).

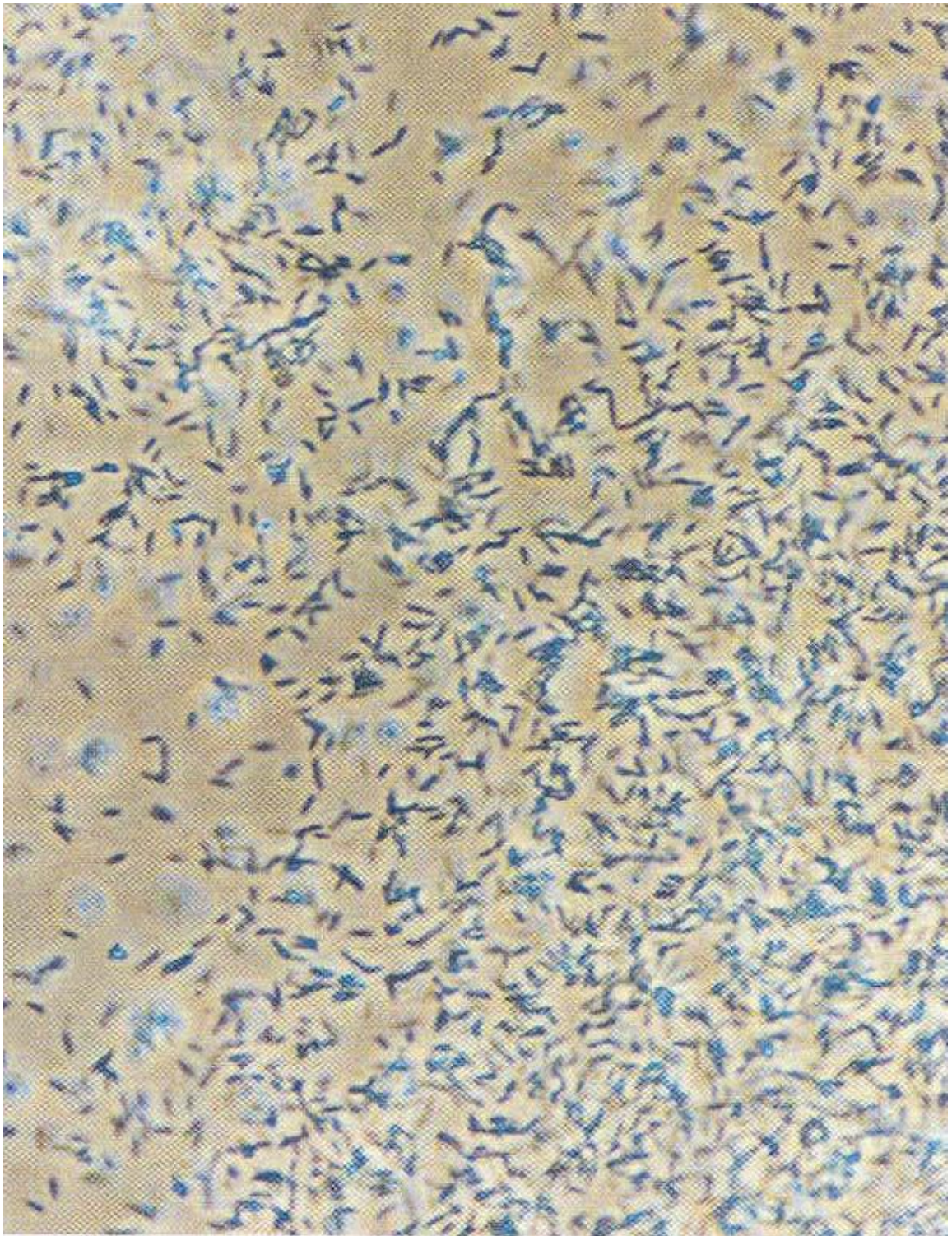


Fig 1: Acetic Acid Bacteria, Picture Provided by (Frings Company, 2005)

2.5.1 Vinegar History

Vinegar is the world's oldest cooking ingredient and food preservation method. According to the Vinegar Institute (Vinegar Institute, 2005), vinegar's use can be traced back over 10,000 years. In fact, flavored vinegars have been manufactured and sold for almost 5,000 years. The wide variety of vinegars available today is nothing new. Until the sixth century BC, the Babylonians were making and selling vinegars flavored with fruit, honey, malt, etc. to gourmets of the time. In addition, the Old Testament and Hippocrates recorded the use of vinegar for medicinal purposes (Conner and Allgeier 1976).

2.5.2 Processing of vinegar

Vinegar is made by fermenting ethanol which produces acetic acid. The ethanol that is used in the process can be derived from wine, beer, fermented fruit juice or cider (Morales, 2003). Along with acetic acid, vinegar has amounts of tartaric acid, citric acid, and other acids. There are various kinds of vinegar including malt vinegar, wine vinegar, apple cider vinegar, fruit vinegar, balsamic vinegar, rice vinegar, coconut vinegar, palm vinegar, cane vinegar, and raisin vinegar (Fings Company, 2005). Acetic acid fermentation, it is an aerobic biological oxidation process which is thermodynamically favorable. The substrate with an ethanol concentration of 50-100 g/l is partially oxidized by the acetic acid bacteria to produce acetic acid and water (Morales, 2003). The result of transformation of the ethanol to acetic acid— the stoichiometry for the conversion of substrate into product is 1:1, and low residual quantities of non-converted ethanol and moreover, a wide variety of secondary compound (Fings Company, 2005). Vinegar has been made from different sources of derived ethanol i.e. wine, cider, beer, fermented fruit juice or it may be made synthetically from natural gas and petroleum derivatives (Morales, 2003). The traditional balsamic vinegar is a natural product prepared from grape must. It contains

polyphenol compound which shows the antioxidant activity (Morales, 2003). In Japan, two traditional rice vinegars i.e. Komesu and Kurosu are produced by a traditional static fermentation process. The Komesu is produced from polished amber rice and Kurosu, which is unpolished black rice vinegar. These vinegars are known for their health benefits via the prevention of inflammation and hypertension (Ebner, 1982). In acetic acid fermentation most of the important physical parameters which affect the growth of the *A. aceti* in fermentation are temperature and pH. It is believed that at lower pH of wine inhibits the *A. aceti*'s growth. It also found that cell numbers of *A. aceti* decrease faster at pH- 3.4 than at pH-3.8 under strict anaerobic condition (Morales, 2003). The optimum pH for the growth of *A. aceti* is pH- 5.5-6.3 (Fings Company, 2005). The temperature 25-300 C is the optimum for *A. Aceti*'s growth, the thermotolerant *A. aceti* are able to grow at 37-400 C. It has been observed that *A. aceti* could not grow below 80 C (Ebner, 1982).

2.5.3 Type of vinegar

The predominant type of vinegar in the United States is white or distilled vinegar. Vinegar is usually described in terms of grain strength, the grain being ten times the acid percentage. For example 10% acid is referred to as 100 grain (Morales, 2003). According to the Crisco Company, vinegar varieties vary greatly from country to country. Some of the most popular vinegars and their characteristics are shown below (Fings Company, 2005):

Balsamic vinegar Balsamic vinegar is brown in color with a sweet-sour flavor. It is made from the white Trebbiano grape and aged in barrels of various woods. Some gourmet Balsamic vinegars are over 100 years old.

Cane vinegar is made from fermented sugarcane and has a very mild, rich-sweet flavor. It is most commonly used in Philippine cooking.

Champagne vinegar has no bubbles. It's made from a still, dry white wine

made from Chardonnay or Pinot Noir grapes (both of which are used to make Champagne).

Cider vinegar is made from apples and is the most popular vinegar used for cooking in the United States.

Coconut vinegar is low in acidity, with a musty flavor and a unique aftertaste.

Distilled vinegar is harsh vinegar made from grains and is usually colorless. It is best used only for pickling.

Malt vinegar is very popular in England. It's made from fermented barley and grain mash, and flavored with woods such as beech or birch. It has a hearty flavor and is often served with fish and chips.

Rice wine vinegar has been made by the Chinese for over 5,000 years. There are three kinds of rice wine vinegar: red (used as a dip for foods and as a condiment in soups), white (used mostly in sweet and sour dishes), and black (common in stir-fries and dressings).

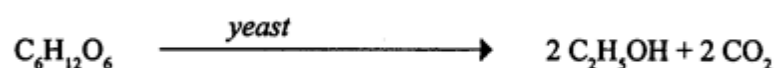
Sherry vinegar is aged under the full heat of the sun in wooden barrels and has a nutty-sweet taste.

Wine vinegar can be made from white, red, or rose wine. These vinegars make the best salad dressings.

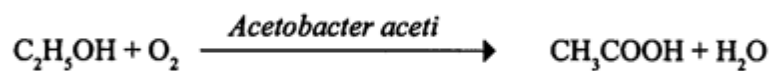
2.5.4 Physical and chemical properties

The physical and chemical properties of vinegar reflect the fact that vinegar is mainly a dilute aqueous solution of acetic acid. This acid liquid which we call vinegar, is the product of two biochemical processes:

1. Alcoholic fermentation, which converts natural sugars into alcohol



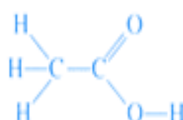
Acid fermentation in which acetobacter, microorganisms present in the air we breathe, converts the alcohol into acid.



it is acid, which imparts the sour taste to vinegar along with its cleaning and antiseptic or germ killing properties (Morales, 2003). Of course most vinegars are much more than dilute solutions of acetic acid. Depending on the fruit or feed stock they are made from, and the amount of processing, they will contain various amounts of minerals, vitamins, fiber, enzymes and other organic compounds (Morales, 2003). These are all however, minor components in the vinegar even though they are major contributors to the product's flavor and aroma as well as its overall nutrition and health benefits (Fings Company, 2005).

2.5.5 Chemical Formula for Vinegar

As far as chemical reactions are concerned, vinegar is a dilute solution of acetic acid, so it has the same chemical formula as acetic acid. A molecule of acetic acid contains two carbon, four hydrogen and two oxygen atoms which is often written as **CH₃COOH** to reflect its actual molecular structure (Morales, 2003):



2.5.6 pH of Vinegar

The term "**pH**" is derived from "potential hydrogen" and refers to the amount of hydrogen ions present in solution. Mathematically, pH is equal to the negative logarithm (base 10) of the hydrogen ion concentration in moles per liter, so if the pH of a solution decreases by 1 pH unit then its hydrogen ion concentration increases by ten times. Pure water has a pH of 7 and is neutral whereas anything with a pH less than 7 is acidic and anything with a pH greater than 7 is basic. The pH of vinegar depends upon how much acid is

present, but most commercial distilled white vinegars contain 5% acetic acid and have a pH of about 2.4 ().

2.5.6 Health Benefits of Acetic acid

2.5.6.1 Increasing Calcium Absorption

Acetic acid, like other acids, can increase the body's absorption of important minerals from the foods we eat. Therefore, including apple cider vinegar in meals or possibly even drinking a mild tonic of vinegar and water (up to a tablespoon in a glass of water) just before or with meals might improve your body's ability to absorb the essential minerals locked in foods. Vinegar may be especially useful to women, who generally have a hard time getting all the calcium their bodies need to keep bones strong and prevent the debilitating, bone-thinning disease osteoporosis. Although dietary calcium is most abundant in dairy products such as milk, many women (and men) suffer from a condition called lactose intolerance that makes it difficult or impossible for them to digest the sugar in milk. As a result, they may suffer uncomfortable gastrointestinal symptoms, such as cramping and diarrhea, when they consume dairy product (FDA, 2009).

2.5.6.2 Controlling Blood Sugar Levels

Vinegar has recently won attention for its potential to help people with type 2 diabetes get a better handle on their disease. Improved control could help them delay or prevent such complications as blindness, impotence, and a loss of feeling in the extremities that may necessitate amputation. Also, because people with diabetes are at increased risk for other serious health problems, such as heart disease, improved control of their diabetes could potentially help to ward off these associated conditions, as well. With type 2 diabetes, the body's cells become resistant to the action of the hormone insulin. The body normally releases insulin into the bloodstream in response to a meal. Insulin's job is to help the body's cells take in the glucose, or sugar, from the

carbohydrates in food, so they can use it for energy (Johnston and Gaas 2006).

2.5.6.3 Replacing Unhealthy Fats and Sodium

There are some delicious varieties of vinegar available. Each bestows a different taste or character to foods. The diversity and intensity of flavor are key to one important healing role that vinegar can play. Whether you are trying to protect yourself from cardiovascular diseases, such as heart disease, high blood pressure, or stroke, or you have been diagnosed with one or more of these conditions and have been advised to clean up your diet, vinegar should become a regular cooking and dining companion. That's because a tasty vinegar can often be used in place of sodium and/or ingredients high in saturated or trans fats to add flavor and excitement to a variety of dishes (Fushimi *et al.*, 2006). Saturated and trans fats have been shown to have a detrimental effect on blood cholesterol levels, and experts recommend that people who have or are at risk of developing high blood pressure cut back on the amount of sodium they consume. So using vinegar as a simple, flavorful substitute for these less healthful ingredients as often as possible can help people manage blood cholesterol and blood pressure levels and, in turn, help ward off heart disease and stroke. We'll find detailed advice about including more vinegar in your diet in chapter four, and you'll discover delicious, good-for-you recipes at the end of the book that put vinegar to use (Fushimi *et al.*, 2006).

2.5.6.4 Making a Healthy Diet Easier to Swallow

Some of our strongest natural weapons against cancer and aging are fruits and vegetables. The antioxidants and phytochemicals they contain seem to hold real promise in lowering our risk of many types of cancer. Their antioxidants also help to protect cells from the free-radical damage that is thought to underlie many of the changes we associate with aging. Protected cells don't

wear out and need replacing as often as cells that aren't bathed in antioxidants (Liljeberg, 1998).

2.5.6.5 Removing Harmful Substances from Produce

Some people are concerned that eating large amounts of fruits and vegetables may lead to an unhealthy consumption of pesticide and other farm-chemical residues. Vinegar can lend a hand here, too. Washing produce in a mixture of water and vinegar appears to help remove certain pesticides, according to the small amount of research that has been published. Vinegar also appears to be helpful in getting rid of harmful bacteria on fruits and vegetables (Liljeberg, 1998).

2.5.6.6 Possible cholesterol and triacylglycerol effects

A 2006 study concluded that a test group of rats fed with acetic acid (the main component of vinegar) had "significantly lower values for serum total cholesterol and triacylglycerol", among other health benefits, rats fed vinegar or acetic acid have lower blood pressure than controls, although the effect has not been tested in humans, reduced risk of fatal ischemic heart disease was observed among participants in a trial who ate vinegar and oil salad dressings frequently (Fushimi *et al.*, 2006)

2.5.6.7 Infections

Vinegar has been used to fight infections since Hippocrates, who lived between 460-377 BC, prescribed it for curing persistent coughs. As a result, vinegar is popularly believed to be effective against infections. While vinegar can be an effective antibacterial cleaning agent on hard surfaces such as washroom tiles and countertops (Johnston and Gaas 2006).

2.6 Microbiology

Meat being a good material for bacterial growth, its quality depend on the initial bacterial contamination. This contamination causes meat deterioration, lower quality, and some time illness may be caused by bacterial pathogens or

their toxins (Jay, 2000). Microorganisms are transferred through direct contact with the hide or indirectly through contact with workers' hands or equipment used, and also via aerosols and dust generated from the hide during removal process (Huffman, 2002).

2.6.1 *Escherichia coli*

E. coli is gram negative, lactose fermenting, facultative aerobic short rod. First documented outbreak of *E. coli* food-borne gastroenteritis occurred in the U.S. in 1971 (Jay, 2000). The first outbreaks of food-borne hemorrhagic colitis in the U.S. was in 1982 (Jay, 2000). *E. coli* 0157:H7 was found to be the cause of two severe outbreaks characterized by hemorrhagic colitis (HC) and hemolytic uremic syndrome (HUS) Rily *et al.*, (1983). The first case of *E. coli* 0157:H7 infection in Italy was reported in 1988. *E. coli* 0157:H7 is one of the enterohemorrhagic *E. coli* (EHEC) serotypes that produce verocytotoxins (VTEC). These pathogen types were identified in 1977 and have been associated with several diseases in both humans and animals (Conedera *et al.*, 1995). *E. coli* 0157:H7 is able to produce toxins which can cause very serious illness in humans, such as HC and HUS (Shapon and Shapon, 1994). The largest recorded food borne outbreak was associated with ground beef, and all raw meat should be considered a possible vehicle for hemorrhagic colitis (Jay, 2000). *Escherichia coli* O157:H7 is commonly found among the intestinal flora of cattle which are the primary reservoir (Shapon and Shapon, 1994). The presence of *E. coli* in meat products indicates fecal contamination of the meat (FAO, 1992).

2.6.2 *Staphylococci aureus*

Staphylococcus aureus is one of the most frequent pathogen that cause food - borne outbreaks. It is responsible for staphylococcal food poison (SEF) by producing heat stable toxin (Shapon and Shapon, 1994). *Staphylococcus aureus* is a major pathogen for humans, ranging in severity from food

poisoning or minor skin infections to severe life-threatening infection (Jawetz *et al.*, 2001).

2.6.3 Salmonella

Microbial contamination of poultry carcasses is a natural result of different procedures necessary to produce retailed products from living birds. Most of bacterial contaminants are non pathogenic; however, poultry are known to harbour a large number of bacteria that are pathogenic to human being (Zhang *et al.*, 2001). Food-borne pathogens have been isolated from processed poultry including salmonella serovars and *S.aureus* which are of the major concern. *S.aureus* as a food poisoning microorganism is considered as a good indicator for inadequate sanitation, less temperature control and the possible presence of enterotoxin-producing strains (Waldroup, 1996). Poultry has been identified as a primary reservoir for these salmonella serovars which are harbored in skin and feathers as well as in the gastrointestinal tract, consequently, salmonella can persist on final raw products. Disease can result when these products handled without good hygienic practices, not properly cooked, and/or subjected to temperature abuse (Zhang *et al.*, 2001). Three human disease syndromes may be caused by Salmonella spp; typhoid fever and Paratyphoid fever which may be transmitted from human to human and human is the only reservoir. In contrast, the third is gastroenteritis which is usually caused by Salmonella enterica serovars which are found in the intestinal tract of both human and animals .

2.7 Non protein

Is term used in animal nutrition to refer collectively to components such as urea, biuret and ammonia which are not proteins but can be converted into protein by microbes in the ruminant stomach.

Due to their lower cost compared to plant and animal proteins their inclusion in diet can result in economic gain, but at too high levels cause a depression in

growth and possible ammonia toxicity (microbes convert to ammonia first before using that to make protein).

Can also be used to artificially raise crude protein values , which are Measured based on nitrogen content as protein is about 16% nitrogen, put , for example, urea is 47% nitrogen .the source of non is typically Chemical feed additive or sometimes chicken waste and cattle

2.8 Processing meat chickens

Chicken are taken directly from the growing farms to the processing plant where they are unloaded from their transport crates or modules, slaughtered, packaged and frozen or chilled, or processed further in to various products prior to packaging and sale to distributors.

Processing plants are very large, highly mechanized operations .much of the improvement in industry's efficiency over the past five decades is due to increasingly automated poultry plants.

For example, in 1962 a typical 6000 bird per hour processing plant employed approximately 300 people from live –bird handling to distribution, whereas today the same plant would employ 100 people .Australia's largest poultry processing establishment kills and processes 33 million birds per year ,or 630,000 birds per week.

All significant poultry processing operations in Australia have a systematic preventative approaches to managing food safety risks, with approved and regularly audited hazard analysis and critical control points (haccp) programs.

(Australian chicken meat 2013).

2.9 Frozen Shelflife;

The shelf life of frozen poultry is influenced by many factors ,as determined by rancidity and off-flavor development or by dehydration of surface areas.

Among the more important variables is freezer temperature, packaging, handling prior to freezing, and type of product.(Paul Dawson)

The consumer pack chicken roasters is 12-15 months and consumer pack chicken broilers, whole is 16 months.

Fresh; according to USDA regulations, to be labeled 'fresh' poultry must never be stored at a temperature of less than 26 F (-3.3°C).

Keep Frozen; when on a poultry product label, the product must be kept frozen at 0 F (-18°C), 10 F (5.5°C) tolerance at all times

Keep Refrigerated; when on a poultry product label, the product must be kept refrigerated at a temperature of less than 40 F (4.4°C), no tolerance.

Product should not be frozen.

Keep Refrigerated or Frozen: When on a poultry product label the product may be either refrigerated, but could be frozen (topic.)

Shelf life of ground poultry meat under modified atmosphere;

The shelf life of ground chicken and turkey meat packaged under a modified atmosphere containing O₂ and high level of CO₂ (62%, 8% O₂ and 30% N₂ gas-2) or gas mixture without O₂ (20% CO₂ and 80% N₂ gas) was evaluated for 20 weeks. At 4°C meat packaged under gas-2 maintained higher value (redness) throughout total aerobic mesophilic counts were higher in chicken meat than in turkey throughout storage. *Coli* forms and *E. coli* counts were lower in meat packaged under gas-1 than meat packaged under mixtures tested. Had a similar account for presumptive pseudomonads, *Staphylococcus aureus*, and lactic acid bacteria. These results indicate that an appropriate gas mixture can maintain a desirable color in ground poultry meat but offers no guarantees with respect to the microbial profile of meat.

CHAPTER THREE

MATERIALS AND METHODS

3.1 materials

The fresh chicken was obtained from Arabic Company at Khartoum and transferred immediately to the Animal Production Department (National Food Research Center NFRC) where it is frozen and kept frozen at $-10 \pm 1^{\circ}\text{C}$. Acetic acid was obtained from the Food Technology and stored at 4°C . Chemicals and reagents used were obtained from Central Lab store of National Food Research Center (NFRC).

3.2 Methods

3.2.1 Raw material preparation

3.2.1.1 Meat preparation

Stored chicken meat was thaw at -3°C over night, sliced to small pieces.

3.2.1.2 Acetic acid preparation

Acetic acid where prepared at three levels 0 %,1%, 2 % and 3 %.

3.2.1.3 Chicken preparation of samples

It was purchased 4 kg chicken breast immediately after slaughter and refrigerated, transported in containers sterilized almost detective then the skin has been removed and the meat is cut in to four units each with the size of 25 grams,each unit was divine in to each of other of acetic acid rate 1%,2%,3° part of them in acetic acid solution concentration of 1% and asection at concentration of 2%and another at aconcentration 3% for 30 seconds ,exceptcontrol remained without treatment and purified chicken pieces of

acetic acid to ensure removed of the remaining of it, then the samples stored in the freezer at temperature $10 \pm 1^\circ\text{C}$ for 21 days.

Analysis is performed microbial, chemical and sensory samples during storage period at 0, 7, 14, 21 - days.

Frozen chicken meat was thawed and cut into small pieces



The spices chicken was divided into four groups (treatments)



The acetic acid was added to different treatment and mixed well



The product was packed in plastic packs and stored in a freezer $4 \pm 1^\circ\text{C}$

Fig 2:Chicken spices enriched acetic acid

3.3.2 pH measurement

This test was carried out according to the method described by A.O.A.C (2003). Ten gram of the samples was placed in blender jar and 100 ml of distilled water were added, the mixture was blended at high speed for 1 min. The pH of the mixture was measured by using a recalibrated pH meter model (HI 8521 microprocessor bench pH / MV / °C meter). This has been calibrated with two standard buffers (6.8 and 4.0).

3.4 Microbial tests

3.4.1 Collection of samples

Samples of chicken were withdrawn from each treatment, kept in sterile containers in ice and transferred immediately to the microbiology laboratory, Faculty of Agric, University of Khartoum.

3.4.2 Sterilization of glassware

Glassware was washed thoroughly, left to dry and sterilized in a hot air oven at 160 °C for at least 3 hours (Harrigan and McCance, 1976). Instruments such as loops, needles, forceps, spoons and Knives were sterilized by flaming directly after dipping in spirit.

3.4.3 Culture media used

3.4.3.1 Nutrient agar (oxoid)

The nutrient agar was used for cultivation of bacteria. Twenty- eight grams of dehydrated nutrient agar were suspended in a liter of distilled water, steamed to dissolve completely, the pH was adjusted to 7.4 then the medium was sterilized by autoclaving at 121 °C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.2 Plate count agar (oxoid)

The plate count agar medium was used to determine total bacterial count. Seventeen and half grams of this media were suspended in a liter of distilled water, dissolved by bringing to boiling with frequent stirring, mixed and distributed into conical flasks sterilized by autoclaving at 121°C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.3 MacConkey broth (oxoid)

The MacConkey broth medium was used for the primary isolation of coliform bacteria. Forty grams of this media were suspended in a liter of distilled water, the medium was distributed in test tubes with inverted Durham tubes, the pH was adjusted to 7.0 and then the medium was sterilized by autoclaving at 121°C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.4 Brilliant green bile lactose broth (oxoid)

The Brilliant green bile lactose broth medium was used to confirm the presence of coliform bacteria by multiple tube technique. Forty grams of dehydrated media were suspended in a liter of distilled water, the pH was adjusted to pH7.4, distributed in the test tubes with inverted Durham tubes and then the medium was sterilized by autoclaving at 121°C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.5 Eoism methylene blue agar (oxoid)

The Eoism methylene blue agar medium was used for the differentiation of *Escherichia coli* and *Aerobacter aerogenes*. Thirty seven and half grams of dehydrated Eoism methylene blue agar were suspended in a liter of distilled water, steamed to dissolve completely, the pH was adjusted to 6.8 and then the medium sterilized by autoclaving at 121 °C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.6 Staphylococcus medium No.110 (oxoid)

A selective *Staphylococcus* medium No.110 was used for isolation and differentiation of pathogenic *Staphylococci*. One hundred and fifty gram of this media were suspended in a liter of distilled water, steamed to dissolve completely, the pH was adjusted to 7.0 and then the medium was sterilized by autoclaving at 121°C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.7 Nutrient broth (oxoid)

The nutrient broth medium was used for the cultivation of microorganisms which are exacting in their food requirements. Thirteen grams of dehydrated nutrient broth were suspended in a liter of distilled water, mixed well, the pH was adjusted to 7.4 and then the medium was sterilized by autoclaving at 121 °C for 15 minutes (Harrigan and McCance, 1976).

3.4.3.8 Selenite broth

The Selenite broth medium was used as an enrichment medium for the isolation of *Salmonella*. Nineteen grams of dehydrated selenite broth were suspended in one liter distilled water, 4 grams of sodium biselenite has been added, and then the medium was sterilized by boiling in a water bath at 100°C for 10 minutes (Harrigan and McCance, 1976).

3.4.3.9 Bismuth sulphite agar

The Bismuth sulphite agar medium was used for the isolation and preliminary identification of *Salmonella*. Fifty two grams of dehydrated Bismuth sulphite agar were suspended in a liter of distilled water, steamed to dissolve completely, the pH was adjusted to 7.0 and then the medium sterilized by boiling in a water bath at 100 °C for 10 minutes (Harrigan and McCance, 1976).

3.4.3.10 Triple sugar iron agar

The Triple sugar iron agar medium was used for the different of *Enterobacteriaceae* according to their ability to fermentation lactose, sucrose, dextrose and to produce hydrogen sulphide. Sixty five grams of dehydrated triple sugar iron agar were suspended in a liter of distilled water, steamed to dissolve completely and then the medium was sterilized by autoclaving at 121 °C for 15 minutes (Harrigan and McCance, 1976).

3.5 Microbial analysis

3.5. 1 Preparation of serial dilution

Thirty grams from each chicken treatment were weighted aseptically in a sterile bottle and then blended with 270 ml sterile distilled water by using an electric blender (Homogenizer MSE). The emulsion was blended for 3 minutes to give 1/10 dilution as described by (Harrigan and McCance, 1976).

3.5.2 Microbial parameters studies

Total viable count was carried out by using the standard plate count method as described by Harrigan and McCance (1976). One ml from the suitable dilution was transferred aseptically into sterile Petri dishes. To each dilution 10-15 ml of (melted and cooled 45°C) plate count agar were added. The inoculums was mixed with medium and allowed to solidify. The plates were then incubated at 37°C for 48 h rs. A colony counter (Quebec colony Counter and Hand Tally) was used to count the viable bacteria.

3.5. 3 Determination of coliform bacteria

3.5.3.1 Presumptive *E.coli* form test

Five tubes each containing nine ml of MacConkey (enrichment medium), fitted with Durham tubes, were inoculated with 0.1 ml from suitable dilutions of chicken samples at 37 °C 48 hrs. Growth and gas production after 24 and

28 hrs were recorded. Gas production constituted a positive test(Harrigan and McCance, 1976).

3.5.3.2 Confirmed *E.coli* form test

All fermentation tubes from the presumptive test showing gas with 24 hrs at 37°C were utilized in the confirmation test. The medium used in this test was Brilliant Green Bile lactose broth BGB. Each tube contained 10 ml of medium fitted with Durham tubes. Presumptive test tubes were transferred to each BGB tubes, and then incubated at 37°C for 48 hrs. Faecal coliform were calculated from the most probable number (MPN) via (MPN) tables (FAO, 1992).

3.5.3.3 Isolation of *E.coli*

For further confirmation of faecal coliform in tubes giving positive reaction on *Escherichia coli* media EC at 44.5°C for 28 hrs were streaked on Eosin Methylene Blue (EMB). Colonies with green metallic shine gave a positive test (Harrigan and McCance, 1976).

3.5.4 *Staphylococcus*

From suitable dilutions of chicken samples, one ml was aseptically transferred to a sterile Petri dish. Fifteen ml of Staphylococcus medium No. 110 were added. The inoculum was mixed with medium and allowed to solidify. Plates were then incubated at 37°C for 48 hours and count was expressed as Colony Forming Unit (CFU) per gram.

3.5.5 Presence of *Salmonella*

Twenty five grams of samples were aseptically weighed and mixed well with 250 ml sterile nutrient broth, then incubated at 37°C for 24 hours. Then 10 ml were aseptically drawn and added to 100 ml selenite broth. The broth was incubated at 37°C for 24 hours. Using a loopfull, streaking was carried out

into solidified Bismuth sulphite agar plates. The plates were incubated at 37°C for 72 hours. Black metallic shine discrete colonies indicated the presence of *Salmonella*. A confirmatory test was carried out by taking a discrete black metallic sheene colonies and subcultured it in triple sugar iron agar tubes (Harrigan and McCance, 1976).

3.6 Sensory evaluation

The panelists were M.Sc. and Ph.D. students of Food Science and Technology Department, College of Agricultural Studies, Sudan University of Science, semi-trained according to the procedure of (Cross *et al.*, (1978). The panelists evaluated the prepared chicken breast samples for color, flavor, taste, texture, juiciness, tenderness, over all acceptability, using a hedonic scale of 7 points (7 extremely like, 1 extremely dislike).

3.7 Statistical analysis

The data collected from the different treatments were subjected to analysis of Variance and whenever appropriate the mean separation procedure of Duncan was employed (Steel and Torrie, 1980). The SAS program (SAS, 2002), was

3.2.1.2 Determination of crude protein;

The crude protein sample was determined using modified kjeldal method described by A O A C (2000) whereby 2g of the samples were transferred into a clean 25ml kjeldal digestion flask. 2g of the catalyst mixture was added and 25ml concentrated H_2SO_4 was also added. The mixture digested for about 5 hours when the pale-blue color appeared. The Content of the digestion flask was transferred to 100cm³ volumetric flask and adjusted to the mark .blank was also prepared on the same way (20cm³ of 2%) Boric acid was transferred into conical flask and 4 drops of mixed indicator were added, a 50cm³ burette was filled with 0.1 m HCL. The distillation assembly was turned on but the steam trap was left opened .the condenser tip was immersed

in to the boric acid, 10ml of blank digest was introduced from the sample introduction cork and the funnel was rinsed with 3ml of 30% NaOH was introduced. The cork was closed after rinsing with 2ml of distilled water and the steam trap was also closed. When the color of the boric acid was changed, the condenser tip was washed with distilled water and the boric acid mixture in the flask was titrated with standard 0.1M HCL until the color disappeared. The procedure was repeated two times with the blank and two times with the sample digests and the averages or the titers were calculated.

Calculation;

$$\text{Nitrogen (\%)} = \frac{\text{vol}(\text{sample-blank}) \text{Hcl} * \text{normality of Hcl} * 0.014 * 100}{\text{Weight of sample}}$$

$$\text{Protein (\%)} = \text{n(\%)} \times 6.38$$

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1. Effect of acetic acid and storage period on chicken meat quality

4.1.1 Chemical composition

4.1.1.1 Non-nitrogen protein (%)

The effect of acetic acid and storage period on the non-nitrogen protein (NNP) contain are shown in Table 1. The NNP of samples decreased ($P > 0.05$) With the progress of storage period and acetic acid concentration. Initially (0 day) Non-nitrogen protein of 3% acetic acid sample was lower (2.37%) than that extended with 1% acetic acid (2.75) or 2% acetic acid (2.58) with the storage progress, 7th day, samples treated with 0%, 1%, 2% and 3% had Non-Nitrogen protein content of 2.36%, 2.44%, 2.41% and 2.16 % respectively. On the 14th day of storage, the NNP contain of these samples (0%, 1%, 2% And 3% acetic acid) continued decrease to 3.59%, 2.22%, 2.20% and 1.95% respectively. The result is in agreement with similar studies using different treatments (Azizih *et al.*, (2013) who reported that the addition of acetic acid (1% and 2%) to breast chicken had decreased the NNP contain (12.36 and increased effectiveness enzyme aldayamnz .in his study result showed that (12.16) compared to control (12.26) on day 0. results in line with that the mentioned that control sample convergent with samples through table .(1)

Nitrogen contain increased in all samples with increasing the storage period agree with Socool, oetterer (2005), because the accumulation of NNP in the middle with increasing duration of storage to the breakdown of protein and free amino acid degrade in to other compounds using total disarmament by the remove main acid by enzyme aldayamnz, and the carboxyl by enzyme aldekkerbokilaz (OzoguI, 2000) it found that the more alkaline the middle

treatment of the samples with acetic acid reduced pH values then reduced effectiveness enzyme aldayamnz, that lead to decrease NNP contain in all samples compared with control.

4.1.2 The effect of acetic acid and storage period on pH of chicken breast

Significant differences ($p \leq 0.05$) were detected among storage period (2). The pH of samples decreased with the progress of storage period and acetic acid concentration. The highest pH value was 5.61 showed at initial time of storage period; it was significantly greater than 7th, 14th and 21th days . With the storage progress, 7daysamples treated with 1%, 2%, and 3%acid had pH values of 5.63, 5.62, 5.22 and 5.71 respectively, continued decrease to 5.58, 5.49, 5.16 and 5.60 respectively also among the different levels of acetic acid, the highest value was 5.66 recorded for untreated sample, however 5.25 was recorded at 3% acetic acid. clearly, increase of acetic acid level lead to decrease in pH value, these may be due to the state of organic acid, minerals, vitamins, fiber, and enzymes. This result is not far from that reported by Harastani *et al.*, (2013) who indicated that concentration of acetic acid 0%, 1% and 2% of breast chicken had 5.75, 5.46 and 5.23pH values respectively. With respect to the treatment combination, the highest pH content 5.71 was recorded for zero % acetic acid at seven day.

However, the lowest value was showed 5.16 was recorded at 3% acetic acid level at twenty one day after processing.(Byrne *et al.*, (2000) stated that meat pH, as affected by post-mortem glycol sis in muscle tissue, has a profound influence on meat quality since it determines traits responsible for the processing suitability and eating attributes of meat. This is also the simplest parameter characterizing the course of post-mortem changes in muscle.

4.1.3: The effect of acetic acid and storage period on sensory Characteristic of breast chicken

The effect enrichment of acetic acid to breast chicken on the sensory characteristic is shown in Table (3), the zero time samples ranked according to appearance as a 0%, 3%, 2%, and 1% acetic acid. These observations agree with (Harastani *et al.*, 2013).

At any storage period, appearance score decreased by increase storage time, for any case, the highest score recorded at zero time 4.62 while, the lowest score recorded after 21 day after processing 4.39, this observation is in on line with (Harastani *et al* .,(2013), who found significant decrease in appearance during storage.

The mean results of flavor are shown in Table (4), generally flavor increased with the increase acetic acid at 3% acetic acid has a higher score followed by 2%, 1% finally 0% acetic acid.

clearly, the flavor scores decrease with the increase in storage period, the highest score was 5.17 which recorded at zero time, but the lowest mean score was 4.97reported after 21 day after processing. Similar finding was observed (Habbal *et al.*,2013).

The panelists detected that the addition of acetic acid decreased juiciness of the chicken breast, 6.54 had the highest score mean of juiciness at all treatments. While the lowest score 3.56 reported at 3%of acetic acid. Within each treatment juiciness decreased with increase in storage period. (Sebsibe (2006) observed that the better juiciness, the lower cooking losses.

Addition of acetic acid to chicken breast resulted in substantial decreases in texture particularly as the level of acetic acid is increased. Numerically 3% had less score texture than the 2%, followed by 1% then 0% acetic acid.

Similar studies using the same experiment, (Azizih *et al.*, (2013) reported that the texture score of chicken breast was 4.90, 4.87 and 4.74 for samples with 0%, 1% and 2% of acetic acid.

Sensory panelists rating for tenderness indicated that chicken breast enrichment with acetic acid at 7 day were more tender than 14 and 21 day after processing. In all cases, the control sample was found to be less hard and juicier than the other treatments.

In the present study, addition of acetic acid to enrich breast chicken resulted in a product as acceptable as that of control.

4.1.4 The effect of acetic acid and storage period on chicken breast

Total viable count

Total viable count of bacteria (TVC) decreased ($P > 0.05$) with the increase of acetic acid concentration. Chicken breast without acetic acid *had* the highest ($P \leq 0.05$) TVC form count ($\log_{10}$ cfu/g) compared to samples treated with acetic acid. The total viable count of bacteria of the control samples measured progressively from 3.590, 3.593, 3.594, and 3.569 ($\log_{10}$ cfu/g) during storage period 0, 7, 14 and 21 days respectively. Obsessively,

Highest mean value 3.586 ($\log_{10}$ c fu/g) was recorded when no acetic acid was added. However, the lowest mean value 3.531 was recorded at 3% acetic acid

Stated that the growth of TVC bacteria decreased with the increase of acetic acid concentration from 4.57 ($\log_{10}$ c fu/g) to 2.61($\log_{10}$ c fu/g), but increase with the increase of storage period.

4.1.4.1. *E.Coli*

As shown on Table (5).All the treatments had homogeneous variance($P >$

0.05), except it presented at 21 day after processing. Similar finding were latter confirmed by (SSMO, (2008), who mentioned that *E.coli* counts

should be limited to $< 6.8\text{MPN}/100\text{g}$. Harris and Savills (2005) mentioned that *E.coli* is the best indicator of fecal contamination or state hygiene.

4.1.4.2 *Salmonella*

The results in Table (6) showed absent of *Salmonella* during the storage time and among the treatments, all these had homogenized variance ($P > 0.05$). These results within Sudanese standard, which mentioned that meat suitable for human consumption, must be *Salmonella* free (SSMO, 2010). Also similar results were obtained by (Harastani *et al*, (2013) who did not detect *Salmonella* in samples under investigation. The presence of salmonella indicates poor food preparation health status (Tompkin, 1994).

4.1.4.3 *Staphylococcus aureus*

Staphylococcus aureus levels are shown on Table (7), all the treatments had Homogeneous variance ($P > 0.05$). These results were closed to that reported by (Sadish (2011), and accordance with the Department of Health (1997), *Staphylococcus aureus* counts should be limited to $<100/\text{g}$.

For the storage period, *Staphylococcus aureus* increased progressively with the time increase, the highest count had 3.099 reported at 14 day of storage.

On the other hand the lowest score reported on 3% acetic acid was 3.071. Then on 2% acetic acid was 3.080 and the last on 1% was 3.096 compared with control 3.110.

Staphylococcus aureus load is less than that reported by (Jalal (2013), these may be due to the effect of treatment. (OFAO (1992) reported that.

4.1.11.5 Yeasts and Moulds growth

Yeast and molds count in Table (8) showed growth at zero time and 21 day no growth during the storage time and among the treatments except at 1% acetic acid.

Table 1: The effect of acetic acid and storage period on chicken breastnon, nitrogen protein

Sample	Storage period (days)				Overall
	0	7	14	21	
A	2.75 ^d ±0.02	2.44 ^f ±0.04	2.22 ^j ±0.02	2.02 ^m ±0.01	2.36 ^B
B	2.58 ^e ±0.03	2.41 ^g ±0.02	2.20 ^k ±0.03	1.83 ^o ±0.02	2.25 ^C
C	2.37 ^h ±0.02	2.16 ⁱ ±0.03	1.95 ⁿ ±0.01	1.74 ^p ±0.00	2.05 ^D
D	3.15 ^c ±0.01	2.36 ⁱ ±0.01	3.59 ^b ±0.01	3.68 ^a ±0.02	3.19 ^A
Overall	2.71 ^A	2.34 ^C	2.49 ^B	2.32 ^D	
Lsd _{0.05}	0.0005259**				
SE±	0.0001826				

Values are means±SD

Mean(s) sharing same superscript(s) in columns and rows are not significantly different (P>0.05) according to DMRT

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

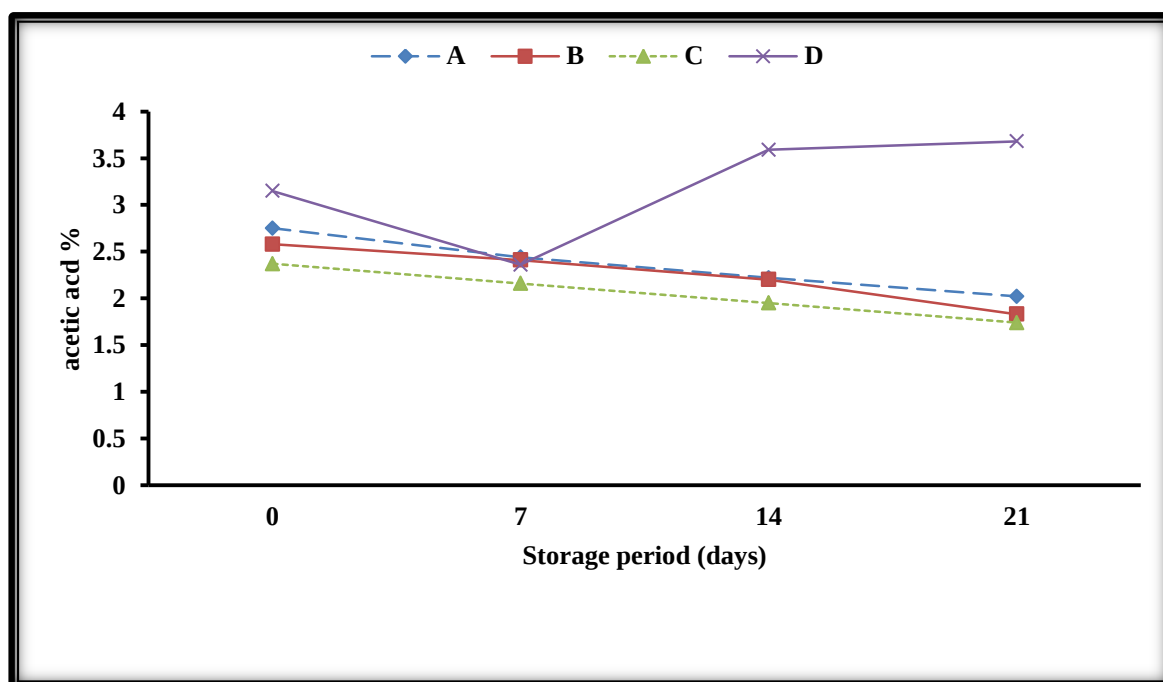


Fig 3: Non nitrogen protein

Table. 2: The effect of acetic acid and storage period on pH of breast chicken

Sample	Storage period (days)				Overall
	0	7	14	21	
A	5.65 ^c ±0.07	5.63 ^e ±0.02	5.59 ^h ±0.02	5.58 ⁱ ±0.04	5.62 ^B
B	5.64 ^d ±0.09	5.62 ^f ±0.03	5.52 ^j ±0.02	5.49 ^k ±0.01	5.57 ^C
C	5.45 ^l ±0.06	5.22 ^m ±0.05	5.18 ⁿ ±0.03	5.16 ^o ±0.02	5.25 ^D
D	5.69 ^b ±0.04	5.71 ^a ±0.01	5.63 ^e ±0.02	5.60 ^g ±0.02	5.66 ^A
Overall	5.61 ^A	5.54 ^B	5.48 ^C	5.46 ^D	
Lsd_{0.05}	0.0005259**				
SE±	0.0001826				

Values are means ±SD

Mean(s) sharing same superscript(s) in columns and rows are not significantly different (P>0.05) according to DMRT

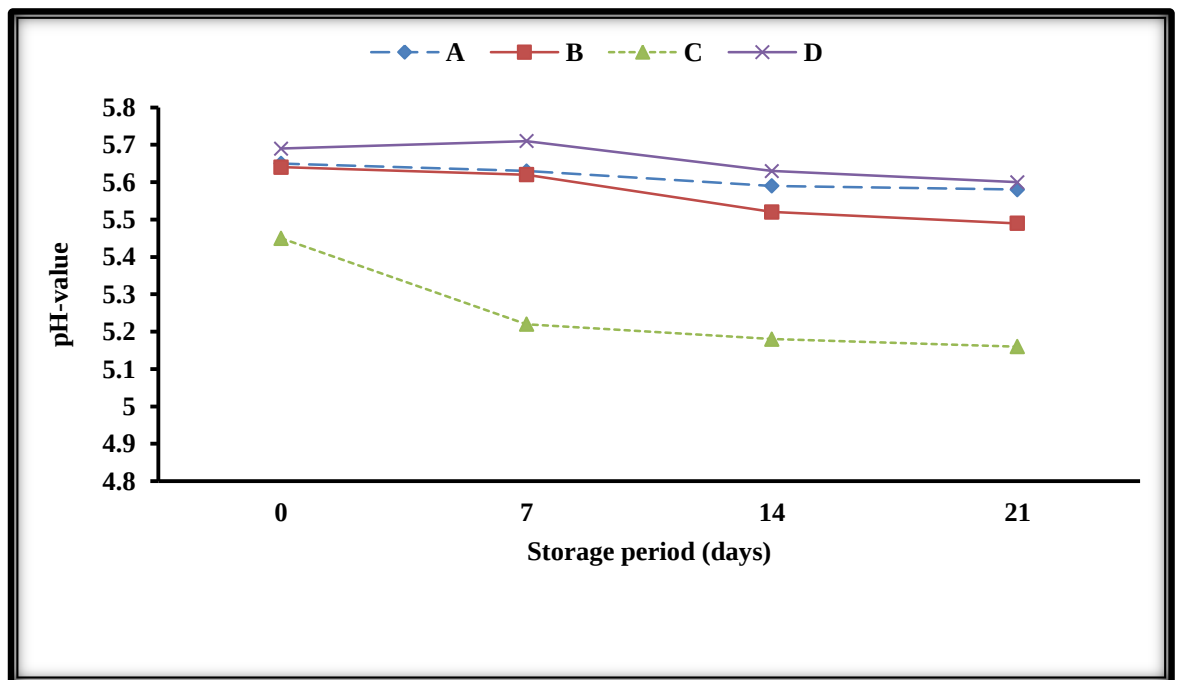


Fig 4: Effect of acetic acid and storage period on chicken breast of pH

Table 3: Sensory evaluation of acetic acid and storage period on chicken breast

Colour						Flavour						Taste					
Sample	Storage period (days)				Overall	Sample	Storage period (days)				Overall	Sample	Storage period (days)				Overall
	0	7	14	21			0	7	14	21			0	7	14	21	
	Scores						Scores						Scores				
A	4.57 ^f ±0.03	4.43 ^g ±0.02	4.34 ^{gh} ±0.01	4.26 ^{hi} ±0.01	4.400 ^B	A	4.84 ^e ±0.02	4.64 ^f ±0.01	4.64 ^f ±0.01	4.58 ^f ±0.01	4.678 ^C	A	4.57f ±0.03	4.43g ±0.02	4.34gh ±0.01	4.26hi ±0.01	4.400B
B	5.88 ^{ab} ±0.01	5.74 ^{cd} ±0.01	5.71 ^{cd} ±0.02	5.63 ^{de} ±0.01	5.742 ^A	B	5.75 ^b ±0.03	5.62 ^{cd} ±0.03	5.62 ^{cd} ±0.03	5.54 ^d ±0.00	5.632 ^B	B	5.88ab ±0.01	5.74cd ±0.01	5.71cd ±0.02	5.63de ±0.01	5.742A
C	5.95 ^a ±0.02	5.82 ^{bc} ±0.02	5.77 ^{bc} ±0.02	5.60 ^e ±0.00	5.785 ^A	C	5.95 ^a ±0.03	5.76 ^b ±0.02	5.76 ^b ±0.00	5.66 ^c ±0.02	5.782 ^A	C	5.95a ±0.02	5.82bc ±0.02	5.77bc ±0.02	5.60e ±0.00	5.785A
D	4.25 ^{hi} ±0.01	4.21 ⁱ ±0.02	4.21 ⁱ ±0.03	4.17 ⁱ ±0.02	4.210 ^C	D	4.16 ^g ±0.01	4.19 ^g ±0.02	4.15 ^g ±0.03	4.11 ^g ±0.01	4.153 ^D	D	4.25hi ±0.01	4.21i ±0.02	4.21i ±0.03	4.17i ±0.02	4.210C
Overall	5.162 ^A	5.052 ^B	5.008 ^B	Overall		Overall	5.177 ^A	5.051 ^B	5.043 ^B	4.973 ^C		Overall	5.162 ^A	5.052 ^B	5.008 ^B	4.915C	
Lsd _{0.05}	0.1052 [*]					Lsd _{0.05}	0.07438 ^{**}					Lsd _{0.05}	0.1052 [*]				
SE±	0.03651					SE±	0.02582					SE±	0.03651				

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

Table. 4: Sensory evaluation of acetic acid and storage period on chicken breast

Juiciness					Texture						Overall acceptability						
Sample	Storage period (days)				Overall	Sample	Storage period (days)				Overall	Sample	Storage period (days)				Overall
	0	7	14	21			0	7	14	21			0	7	14	21	
	Scores						Scores						Scores				
A	5.57 ^c ±0.08	5.41 ^c ±0.04	5.40 ^c ±0.01	5.38 ^c ±0.02	5.441 ^B	A	5.57 ^c ±0.04	5.50 ^c ±0.00	5.48 ^c ±0.04	5.42 ^c ±0.01	5.493 ^B	A	4.37 ^c ±0.03	4.28 ^c ±0.02	4.25 ^c ±0.02	4.22 ^c ±0.00	4.280 ^C
B	4.59 ^d ±0.06	4.39 ^e ±0.05	4.34 ^e ±0.02	4.31 ^e ±0.03	4.407 ^C	B	4.35 ^d ±0.02	4.25 ^d ±0.02	4.22 ^d ±0.01	4.21 ^d ±0.01	4.254 ^C	B	4.67 ^b ±0.02	4.47 ^{bc} ±0.01	4.47 ^{bc} ±0.01	4.42 ^{bc} ±0.01	4.508 ^B
C	3.68 ^f ±0.04	3.62 ^{fg} ±0.02	3.54 ^{fg} ±0.05	3.44 ^g ±0.02	3.569 ^D	C	3.21 ^e ±0.03	2.83 ^f ±0.01	3.12 ^e ±0.02	3.12 ^e ±0.02	3.071 ^D	C	5.46 ^a ±0.01	5.43 ^a ±0.02	5.47 ^a ±0.02	5.43 ^a ±0.01	5.447 ^A
D	6.77 ^a ±0.07	6.52 ^b ±0.04	6.51 ^b ±0.04	6.38 ^b ±0.03	6.546 ^A	D	6.68 ^a ±0.01	6.59 ^{ab} ±0.02	6.53 ^{ab} ±0.04	6.42 ^b ±0.01	6.557 ^A	D	4.30 ^c ±0.02	4.25 ^c ±0.03	4.20 ^c ±0.05	4.19 ^c ±0.06	4.236 ^C
Overall	5.153 ^A	4.985 ^B	4.947 ^{BC}	4.878 ^C		Overall	4.951 ^A	4.794 ^B	4.838 ^B	4.792 ^B		Overall	4.700 ^A	4.608 ^A	4.597 ^A	4.566 ^A	
Lsd _{0.05}	0.1968 [*]					Lsd _{0.05}	4.951 ^A					Lsd _{0.05}	0.2577 [*]				
SE±	0.06831					SE±	4.951 ^A					SE±	0.08944				

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

Table. 5:Effect of acetic acid and storage period on chicken breast
Total viable count of bacteria (TVC)

Sample	Storage period (days)				Overall
	0	7	14	21	
A	3.567 ^g ±0.04	3.570 ^e ±0.02	3.576 ^d ±0.02	3.558 ⁱ ±0.01	3.568 ^B
B	3.554 ^j ±0.02	3.565 ^h ±0.01	3.567 ^g ±0.00	3.540 ^l ±0.03	3.557 ^C
C	3.525 ^o ±0.03	3.530 ^m ±0.01	3.541 ^k ±0.02	3.528 ⁿ ±0.02	3.531 ^D
D	3.590 ^c ±0.01	3.593 ^b ±0.04	3.594 ^a ±0.03	3.569 ^f ±0.01	3.586 ^A
Overall	3.559 ^C	3.564 ^B	3.570 ^A	3.549 ^D	
Lsd _{0.05}	0.0005259**				
SE±	0.0001826				

Values are means ±SD

Mean(s) sharing same superscript(s) in columns and rows are not significantly different (P>0.05) according to DMRT

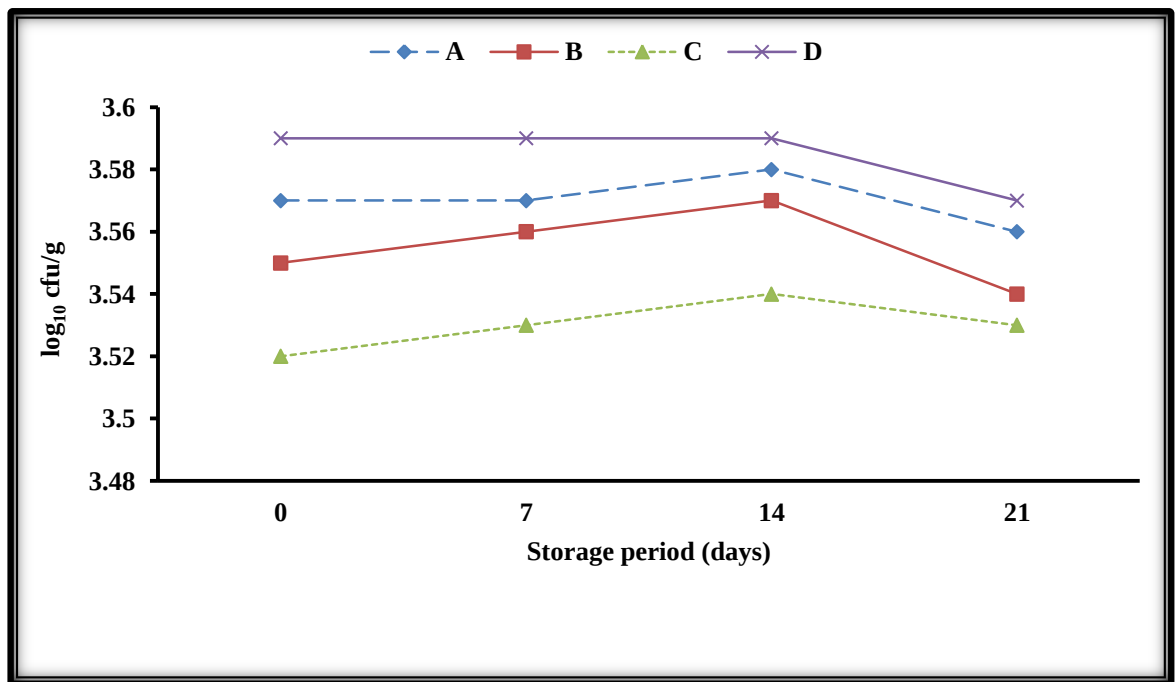


Fig5: Total viable count of bacteria (TVC)

Table 6: The effect of acetic acid and storage on chicken breast

***E.coli* (MPN/g)**

Sample	Storage period (days)			
	0	7	14	21
A	-	-	-	+
B	-	-	-	+
C	-	-	-	+
D	-	-	-	+

Table 7: Effect of acetic acid and storage period on chicken breast *Salmonella* (MPN/g)

Sample	Storage period (days)			
	0	7	14	21
A	-	-	-	-
B	-	-	-	-
C	-	-	-	-
D	-	-	-	-

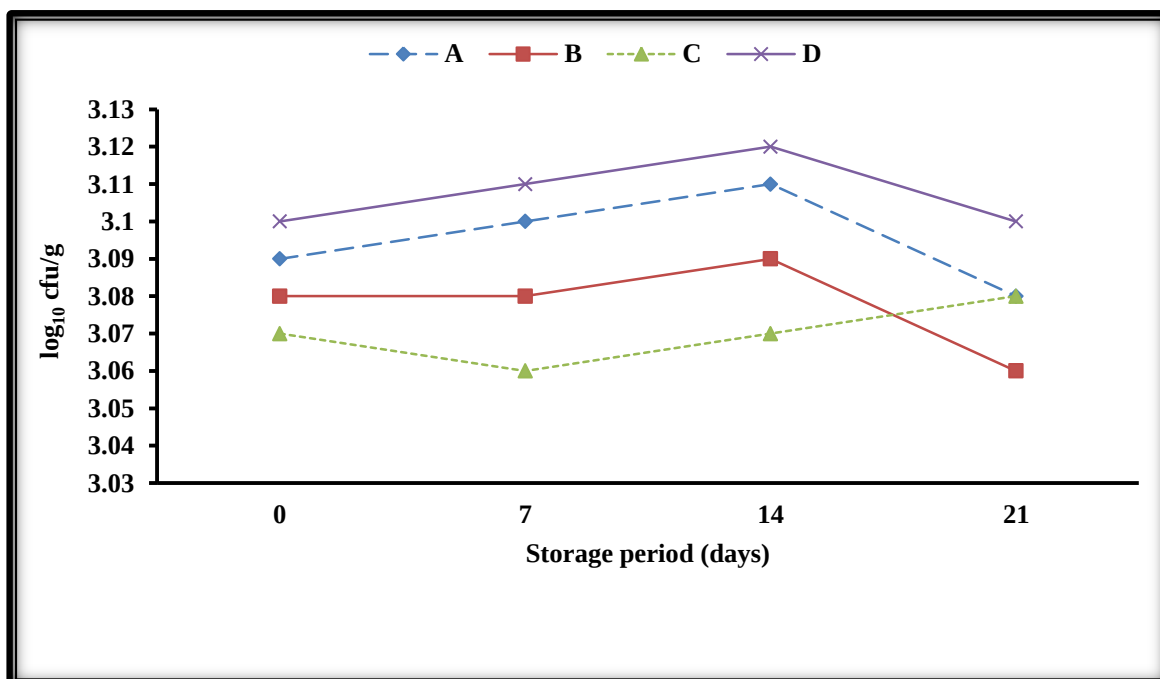
Table 8: Effect of acetic acid and storage period on chicken breast *Staphylococcus aureus* ($\log^{10}$ c fu/g)

Sample	Storage period (days)				Overall
	0	7	14	21	
A	3.089 ^g ±0.01	3.105 ^d ±0.04	3.111 ^c ±0.04	3.080 ^j ±0.01	3.096 ^B
B	3.076 ^k ±0.02	3.085 ^h ±0.01	3.093 ^f ±0.02	3.064 ⁿ ±0.02	3.080 ^C
C	3.073 ^l ±0.01	3.059 ^o ±0.02	3.071 ^m ±0.02	3.082 ⁱ ±0.04	3.071 ^D
D	3.105 ^d ±0.02	3.112 ^b ±0.03	3.119 ^a ±0.00	3.104 ^e ±0.01	3.110 ^A
Overall	3.086 ^C	3.090 ^B	3.099 ^A	3.083 ^D	
Lsd _{0.05}	0.0005259 ^{**}				
SE±	0.0001826				

Values are means ±SD

Mean(s) sharing same superscript(s) in columns and rows are not significantly different ($P>0.05$) according to DMRT

Fig 6: *Staphylococcus aureus*



Key :

A ≡ Sample treated with 1% acetic acid

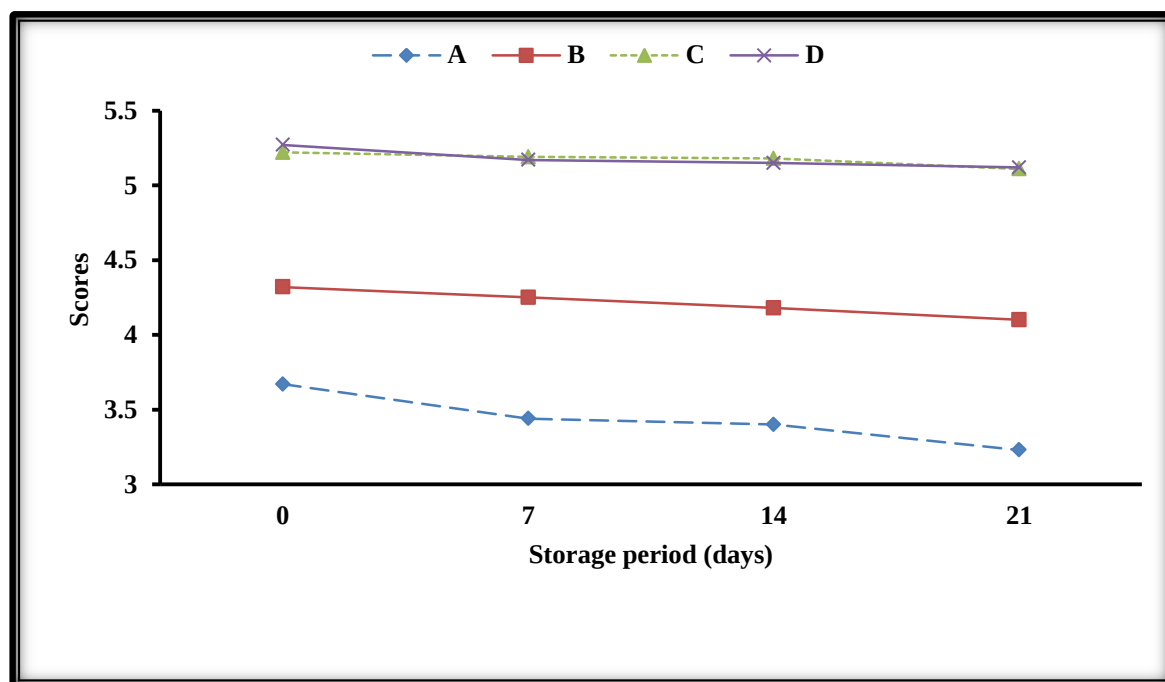
B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

Table 9: Effect of acetic acid and storage period on chicken breast Yeasts and moulds (MPN/g)

Sample	Storage period (days)			
	0	7	14	21
A	-	-	-	-
B	+	-	-	+
C	+	-	-	+
D	+	-	-	+



Sensory. Colour

Fig 7: Effect of acetic acid and storage period on chicken breast

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

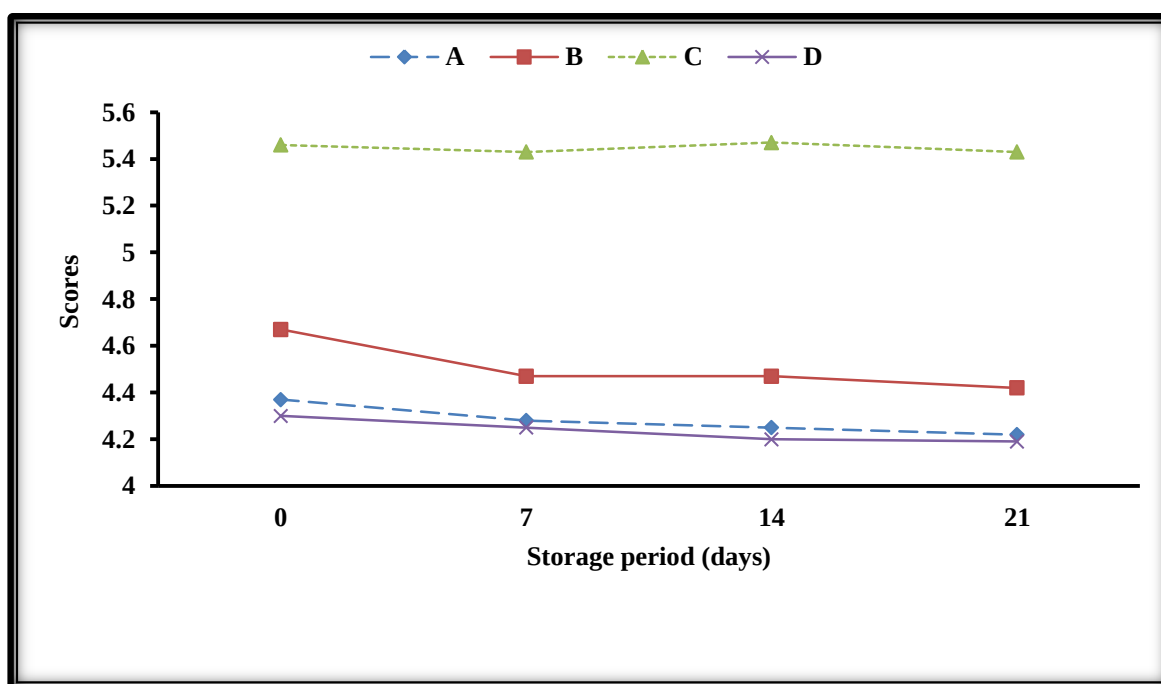


Fig 8:Effect of acetic acid and storage period on chicken breast

Sensory. Flavor

Key:

- A ≡ Sample treated with 1% acetic acid
- B ≡ Sample treated with 2% acetic acid
- C ≡ Sample treated with 3% acetic acid
- D ≡ Control

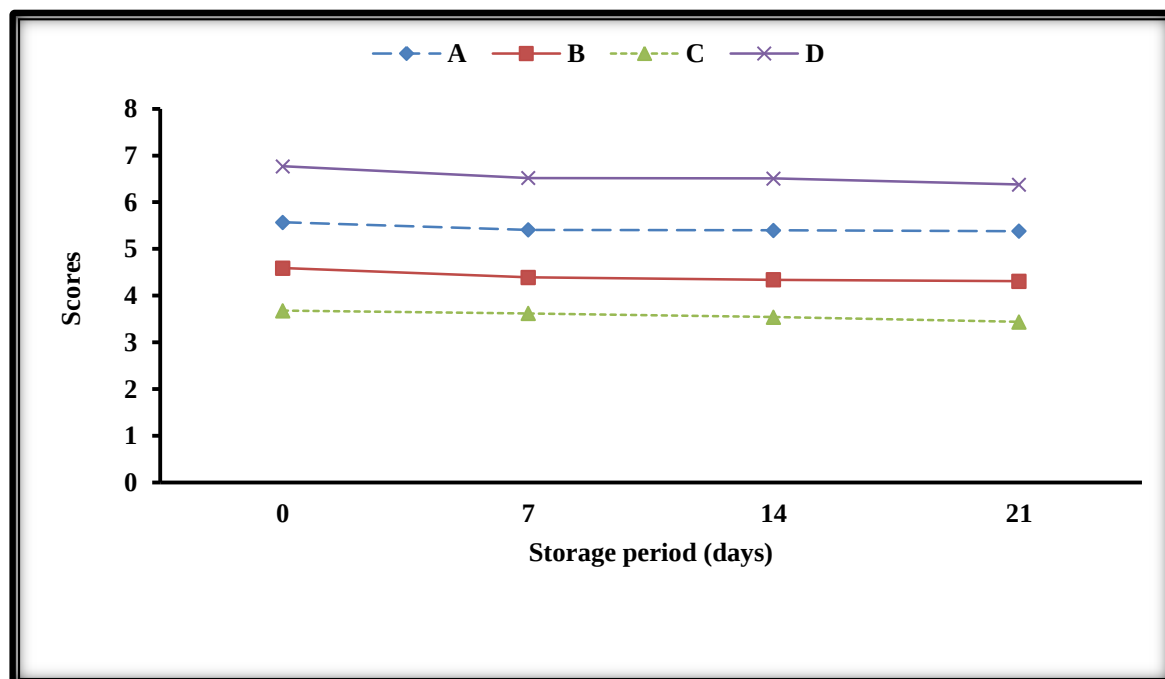


Fig 9:Effect of acetic acid and storage period on chicken breast

Sensory. Juiciness

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

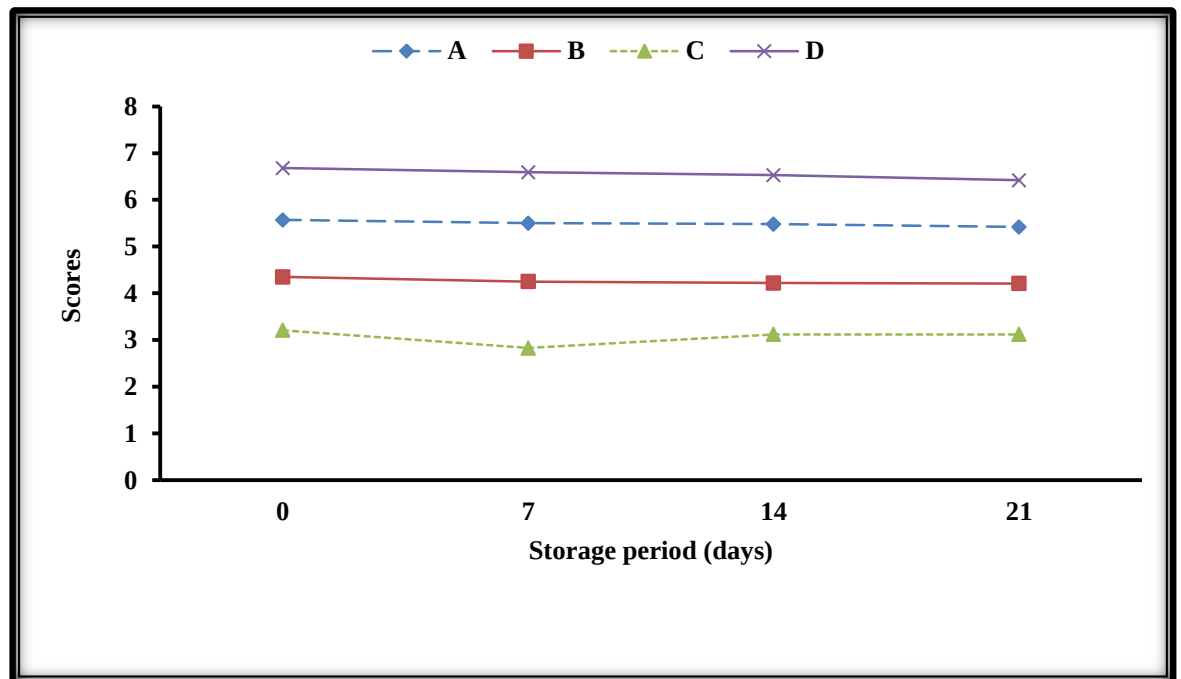


Fig 10:Effect of acetic acid and storage period on chicken breast

Sensory. Texture

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

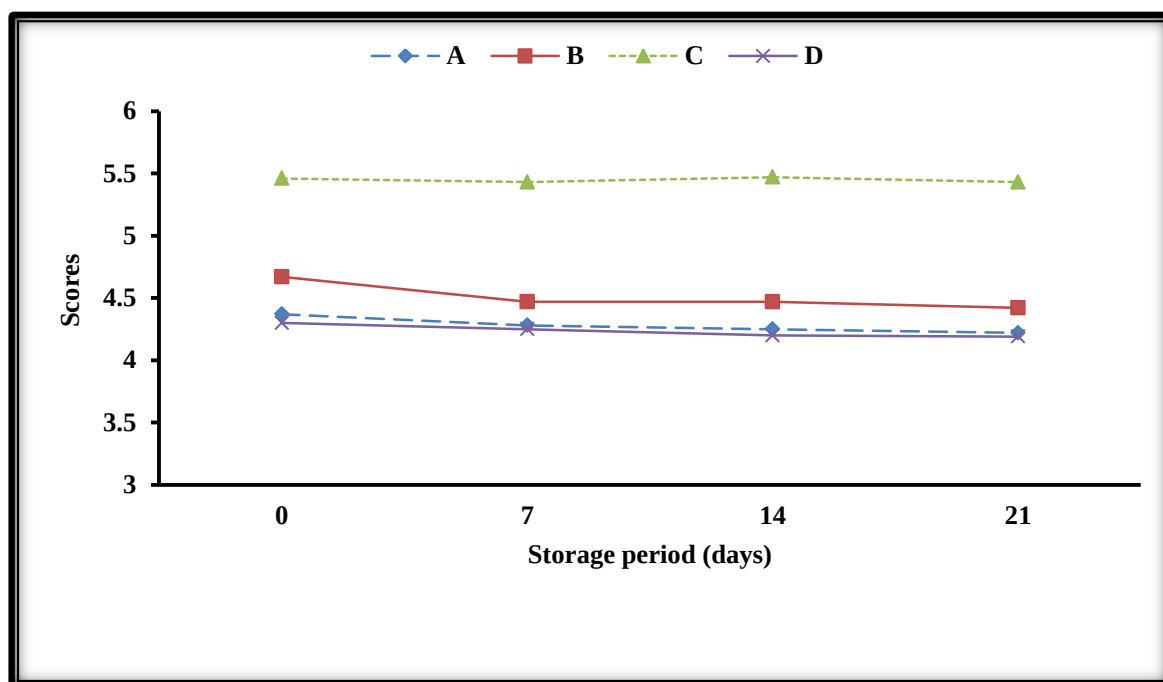


Fig 11:Effect of acetic acid and storage period on chicken breast

Sensory. Overall acceptability

Key:

A ≡ Sample treated with 1% acetic acid

B ≡ Sample treated with 2% acetic acid

C ≡ Sample treated with 3% acetic acid

D ≡ Control

CHAPTER FIVE

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

Through this study concluded that the treatment of chicken breast by acetic acid 3% reduced the total count of bacteria, yeast and moulds while preserved the qualities of sensory and chemical to chicken breast addition to refrigerator in freezer for at least 14 days without changing dynamic meat qualities.

5.2 Recommendations

- Acetic acid can be utilized as functional additive to preserve chicken meat against microbial growth.
- Finding showed that acetic acid exhibit significant antimicrobial and antibacterial activities.
- Finally .we recommends using vinegar concentration (1% to 3%).

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Appendices

Appendix 1

Sensory evaluation chart

The parameters	A	B	C	D
Color				
Aroma				
Taste				
Texture				
Juiciness				
Overall acceptability				

7-Extremely like

6-moderately like

5-Like

4-Slightly like

3-Like

2-Dislike

1-Extremely dislike

If you have any question please ask.

Appendix (2); Effect of acetic acid on microbial analysis

1

The parameters	A	B	C	D
TVC	3.68x10 ³ 3.71x10 ³ 3.69x10 ³	3.60x10 ³ 3.58x10 ³ 3.57x10 ³	3.33x10 ³ 3.36x10 ³ 3.35x10 ³	3.88x10 ³ 3.90x10 ³ 3.89x10 ³
E.coli	-/-/-	-/-/-	-/-/-	+ /+ /+
<i>Staphylococcus</i>	1.23x10 ³ 1.24x10 ³ 1.21x10 ³	1.20x10 ³ 1.19x10 ³ 1.18x10 ³	1.16x10 ³ 1.20x10 ³ 1.19x10 ³	1.28x10 ³ 1.29x10 ³ 1.25x10 ³
Salmonella	-/-/-	-/-/-	-/-/-	-/-/-
Yeast @ mould	-/-/-	-/-/-	-/-/-	-/-/-

The parameters	A	B	C	D
TVC	3.73×10^3	3.68×10^3	3.39×10^3	3.92×10^3
	3.72×10^3	3.67×10^3	3.38×10^3	3.92×10^3
	3.70×10^3	3.66×10^3	3.39×10^3	3.91×10^3
E.coli	-/-/-	-/-/-	-/-/-	+ / + / +
<i>Staphylococcus</i>	1.27×10^3	1.24×10^3	1.17×10^3	1.29×10^3
	1.29×10^3	1.21×10^3	1.14×10^3	1.31×10^3
	1.26×10^3	1.20×10^3	1.13×10^3	1.28×10^3
Salmonella	-/-/-	-/-/-	-/-/-	-/-/-
Yeast @ mould	+ / + / +	-/-/-	-/-/-	+ / + / +

The parameters	A	B	C	D
TVC	3.68x10 ³ 3.71x10 ³ 3.69x10 ³	3.60x10 ³ 3.58x10 ³ 3.57x10 ³	3.33x10 ³ 3.36x10 ³ 3.35x10 ³	3.88x10 ³ 3.90x10 ³ 3.89x10 ³
E.coli	-/-/-	-/-/-	-/-/-	+ /+ /+
<i>Staphylococcus</i>	1.23x10 ³ 1.24x10 ³ 1.21x10 ³	1.20x10 ³ 1.19x10 ³ 1.18x10 ³	1.16x10 ³ 1.20x10 ³ 1.19x10 ³	1.28x10 ³ 1.29x10 ³ 1.25x10 ³
Salmonella	-/-/-	-/-/-	-/-/-	-/-/-
Yeast @ mould	-/-/-	-/-/-	-/-/-	-/-/-

The parameters	A	B	C	D
TVC	3.63×10^3	3.47×10^3	3.38×10^3	3.70×10^3
	3.63×10^3	3.48×10^3	3.39×10^3	3.71×10^3
	3.59×10^3	3.46×10^3	3.36×10^3	3.72×10^3
E.coli	-/-/-	-/-/-	-/-/-	+/+/+
<i>Staphylococcus</i>	1.20	1.15	1.21	1.27
	1.21	1.16	1.20	1.28
	1.20	1.17	1.21	1.26
Salmonella	-/-/-	-/-/-	-/-/-	-/-/-
Yeast @ mould	+/+/+	-/-/-	-/-/-	+/+/+

Appendix(2)

Effect of acetic acid enrichment and storage period on the sensory characteristic of chicken meat.

The parameters	A	B	C	D
Color	3.55	4.23	5.33	5.30
	3.50	4.37	5.21	5.28
	3.97	4.36	5.12	5.24
Taste	4.67	5.89	5.99	4.26
	4.69	5.88	5.98	4.25
	4.34	5.86	5.89	4.24
Flavor	4.81	5.78	5.98	4.20
	4.89	5.72	5.90	4.16
	4.83	5.75	5.98	4.13
Juciness	5.46	4.50	3.98	6.63
	5.62	4.38	3.43	6.73
	5.64	4.89	3.64	6.94
Texture	5.48	4.30	3.13	6.64
	5.60	4.36	2334	6.75
	5.63	4.38	3.16	6.64
Overall acceptability	4.38	4.47	5.88	4.36
	4.38	4.85	5.18	4.28
	4.34	4.69	5.32	4.27

Stage1

The parameters	A	B	C	D
Color	3.44	4.20	5.20	5.18
	3.43	4.28	5.21	5.17
	3.45	4.26	5.16	5.16
Taste	4.37	5.79	5.89	4.22
	4.49	5.68	5.78	4.21
	4.44	5.76	5.79	4.20
Flavor	4.61	5.68	5.78	4.18
	4.69	5.52	5.71	4.16
	4.63	5.65	5.78	4.22
Juciness	5.40	4.30	3.68	6.50
	5.42	4.48	3.53	6.53
	5.40	4.39	3.65	6.54
Texture	5.48	4.20	3.10	6.64
	5.50	4.26	2.30	6.55
	5.53	4.28	3.10	6.59
Overall acceptability	4.28	4.37	5.78	4.30
	4.27	4.55	5.20	4.20
	4.28	4.49	5.32	4.25

Stage**3**

The parameters	A	B	C	D
Color	3.40	4.16	5.19	5.16
	3.41	4.19	5.20	5.15
	3.38	4.20	5.15	5.14
Taste	4.33	5.70	5.83	4.22
	4.34	5.70	5.73	4.21
	4.36	5.74	5.74	4.20
Flavor	4.61	5.68	5.78	4.15
	4.69	5.52	5.71	4.13
	4.63	5.65	5.78	4.18
Juciness	5.40	4.30	3.63	6.50
	5.40	4.38	3.43	6.530
	5.40	4.34	3.55	6.50
Texture	5.43	4.20	3.10	6.56
	5.50	4.23	2.14	6.52
	5.51	4.22	3.13	6.52
Overall acceptability	4.24	4.37	5.68	4.20
	4.26	4.55	5.50	4.20
	4.26	4.49	5.22	4.20

The parameters	A	B	C	D	Stage4
Color	3.20	4.11	5.10	5.13	
	3.21	4.10	5.11	5.12	
	3.28	4.10	5.12	5.12	
Taste	4.23	5.60	5.63	4.21	
	4.24	5.66	5.53	4.20	
	4.30	5.64	5.64	4.10	
Flavor	4.56	5.58	5.68	4.11	
	4.59	5.50	5.66	4.11	
	4.59	5.55	5.64	4.11	
Juciness	5.38	4.30	3.53	6.40	
	5.38	4.30	3.33	6.30	
	5.39	4.32	3.45	6.45	
Texture	5.33	4.20	3.10	6.44	
	5.43	4.22	2.12	6.42	
	5.50	4.20	3.13	6.41	
Overall acceptability	4.21	4.37	5.58	4.18	
	4.22	4.45	5.49	4.19	
	4.24	4.45	5.21	4.20	

Effect of acetic acid enrichment and storage period on the pH of chicken meat.

The parameters	A	B	C	D
Ph	5.66	5.64	5.45	5.69
<i>Stage1</i>	5.63	5.64	5.44	5.69
	5.66	5.64	5.47	5.69
Ph	5.62	5.63	5.21	5.68
<i>Stage2</i>	5.63	5.62	5.23	5.70
	5.63	5.62	5.22	5.74
Ph	5.60	5.53	5.18	5.63
<i>Stage3</i>	5.59	5.52	5.17	5.62
	5.59	5.51	5.19	5.63
Ph	5.58	5.50	5.15	5.60
<i>Stage4</i>	5.58	5.49	5.16	5.61
	5.59	5.49	5.16	5.60

Appendix 4:

Effect of acitic acid on chicken non-nitrogen protein analysis

Treatment	0 time	7 day	14 day	21 day
A	2.54	2.84	2.01	1.31
B	2.12	3.20	2.95	3.40
C	3.53	3.35	3.29	3.55
D	3.15	3.08	3.36	3.19