EFFECTS OF HIGH PRESSURE PROCESSING AND ETHYL LAUROYL ARGINATE ON THE SHELF-LIFE OF READY-TO-EAT SLICED CHICKEN BREAST ROAST

A Thesis

submitted in partial fulfillment of the requirements for the degree of

Master of Food Technology

Massey University
Albany, New Zealand

Sadia Seemeen

August 2011

ABSTRACT

Title: Effects of high pressure processing and ethyl lauroyl arginate on the shelf-life of ready-to-eat sliced chicken breast roast.

High pressure processing (HPP) is becoming increasingly popular in commercial food processing as it offers great potential within the food industry. The popularity of the technology is driven by the need to provide minimally processed foods which are safe, wholesome and have extended shelf-life that challenge traditional methods of food processing. High pressures of upto 900 MPa can be used to kill or inhibit microorganisms without changing the nutritional and sensory properties of the food. However, the inherent high resistances of bacterial endospores and food enzymes are the major challenges for the broader application of HPP. Therefore, a hurdle approach is almost axiomatic for significant widespread use of HPP in commercial food processing. Therefore, several antimicrobial compounds have been used in conjunction with HPP in a hurdle approach to improve the overall quality of the products. Ethyl lauroyl arginate (LAE) has not been investigated in combination with HPP. LAE is a novel antimicrobial compound derivative of lauric acid, L-arginine and ethanol, all of which are naturally occurring substances. LAE can extend the shelf-life of products due to its antimicrobial action on spoilage microorganisms during refrigerated storage. Therefore, the objective of this study was to investigate the effects of HPP and LAE on the shelf-life of ready-to-eat (RTE) cooked chicken breast roast during storage at 4°C for 16 weeks. The RTE cooked chicken breast roast was prepared using portions (samples) of freshly marinated chicken breasts, which were cooked to an internal temperature of 75°C for 5 minutes, and then cooled (4°C), sliced (60 mm) and vacuum-packaged. The study was conducted in two phases, each carried out for 16 weeks. The first phase comprised of fourteen unique treatments which were screened by microbial and instrumental analysis. Based on the results of the first phase, five treatments were selected for further work. Similar tests were carried on these treatments, in addition to sensory evaluation.

The effects of HPP at 450 MPa and 600 MPa pressures at 1 min, 3.5 min and 5 min hold times respectively, on the shelf-life of RTE sliced chicken breast roast were studied for 16 weeks during storage at 4°C. HPP in combination with LAE (200 ppm) was also investigated using similar treatment pressures, hold times and storage conditions. The effects of LAE (200 ppm & 315 ppm) alone on the shelf life of RTE sliced chicken breast roast was studied for 16 weeks when stored at 4°C. RTE sliced chicken breast roast samples without any preservative and/or HPP treatment served as the controls. Aerobic plate counts (APCs), lactic acid bacteria (LAB) and yeasts and moulds (Y&M) were analyzed in five samples from each of the treatments at regular intervals for upto 16 weeks. Instrumental analyses of color and texture were also conducted on the samples to determine any significant changes during storage at 4°C. Five sample treatments were selected after screening and evaluated by consumer sensory analysis using a 9-point hedonic scale. Analyses for APCs, LAB, Y&M, color and texture were also conducted on the selected samples during refrigerated storage. Survival analysis methodology was used to estimate the consumer sensory shelf-life of the selected treatments at 25% and 50% rejection probability.

The results showed the potential of using HPP to extend the microbiological and consumer sensory quality of the products. Samples treated with HPP alone, and HPP in combination with LAE (200 ppm) at 600 MPa inhibited the growth of APCs for 16 weeks when stored at 4°C. However, there was no significant (P>0.05) difference in the microbial shelf-life of samples treated with 200 ppm or 315 ppm LAE. No significant (P>0.05) changes in color and texture were detected in all the treatments. Further, no LAB or Y&M were detected in all the sample treatments for the entire storage period at 4°C. Samples treated with HP at 600 MPa for 1 min and 5 min, HPP+LAE (200 ppm) at 600 MPa for 1 min, LAE at 200 and 315 ppm were evaluated by a consumer sensory panel at different storage times. The results of the consumer sensory analysis showed no changes in color, texture, flavour and freshness of the HP-treated and HPP+LAE (200 ppm)-treated samples. LAE-treated (200 and 315 ppm) samples were not acceptable by a consumer panel at week 12. A maximum sensory shelf-life of >16 weeks at 50% and 13.8 weeks at 25% rejection probability was obtained for samples treated with HPP at 600

MPa for 1 min. Therefore, samples treated at 600 MPa for 1 min had stable sensory properties and were well-accepted by a consumer panel. Also, the samples had good microbiological quality.

ACKNOWLEDGEMENTS

There are several people and organizations I would like to thank for their help when I pursued the Master’s in Food Technology at Massey University. Without their support, my stay in New Zealand would not have been fruitful.

I would like to thank God for gifting me this beautiful life and giving me the ability to reason, judge and be a good human being.

I would like to acknowledge the financial support given by Tegel Foods Ltd to conduct this project and I would like to thank Roy Biggs, Food Safety and Quality Assurance Manager at Tegel Foods Limited, who gave me this wonderful opportunity to work on a commercial project. Also, I would like to specially thank Robyn Marshall for sharing her technical skills, supervision and invaluable contribution. I would like to acknowledge the full team at Tegel Foods Ltd, Christchurch, who contributed towards the physical completion my project.

I would like to express my deepest appreciation and gratitude to my supervisor, Dr. Tony Mutukumira, who continually and convincingly conveyed a spirit of adventure in regard to research. Without his guidance and persistent help this project report would not have been possible. Thank you for believing in me and supporting me throughout my post-graduate studies and life.

I would also like to thank Rachel Liu, Jarod Young and Marjan Asgarlou for their assistance in the laboratory during the course of the project. I would also like to immensely thank Daniel Walsh for his help with the statistics in this project.

Finally, I would like to thank the people who make my life worth living. I would like to express my love and gratitude to my loving family for their endless love and support. I would like to specially thank my best friend Sneha Prakash for her help and support throughout the project. I am very grateful to have Deepak Menon as my partner, who is my source of strength and inspiration.

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ABBREVIATIONS

ANOVA = Analysis of variance
APCs = Aerobic plate counts
CFD = Computational fluid dynamics
FDA = Food and drug administration
FE = Finite Element
GRAS = Generally recognized as safe
HP = High pressure
HPP = High pressure processing
KPa = Kilo pascals
LAB = Lactic acid bacteria
LAE = Lauric arginate
MPa = Mega pascals
RTE = Ready-to-eat
SD = Standard deviation
Y&M = Yeasts and mould

1.0 INTRODUCTION

Cooked poultry meat is a highly perishable product with a shelf-life of 14-15 days when stored at 4°C (Patsias, Chouliara, Badeka, Savvaidis, & Kontominas, 2006). Several factors affect the shelf-life of the cooked chicken product, which include the microorganisms that survive the process of cooking, post-cooking procedures, such as slicing and packaging, and storage temperature (Samelis, Kakouri, & Metaxopoulos, 1998). However, the shelf-life can be extended through the use of appropriate packaging, including modified atmospheres, or through vacuum packaging (Patsias et al., 2006), and more recently advanced technologies such as high pressure processing (Erkan, Üretener, & Alpas, 2011; Polydera, Stoforos, & Taoukis, 2003; Ramirez-Suarez & Morrissey, 2006). Extending the shelf-life of cooked poultry meat to 10-12 weeks has several advantages. Consumers would still regard the product as fresh and minimally processed and the longer shelf-life would allow more flexibility in the manufacturing and distribution supply chain, and potentially, less waste.

High Pressure Processing (HPP) is an emerging preservation technology that is finding wide applications in the food industry. It is a method of food processing wherein the food is subjected to elevated pressures (upto 900 megapascals (MPa)) with or without the application of heat to achieve microbial inactivation or to alter food attributes to achieve consumer-desired qualities (Garriga, Grebol, Aymerich, Monfort, & Hugas, 2004). The technology is commercially adopted to process a wide variety of foods including jams, jellies, fish, oysters, meat, salad dressing, rice cakes, juices, smoothies, and guacamole (Ramaswamy, Balasubramaniam, & Sastry, 2011). HPP is slowly gaining momentum in the meat industry, particularly in the processing of ready-to-eat (RTE) products. HPP has been successfully used as a final preservation step for pre-packed cooked meats (Garriga et al., 2004). The technology is useful as it can inactivate many microorganisms, and thereby give improved product safety and shelf-life, without significantly affecting the sensory or nutritional properties of the food and it is being used commercially in a number of countries in the Europe and USA for this purpose. Hormel Foods, Kraft Foods, Perdue, Foster Farms, Wellshire Farms and Cargill Foods meat processors that have successfully utilized the technology for a wide variety of RTE, minimally processed food products (Balasubramaniam, 2010).

A number of commonly encountered pathogens found in RTE meals are susceptible to inhibition by high pressure (Jofré, Garriga, & Aymerich, 2011). Pathogens such as *Listeria monocytogenes, Salmonella tryphimurium* and *Staphylococcus aureus* are relatively sensitive to pressure and a treatment of 400-600 MPa for a few minutes is sufficient to give a significant reduction in numbers in RTE foods (Aymerich, Jofre, Garriga, & Hugas, 2005; Garriga et al., 2004; Hayman, Baxter, O'Riordan, & Stewart, 2004; Tassou, Nychas, Koutsoumanis, & Skandamis, 2008).

The inactivation of microorganisms by HPP is probably a result of a combination of factors, therefore cell death is due to multiple or accumulated damage inside the cell (Jofre, Aymerich, Grebol, & Garriga, 2009). The various effects of HPP on microbial inactivation can be grouped into cell envelope-related effects, pressure-induced cellular changes, biochemical aspects and effects on genetic mechanisms. However, bacterial spores are extremely pressure-resistant (Torres & Velazquez, 2005), just as they are resistant to many other physical treatments such as heat and irradiation. Typically, spores can survive at pressures >1000 MPa (Cheftel, 1995b) and the use of high pressure alone is unlikely to be successful in reducing numbers to safe levels recommended in food legislation. Such high pressures can adversely affect the acceptance quality of many foods and thus may limit the commercial application of HPP.

A possible approach to overcome this important disadvantage of HPP is the application of hurdle technology, which relies on the synergistic combination of moderate doses of inactivation and or growth-retarding factors (Leistner & Gorris, 1995). Typically, there is an increasing interest in using heat in combination with high pressure (Leadley, Tucker, & Fryer, 2008) and antimicrobials in combination with high pressure to sterilize foods (Jofré et al., 2011). The hurdle concept is being investigated in many applications because it often allows reduced intensity of any antimicrobial treatment while improving the overall antimicrobial protection (Marcos, Aymerich, Monfort, & Garriga, 2008).

HPP has been adopted for the post-packaging pasteurization of sliced meat products, including fermented meats, whole and sliced cured deli ham, dry cured ham, pre-cooked chicken strips, cold cuts, and other delicatessen meat products (Jo et al., 2011). The meat products retain their sensory characteristics and have a significant extension of shelf-life under chilled storage conditions (upto 120 days) (Jofre et al., 2009). Accordingly, the food manufacturers can reduce the levels of added chemical preservatives (like sodium lactate, potassium lactate, sodium metabisulphite etc.) that can create a metallic or salty aftertaste and consequently the meat tastes fresher and better (Doona & Feeherry, 2007). The high levels of inactivation observed with the application of antimicrobial agents and HPP are reported to be due to the combined factors of destabilization of membrane structure or function. The specific modes of action being different for the type of antimicrobial agent used (Houdt, Michiels, & Masschalck, 2001). Antimicrobials that are safe, natural and effective in low to moderate doses such as nisin and lauric arginate (LAE) can be investigated as synergistic combination treatments with HPP.

LAE is a novel antimicrobial compound derivative of lauric acid, L-arginine, and ethanol, all of which are naturally occurring substances. The ingredients of LAE have been self-affirmed as Generally Recognised As Safe (GRAS) (Luchansky et al., 2005). LAE is known to have a broad spectrum of antimicrobial efficacy and is hydrolyzed in the human body by chemical and metabolic pathways, breaking the compound into its natural constituents, which are L-arginine and ethanol. Furthermore, LAE is rapidly metabolized to the amino acid arginine, which then undergoes normal amino acid catabolism, resulting in the production of urea and carbon dioxide.

The antimicrobial properties of LAE are derived from the action of the preservative on the cytoplasmic membranes of microorganisms, which include the reduction of surface tension and the formation of ionic aggregates leading to changes in the conductivity and solubility of cell membranes (Taormina & Dorsa, 2009b). The disruption of proteins in the cellular membrane can lead to leaking of ions and other cellular constituents resulting in permanent alterations in the cell permeability and subsequent inhibition of growth, or inactivation of the microorganism (Taormina & Dorsa, 2009a). LAE is most effective in controlling pathogens such as *L. monocytogenes* in cooked meats (Bakal & Diaz, 2005).

It is known to extend the shelf-life of the products by improving the appearance of the product during refrigerated storage due to its action against lactic acid bacteria which are mainly responsible for deterioration of organoleptic properties of food products (Bakal & Diaz, 2005).

Modern consumers demand fresh, minimally processed, RTE nutritious foods. In order to meet the demands of consumers and expand the market for poultry products, more effective preservative systems are required which involve increased testing of new preservatives and/or combinations of existing and alternative preservatives. The use of a combination of antimicrobial with HPP maybe an efficient preservative system for the poultry industry in terms of improving the microbial quality of the products, whilst retaining natural flavor, texture and color and other sensory attributes of the products.

Preservation of RTE meat products requires the choice of the appropriate combined technologies to achieve the desired levels of microbial inactivation and shelf-life extension (Raso & Barbosa-Canovas, 2003). HPP alone and in combination with LAE was being investigated in this study as a potential hurdle technology to improve the microbiological quality and extend the shelf-life of RTE sliced chicken breast roast. In view of this, the objective of the study was:

1.1 Objective

The main objective of the study was to determine the effect of HPP and LAE on the shelf-life of ready-to-eat cooked chicken breast roast during storage at 4°C for 16 weeks.

1.2 Specific objectives

The specific objectives of the study were:

- To investigate the effects of HPP alone, LAE alone and HPP+LAE combinations on RTE sliced chicken breast roast, based on microbial, color and texture analysis when stored at 4°C for 16 weeks,

- To select treatments that are capable of significantly (*P*<0.05) extending the microbial shelf-life of the product with insignificant (*P*>0.05) changes to color and texture, and
- To determine the consumer acceptance of the selected treatments under refrigerated storage.

2.0 LITERATURE REVIEW

2.1 Introduction

Food processing involves the transformation of raw animal or plant materials into safe consumer-ready products with the objective of stabilizing the products by preventing or reducing any changes in quality. Without these processes, we would neither be able to store food from time of plenty to time of need nor to transport food over long distances (Lund, 2003).

To consumers, the most important attributes of a food product are its sensory characteristics such as texture, flavour, aroma, shape and color. These attributes determine an individual's preference for specific products and minor differences between brands of similar products can have a significant influence on acceptability. An important goal of food manufacturers is to develop and employ processing technologies that retain or create desirable sensory qualities or reduce undesirable changes in food due to processing.

Physical (eg. freezing, heating, dehydration and packaging) and chemical (eg. reduction of pH or use of preservatives) preservation methods continue to be used extensively and technological advances to improve the efficiency and effectiveness of these processes are being made at a rapid rate. The basis of these traditional methods involves reducing microbial growth and metabolism to prevent undesirable chemical changes in food. Probably, the most common method of food preservation used today is thermal treatment (eg. pasteurization, sterilization). Although heating food effectively reduces levels of microorganisms such as bacteria, such processing can alter the natural taste and flavour of food as well as destroy vitamins.

Therefore, alternatives or novel food processing technologies are being explored and implemented to provide safe, fresher-tasting, nutritive foods without the use of heat or chemical preservatives. Innovative non-thermal processes for preservation of food have attracted the attention of many food manufacturers. Consumer demand for minimally-processed food products had presented specific challenges to food processors. These processors all face the same problem; how to keep the food fresh and healthy with high retention of vitamin and nutrient levels, while offering a reasonable shelf-life and convenience and assuring food safety.

In search for new processing methods, particularly for certain products, the application of high-pressure processing (HPP) has shown considerable potential as an alternative technology to heat treatments, in terms of assuring safety and quality attributes in minimally-processed food products (Palou, Lopez-Malo, & Welti-Chanes, 2002).

HPP potentially answers many of these challenges. Unlike heat treatment, HPP does not reduce the quality of foods and pressure is evenly instantaneously and uniformly transmitted throughout the sample, which allows products without over-treated parts to be obtained. HPP can thus facilitate the production of foods that have the quality of fresh foods but the convenience and profitability associated with shelf-life extension (McClements, Patterson, & Linton, 2001).

2.2 Principles of High Pressure Processing

Currently, a great deal of research is being directed towards understanding the effects of high pressure (HP) on food and food ingredients (Balasubramaniam, 2010; Rastogi, Raghavarao, Balasubramaniam, Niranjan, & Knorr, 2007). Studies of the effects of high pressures on foods date back to over a century. In 1899, Bert Hite of the Agriculture Research Station in Morganstown, West Virginia, USA, designed and constructed a high-pressure unit to pasteurize milk and other food products (Hogan, Kelly, & Sun, 2005). Hite constructed a machine that could reach pressures in excess of about 6800 atmospheres (approximately 700 MPa) to examine the potential use of HPP for a wide range of foods and beverages, including the pressure inactivation of viruses. The level of sophistication that was accomplished was remarkable, given the technological disadvantages at that time period regarding processing systems and packaging materials. In 1899, Hite and co-workers reported that treatment at pressures of 450 MPa or greater could improve the keeping quality of milk (Hogan, Kelly, & Sun, 2005). In 1914, Hite and co-workers showed that yeasts and lactic acid bacteria associated with sweet, ripe fruit were more susceptible to pressure than other organisms, especially spore-forming bacteria associated with vegetables (Gilmour, Patterson, Quinn, & Simpson, 1995).

Compared to today's HPP equipment, the prototype system utilized in the 1890s by Hite was very primitive. Today, with advances in computational stress analysis and new materials, high capacity pressure systems can be manufactured to allow reliable HP treatment of food products at even higher pressures (Hoover, 1993).

Although the potential for HPP of foods has thus been known since the late nineteenth century, its application and potential have only been recently widely recognized. While this potential was largely ignored through most of the last century, basic work on the effects of hydrostatic pressures on biological systems steadily developed. In recent years, the use of HPP as a food preservation technique has gained momentum throughout the world as an alternative to traditional heat-based methods, for the reasons cited earlier. Much of the research regarding the use of HP for food preservation has focused on the inactivation of microorganism; the pressure stability of food enzymes is now also beginning to attract increasing attention (Ashie & Simpson, 1996a; Krebbers et al., 2003).

2.2.1 Description of the process

In a high pressure process, the food product intended for treatment is placed in a pressure vessel capable of sustaining the required pressure and the product is submerged in a liquid, which acts as the pressure-transmitting medium. Water may be used as the pressure-transmitting medium, but media containing castor oil, silicone oil, sodium benzoate, ethanol or glycol are also used (Balasubramanian & Balasubramaniam, 2003). The ability of the pressure-transmitting fluid to protect the inner vessel surface from corrosion, the specific HP system being used, the process temperature range and the viscosity of the fluid under pressure are some of the factors that must be considered while selecting the medium (Knoerzer, Buckow, Sanguansri, & Versteeg, 2011; Rasanayagam et al., 2003).

Industrial HP treatment is currently a batch or semi-continuous process (Hogan, Kelly, & Sun, 2005). The selection of equipment depends on the kind of food product to be processed. Solid food products or food with large solid particles can only be treated in a batch mode. Liquids, slurries and other pumpable products have the additional option of semi-continuous production (Ting & Marshall, 2002). Currently, most HP machines in industrial use for food processing are batch systems, whereby the product is placed in a high-pressure chamber and the vessel is closed, filled with pressure-transmitting medium and pressurized either by pumping medium into the vessel or by reducing the volume of the pressure chamber, for example by using a piston. If water is used as the pressurizing medium, its compressibility must be accounted for; water is compressed by up to 15 per cent of volume at pressures above 600 MPa (Knoerzer et al., 2011). Once the desired pressure is reached, the pump or piston is stopped, the valves are closed and the pressure is maintained without further energy input (Gao, Ju, & Wu, 2007). After the required hold time has elapsed, the system is depressurized, the vessel opened and the product unloaded. The system is then reloaded with the product, either by operators or machines, depending on the degree of automation possible (Ting & Marshall, 2002). The total time for pressurization, holding time and depressurization is referred to as the ‘cycle time’. The cycle time and the loading factor (i.e. the percentage of the vessel volume actually used for holding packaged product, primarily a factor of package shape) determine the throughput of the system. In a commercial situation, with this sort of batch process, a short holding time under pressure is desirable in order to maximize throughput of product.

For any HP system, the working pressure is a very important parameter, not only because the initial price of the equipment increases significantly with its maximum working pressure, but also because a decrease in working pressure can reduce significantly the number of failures, increasing the working life of the equipment (Ting & Marshall, 2002). Pressures anywhere between 50 and 1000 MPa are commonly used.

Keeping the sample under pressure for extended periods of time does not require any additional energy (Gao et al., 2007). The work of compression during HP treatment will increase the temperature of foods through adiabatic heating, by approximately 3°C per 100 MPa, depending on the composition of the food (Knoerzer et al., 2011). For example, if the food contains a significant amount of fat, such as butter or cream, the temperature rise can be larger. Foods cool down to their original temperature on decompression if no heat is lost to, or gained through, the walls of the pressure vessel during the hold time at pressure.

While the temperature of a homogenous food will increase uniformly due to compression, the temperature distribution in the mass of food during the holding period at pressure can change due to heat transfer to or from the walls of the pressure vessel. The pressure vessel must be held at a temperature equal to the final food temperature increase from compression for ideal isothermal conditions. Temperature distribution must be determined in the food and reproduced each treatment cycle if temperature is an integral part of the HPP microbial inactivation process specification.

Foods decrease in volume as a function of the imposed pressure (Rastogi et al., 2007). An equal expansion occurs on decompression. For this reason, the packaging used for HPP treated foods must be able to accommodate up to a 15% reduction in volume and return to its original volume, without loss of seal integrity and barrier properties.

Regarding HPP as a food processing technology, the greater the pressure level and time of application, the greater the potential for changes in the appearance of selected foods (Jo et al., 2011). This is especially true for raw, high protein foods where pressure-induced protein denaturation will be visually evident (Rastogi et al., 2007).

2.2.2 Process principles

The governing principles of HPP are based on the assumption that foods which experience HP in a vessel follow the isostatic rule regardless of the size or shape of the food. The isostatic rule states that “pressure is instantaneously and uniformly transmitted throughout a sample whether the sample is in direct contact with the pressure medium or hermetically sealed in a flexible package” (Hugas, Garriga, & Monfort, 2002). Therefore, in contrast to thermal processing, the time necessary for HPP should be independent of the sample size (Rastogi et al., 2007).

The effect of HP on food chemistry and microbiology is governed by Le Chatelier’s principle. The principle states that when a system at equillibrium is disturbed, the system then responds in a way that tends to minimize the disturbance (Pauling, 1964). In other words, HP stimulates some phenomena (eg. phase transition, chemical reactivity, change in molecular configuration, chemical reaction) that are accompanied by a decrease in volume, but opposes reactions that involve an increase in volume. The effects of pressure on protein stabilization are also governed by this principle, i.e. the negative changes in volume with an increase in pressure causes an equillibrium shift towards bond formation. Alongside this, the breaking ions are also enhanced by HP, as this leads to a volume decrease due to the electrostriction of water. Moreover, as hydrogen bonds are stabilized by high pressure, their formation involves a volume decrease. Pressure does not generally affect covalent bonds. Consequently, HP can disrupt large molecules such as enzymes, proteins, lipids or microbial cell structures like cell membranes, and does not affect small molecules such as vitamins and flavour components (Linton & Patterson, 2000).

Due to the work of compression, HPP causes temperature to rise inside the HP vessel. This is known as adiabatic heating and should be given due consideration during the preservation process. The value of the temperature increments in the food and pressure transmitting medium will be different, as they depend on food composition as well as processing temperature and pressure and the rate of pressurization (Otero, Ramos, deElvira, & Sanz, 2007). In food sterilization, adiabatic heating can be used advantageously to provide heating without the presence of sharp thermal gradients at the process boundaries. Knowledge of the engineering concepts of HPP has broadened extensively in recent times. Therefore, relevant engineering principles that promote the capabilities of HP are discussed in the following topics.

The mechanism of cellular inactivation

The effectiveness of a food preservation technique is primarily evaluated on the basis of its ability to eliminate the pathogenic microorganisms that are present. Cellular inactivation is closely associated with morphological changes that occur within individual microbial cells during HPP (Alpas, Kalchayanand, Bozoglu, & Ray, 2000); studies of which are discussed in Section 2.3. Cell disruption is highly specific to the geometry of the bacteria, as opposed to its Gram-type (Schreck, Layh-Schmidt, & Ludwig, 1999), although this has been disputed (Yuste, Capellas, Pla, Fung, & Mor-Mor, 2001). Moreover, presence of a cell wall does not mean pressure resistance is enhanced; in fact, quite the opposite has been hypothesized by Ludwig et al. (2002) who suggested that pressure may induce mechanical stresses on the microbial cell wall, which, in turn, may interact with inactivation mechanisms.

Although the above studies show strong correlations between the physiological state of the microorganisms and degree of pasteurization, cell disruption during processing remains poorly understood at the fundamental level of fluid and cell interactions (Smith, Zhang, & Thomas, 2000a). Up to quite recently, this has been quantified via a cell-wall-strength model which presumes disruption to occur when the fluid stresses that are imparted on a cell wall exceed some defined threshold. This has been successfully applied to animal cells, as these have no proper cell wall (Thomas & Zhang, 1998). Progress, however, has been slower for microbial cells whose well-structured cell walls add considerable complexity. As a consequence, there is a lack of understanding and characterization of the mechanical properties of microbial cell walls (Smith et al., 2000a).

To appreciate the mechanical strength of microbial cells and the factors that contribute to that strength, investigations of cell mechanical properties under periods of pressurization are necessary. As yeast cells are widely used to produce intracellular bio-products of commercial interest, experimental techniques have been employed to evaluate their properties. For example, via micromanipulation, the relationship between bursting force, diameter and the relationship between force and displacement of yeast cells have been established (Mashmoushy, Zhang, & Thomas, 1998). Fortunately, yeast cell walls are structurally complex. Therefore, experimentation may provide scope for understanding mechanisms of inactivation in complex microorganisms such as *Escherichia coli*.

In recent years, it has been found that when three dimensionless parameters, namely the permeability constant, the initial thickness to radius ratio and the initial radial stretch ratio, were found from experiments, the non-unique properties for cell walls of biological cells could be derived (Smith, Moxham, & Middleberg, 1998).

To determine the cell wall properties for yeast cell using these dimensionless parameters, Smith et al. (2000a) conducted compression experiments. They used osmotic theory to interpret measurements of cell volume as a function of external osmotic pressure. Then, they quantified the effect of osmotic pressure and cell compression rates on the bursting force, deformation at bursting and cell diameter.

To determine the intrinsic cell wall properties and cell wall failure criteria, the force-deformation data obtained were used in conjunction with a finite element (FE) mechanical model (Smith, Moxham, & Middleberg, 2000b). Specifically, this model determined the mean Young's modulus (when used in conjunction with simple membrane theory), mean maximum von Mises stress-at-failure and mean maximum von Mises strain-at-failure. Unfortunately, internal organelles of the yeast cell which are also susceptible to stress were not considered, thereby reducing the model's acceptability in the area of HPP. Delgado and Hartmann (2004) addressed this issue by using the above information in the development of a FE mechanical model of a yeast cell during compression phase of HPP (Figure 1), which was experimentally validated with yeast cell volume reduction data from Perrier et al. (1995). Instead of using a volume loss equation as was done in the study of Smith et al. (1998), a reduced form of the Cauchy equation of motion represented the mechanical behavior of the yeast cell.

Major organelles were modeled to investigate the homogeneity of the stress distribution in the cell as well as the cell deformation characteristics. The authors found that at 400 MPa, the critical effective strain upon failure of the organelles membranes of 80% was predicted, correlating well to the experimental studies of Shimada et al. (1993). Most notably, Delgado and Hartmann (2004) predicted a non-homogenous stress distribution in the cell. In addition, through dimensional analysis, the authors found that the compression rates did not influence cellular inactivation.

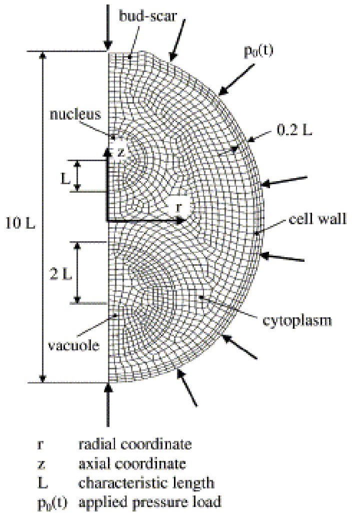

Figure 1: Finite element model of yeast cell under compression (Delgado & Hartmann, 2004).

Thermal-Hydraulic processes in HPP

As HPP often involves heat interactions and fluid flow, thermal-hydraulic investigations, i.e., the study of thermodynamic and fluid-dynamic phenomena, have shown to be of high importance (Hartmann et al., 2004). The thermal-hydraulic processes that occur during the HPP of both fluid and solid food systems can be highly influential on the efficiency and effectiveness of the overall process (Hartmann, 2002; Rademacher, Werner, & Pehl, 2002). The internal energy of the HP system changes during compression/decompression phases, resulting in heat transfer between the internal system and its boundaries. The first experimental observations of fluid temperature in an HP vessel were made by Pehl et al. (2000) who revealed a heterogeneous temperature distribution through high-pressure thermochromatic liquid crystals.

An important contribution to the understanding of thermal-hydraulics in the HPP of a fluid-food system at mild temperatures (i.e. 40°C) was made by Delgado and Hartmann (2002). Computational fluid dynamics (CFD) and dimensional analysis were used to determine the timescales of convection, conduction and bacterial inactivation, the relative values of which contribute to the efficiency and uniformity of conditions during HPP. During the study, conductive and convective timescales were directly compared to the inactivation timescale to provide a picture of the thermal-hydraulic states of HP vessel during bacterial inactivation. Results of high industrial relevance were provided, for example, it was shown for pilot scale systems, that when processed fluids exhibit a larger convective timescale than the inactivation timescale, intensive fluid motion and convective heat transfer resulted in the homogeneous bacterial inactivation.

As regards the HP vessel boundaries, Otero et al. (2000) and Hartmann et al. (2004), showed that the thermal properties of the HP vessel boundaries have considerable influence on the uniformity of the process, and insulated material promoted the most effective conditions. As well as this, the insulated vessel was found to increase the efficiency of HPP by 40% (Hartmann et al., 2004). Kowalczyk and Delgado (2007) suggested that HP systems with a characteristic dimension of 1 m alongside a low viscous medium should be used to avoid heterogeneous processing of the product.

Other studies provided similar solutions to the thermal-hydraulic phenomena in HPP systems containing packaged ultra-heat treatment (UHT) milk (Delgado & Hartmann, 2003), packaged enzyme mixture (Ghani & Farid, 2006) and solid food analogue material (Otero et al., 2007). For example, tylose with similar properties to meat and agar with similar properties to water, were both used. In both of the investigations of Hartmann et al. (2002 & 2003) the most significant results, revealed by validated CFD simulations, showed strong coupling between concentrations of the surviving microorganisms and the spatial distribution of low temperature zones within the food package in the HP vessel. A low thermal conductive package material was also found to improve the uniformity of the processing by preserving the elevated temperature level within the package throughout the pressurization phase; an average difference of about two log reductions was found per tenfold increase in the package thermal conductivity. The two-dimensional CFD simulations of Otero et al. (2007) found that the filling ratio of the HP vessel played a major role in process uniformity, with convective currents having least effect on heat transfer when this ratio is large (Figure 2). They also showed that by anticipating the temperature increase, which results from compression heating, and by allowing the pressure transmitting medium to supply the appropriate quantity of heat, the uniformity of HPP was enhanced when both large and small sample ratios were used (Figure 3).

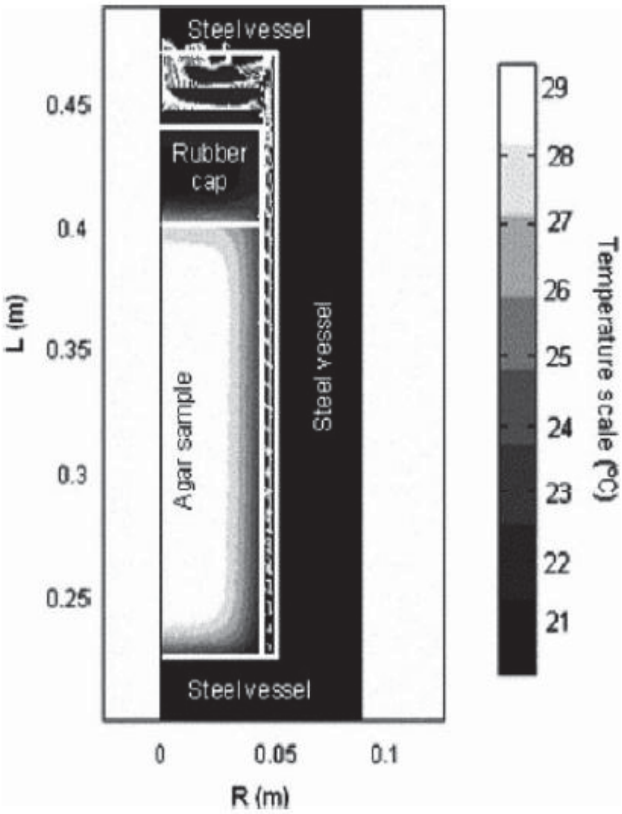

Figure 2: Temperature distributions in an HP chamber with a large filling ratio (Otero et al., 2006).

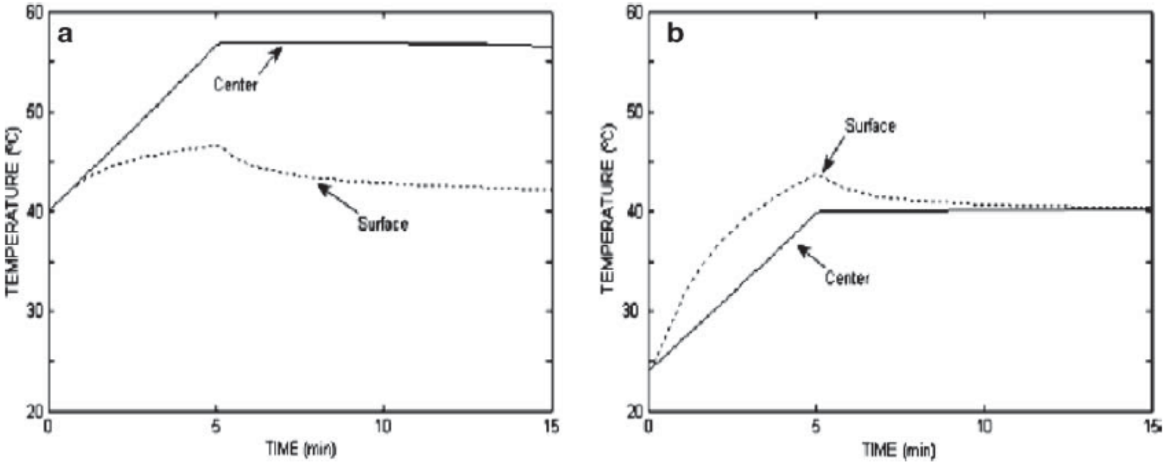

Figure 3: Temperature evolution in tylose sample calculated from the model: the initial temperature of both the tylose sample and the pressure medium was (a) 40°C and (b) 24°C, i.e., showing the benefits of anticipating the adiabatic temperature rise (Otero et al., 2006).

An important feature of the above modeling studies was that contrasting results were possible owing to (i) the HP systems having different operational properties or (ii) numerical modeling limitations. It is evident from the above studies that both temperature and velocity field are transient during the phase of pressure holding, as the fluid velocity distribution influences strongly the temperature distribution and vice versa (Otero et al., 2007). Therefore, to accurately study the relative contributions of forced and natural convection to the effectiveness of HPP, it would be most beneficial to measure velocity as well as temperature and use both to develop a comprehensive validation in future simulation studies.

Thermophysical properties

Designing safe, effective and efficient HPP systems demands the modeling of conceptual designs throughout a range of pressures and temperatures experienced in the food industry. One of the main difficulties when developing or optimizing these systems is the lack of knowledge about the important thermophysical properties of food while under pressure (Hartmann, Delgado, & Szymczyk, 2003). However, such knowledge is important as, from an engineering point of view, theoretically based heat and mass transfer models that allow the accurate prediction of the physical history of food undergoing HP are desirable (Hogan, Kelly, & Sun, 2005). With respect to the thermo-hydraulic studies reviewed in the previous sections, it would not have been possible to evaluate the relative importance of process parameters such as the compression rate (Hartmann, 2002), the size of the HP vessel (Hartmann et al., 2003), the viscosity of the pressure transmitting medium (Delgado & Hartmann, 2002), and the process uniformity (Otero et al., 2007) unless the physical properties of the systems fluids were modeled as functions of pressure and temperature.

In addition, when HPP involves a change of phase, the ice fraction, the enthalpy and the initial freezing point also need to be modeled (Otero, Ousegui, Guignon, LeBail, & Sanz, 2006). Models of these properties during HPP can be derived from (i) additive models considering the food properties under pressure (Otero et al., 2006); (ii) the phase change domain data at atmospheric pressure can be shifted according to the freezing point depression, or an experimentally observed change, associated with the applied pressure (Denys, VanLoey, Hendrickx, & Tobback, 1997; Hartmann et al., 2003) and (iii) the physical property of water under pressure can be multiplied by a constant which represents the ratio of the physical property of the food to that of water at atmospheric pressure (Ghani & Farid, 2006; Hartmann et al., 2003). Another method used by Chen et al. (2007) and Kowalczyk et al. (2005) was to firstly run two-dimensional CFD simulations for a food product undergoing the high temperature with low pressure (HPLT) process and then fit the resulting curves to experimental data by varying the appropriate thermophysical property. A similar technique was followed by Schluter et al. (2004) who allowed coefficients in Weibull distribution of the physical properties to vary in accordance with the prevailing experimental conditions.

2.2.3 General equipment for HPP treatment

Equipment for batch HPP treatment of foods consists of (i) a pressure vessel of cylindrical design, (ii) two end closures, (iii) means for restraining the end closures (for example, yoke, threads, pin), (iv) a low pressure pump, (v) an intensifier which uses liquid from the low pressure pump to generate high pressure process fluid for system compression, and (vi) necessary system controls and instrumentation. The six components of a high pressure processing system can be arranged to treat unpackaged liquid foods in a semi-continuous manner and packaged foods in a batch configuration.

Batch HPP equipment technology

Batch HPP systems are similar in operation to batch thermal processing retort systems in that both process cycles consist of filling the process vessel with product, closing the vessel, bringing the vessel to pressure process conditions, decompressing the vessel and removing the product.(Avure Technologies, 2011). High pressure vessels may operate in a vertical, horizontal, or tilting mode (Farkas & Hoover, 2000). Pressure vessels capable of routine operation at pressures over 400 MPa can be built of two or more concentric cylinders of high tensile strength steel. The outer cylinders compress the inner cylinders such that the wall of the pressure chamber is always under some residual compression at the design operating pressure. Safety codes (ASME Section 8, Division 3 of the Boiler and Pressure Vessel Code) require the inner cylinders to crack to allow leakage to relieve pressure and thus avoid catastrophic failure of the pressure vessel (“leak before break”). The outer cylinder of a pressure vessel may be wire wound or encapsulated in a liquid-filled, permanently pressurized, outer cylinder to ensure a cycle life of over 100,000 cycles at pressures of 680 MPa or higher.

The inner cylinder and all parts exposed to water or food should be made of stainless steel to avoid corrosion. Systems using high tensile strength steel (non-stainless) may use a food-approved oil or water containing FDA- and ISDA-approved lubricants, anti-corrosion agents, and antimicrobial compounds as pressurizing fluids. Packaged foods treated in systems using a lubricant can be protected during HPP treatment by over-wrapping in a sealed bag (Balasubramaniam, 2010). Preferred practice is to design high pressure food processors with stainless steel food contacting parts so that filtered, potable, water can be used as the isostatic compression fluid (Farkas & Hoover, 2000).

Pressure vessels are available as laboratory units with volumes ranging from 0.1 to 2 liters. Pilot plant vessels have capacities of 10 to 25 liters while batch production pressure vessels can be supplied with volumes of several hundred liters (Avure Technologies, 2011).

For batch operation, packaged food is loaded into the pressure vessel, the vessel is sealed, and process water is pumped in the vessel to displace any air. When the vessel is filled, the pressure relief valve is closed, and water is pumped into the vessel until the process pressure is reached (Avure Technologies, 2011). The rate of compression is directly proportional to the horsepower of the low pressure pump driving the intensifier (Hogan, Kelly, Sun, & Da-Wen, 2005). When the process time is completed, the pressure relief valve is opened and the water used for compression is allowed to expand and return to atmospheric pressure. The vessel is opened and the packaged food is removed and is ready for shipment. The displacement of air prior to HPP treatment is done to reduce pumping costs by eliminating air compression. Residual air in the treatment chamber has no effect on microbial inactivation kinetics of HPP-treated, packaged foods. The amount of air in the system is not a critical process factor (Farkas & Hoover, 2000).

A 100-horsepower pump can bring a 50-liter vessel to an operating pressure of 680 MPa in 3-4 min (Hogan, Kelly, Sun et al., 2005). Compression time is a function of pump horsepower. Work must be supplied to compress water at pressures above 200 MPa. Vessel expansion may add several percent to the vessel volume. A filled 100-liter vessel will require an additional 15 liters of water to bring it to a pressure of 680 MPa.

The high cost of pressure vessels, pumps, intensifiers, and sealing systems require that the system cycle as many times per hour as is possible given the hold time at the pressure needed to treat the food. Systems that can perform product loading, vessel sealing, compression, decompression, unsealing and unloading in under 2 min are under design currently (Balasubramaniam, 2010). Target pressure hold times of 5 min or less are desirable. HPP treatments will probably be limited to hold times that are no longer than 10 min. This is in contrast to batch thermal processes which require 60 min to complete a process.

Semi continuous HPP equipment technology

Current semi-continuous systems for treating liquids use a pressure vessel containing a free piston to compress liquid foods. A low-pressure food pump is used to fill the pressure vessel. As the vessel is filled, the free piston is displaced. When filled, the inlet port is closed and high pressure process water is introduced behind the free piston to compress the liquid food. A process pressure of 680 MPa will result in 15% compression of the liquid treated (Knoerzer et al., 2011). After an appropriate process hold time, the system is decompressed by releasing the pressure on the high pressure process water. The treated liquid is discharged from the pressure vessel to a sterile hold tank through a sterile discharge port. A low pressure water pump is used to move the free piston towards the discharge port. The treated liquid food can be filled aseptically into pre-sterilized containers (Avure Technologies, 2011).

Continuous HPP process equipment

A continuous system must compress the liquid food, provide a plug flow hold tube or hold vessel to achieve a specified process time. There must be a means to decompress the liquid such that the liquid is caused to do work to avoid excessive shear and heating. The decompressed, treated liquid could be sent to a sterile hold tank for eventual aseptic filling.

Homogenizers operating above 100 MPa have been proposed as a means for the inactivation of microbes in liquid foods (Moorman, Toledo, & Schmidt, 1996). Experimental data must demonstrate the efficacy of this equipment as a function of operating pressure. Heating effects during decompression must be separated from the contribution made by pressure.

Pulsed HPP processing systems

Semi-continuous and batch equipment can be adapted to pulsed operation by programming a series of treatment cycles of short duration prior to discharging the treated food. Preliminary studies (Aleman et al., 1996) observed an increase in the inactivation rate of yeast with multiple-pulsed pressure treatments. The total pulsed exposure time was equal in duration to a single constant pressure treatment. Pulse frequency, and the ratio of time intervals at pressure and off-pressure, must be considered. Pulse shape (ramp, square, sinusoidal, or other wave form) must also be considered (Hayakawa, Kanno, Tomita, & Fujio, 1994).

2.3 Effects of pressure on microorganisms

The effect of HPP on the degree and mechanism of microbial inactivation has been extensively studied and reviewed (Betti, Omana, & Plastow, 2011; Diez, Santos, Jaime, & Rovira, 2009; Jo et al., 2011; Polydera, Stoforos, & Taoukis, 2005; Ramirez-Suarez & Morrissey, 2006). Most vegetative cells can be inactivated at relatively low pressures, typically 200 to 400 MPa, while bacterial spores are more resistant and need a combination of higher pressures and temperatures (Torres & Velazquez, 2005). High pressure can be used for both pasteurization and sterilization of food products. High pressure pasteurization involves the application of high pressure (between 400-600 MPa) at temperatures up to 60°C (Grauwet, Van der Plancken, Vervoort, Hendrickx, & Van Loey, 2011). Like conventional thermal treatment, high pressure pasteurization treatment inactivates harmful vegetative bacteria in foods effectively (Grauwet et al., 2011) with minimal nutrient degradation. The products pasteurized by HP require refrigeration during storage and distribution. HP sterilization is essentially a pressure accelerated thermal process that requires a combination of elevated pressures (upto 800 MPa) and temperature (up to 120°C) to produce a shelf-stable food free from harmful bacterial spores (Vercammen, Vivijs, Lurquin, & Michiels, 2011). At present, a number of food processors are actively investigating the flexibility of introducing HP sterilized products in commercial markets (Rastogi et al., 2007).

Microbial inactivation by HP has been extensively studied and has been concluded to be the result of a combination of factors. The primary site for pressure induced microbial inactivation is the cell membrane (eg. modifications in the permeability and ion exchange) (Farkas & Hoover, 2000). Microorganisms are resistant to selective chemical inhibitors due to their ability to exclude such agents from the cell, mainly by the action of the cell membrane; however, if the membrane becomes damaged, this tolerance is lost. In addition, HP causes changes in cell morphology and biochemical reactions, protein denaturation and inhibition of genetic mechanisms. Other mechanisms of action which may be responsible for microbial inactivation include the denaturation of key enzymes and the disruption of ribosomes (Katsaros, Katapodis, & Taoukis, 2009). Different organisms react to HPP with different degree of resistance.

2.3.1 Bacteria

Bacteria are relatively simple, single-celled organisms and are among the smallest free-living organisms known. The main bacteria that cause food poisoning are *Campylobacter* sp, *Salmonella* sp, *Listeria monocytogenes, Staphylococcus aureus, Escherichia coli* and *Vibrio* sp. Among these, *Listeria monocytogenes* and *Staphylococcus aureus* are probably the two most intensively studied species in terms of use of HPP. *Listeria monocytogenes* is a Gram-positive rod that is an important pathogen in acidified and other foods such as dairy products and RTE meats. As a food-borne bacteria, *L. monocytogenes* requires particular care for processing and storage because it is moderately heat-resistant and can grow anaerobically under refrigeration. *Staphylococcus aureus* appears to have high resistance to pressure (Wang, Pan, Xie, Yang, & Lin, 2011). *Escherichia coli* O157:H7 also has a high barotolerance and is considered to be an important pathogen that can cause serious illness (Erkmen & Dogan, 2004).

Microbiological safety can be assured in many RTE products if they are processed using HP. Treating food samples using HP can destroy both pathogenic and spoilage microorganisms. However, there is a large variation in the pressure resistance of different bacterial strains and the nature of the medium can even affect the response of microorganisms to pressure. Table 1 compares the different pressures required to inactivate two different types of bacteria at various temperatures in milk and poultry meat.

Table 1: Predicted treatment pressures required at various temperatures for a 5-log inactivation of *Escherichia coli* and *Staphylococcus aureus* in poultry meat and milk, for a treatment time of 15 mins

Temperature (°C) during pressure treatment	Estimated pressure (MPa)			
	Escherichia coli		*Staphylococcus aureus*	
	Poultry meat	UHT milk	Poultry meat	UHT milk
10	850	1014	647	749
20	779	966	694	625
30	681	840	735	602
40	544	638	701	583
50	371	392	524	478
60	125	133	177	196

Source: (Kilpatrick & Patterson, 1998)

The stage of growth of the bacteria is also important in determining the pressure resistance, with cells in the stationary phase being more resistant than those in the exponential phase (Robbins, Balasubramaniam, Stewart, & Ting, 2004). Gram-positive and Gram-negative bacteria differ significantly in terms of chemical structure of their cell walls. The cell walls of Gram-negative bacteria are significantly weaker and Gram-negative bacteria consequently tend to be more pressure-sensitive than Gram-positive bacteria (Erkmen & Dogan, 2004).

Further work is required to understand the factors that can affect the response of microorganisms, including pathogens, to pressure so that treatments can be optimized and microbiological safety can be assured.

2.3.2 Bacterial spores

The elimination of bacterial endospores from food probably represents the greatest food processing and food-safety challenge to the industry. It is well established that spores are the most pressure-resistant life forms known (Baranyi et al., 2006; Enomoto, Nakamura, Hakoda, & Amaya, 1997). In general, only very high pressures (>800 MPa) can kill bacterial spores around ambient temperatures. Alternatively, other processing methods, applied in combination with HP can be effective for elimination of bacterial spores, by achieving a synergistic or hurdle effect. In particular, HPP at elevated temperatures (at up to 90°C) is very effective in the elimination of bacterial spores in foods. Pressure-induced inactivation of bacterial spores is also markedly enhanced at temperatures of 50-70°C and perhaps also at or below 0°C (Crawford, Murano, Olson, & Shenoy, 1996).

The most heat-resistant pathogenic bacterium is *Clostridium botulinum* and its spores are also among the most pressure-resistant spores known (Gao & Ju, 2008). Among other spore-forming bacteria of concern, *Bacillus cereus* has been widely studied because of its anaerobic nature and very low rate of lethality (Gao, Ju, & Jiang, 2006). *Bacillus cereus* is a spore-forming food-borne pathogen, which is ubiquitous in nature and hence occurs frequently in a wide range of food raw materials. It is recognized as a leading cause of bacterial food poisoning, with a variety of proteinaceous and starchy foods being implicated (Guinebretiere, Girardin, Dargaignaratz, Carlin, & Nguyen-The, 2003; Likimani & Sofos, 1990)

An alternative to using treatments combining heat and pressure for enhanced killing of bacterial spores is first to activate bacterial spore germination and then use high pressure to kill the pressure-sensitive vegetative cells. Germination is a process by which a dormant microbial spore changes into a vegetative cell. Interestingly, bacterial spores can be stimulated to germinate by treatment at relatively low pressures, (30-50 MPa). The germinated spores can then be killed by relatively mild heat treatments or high pressure treatments (Gao et al., 2006). Process temperatures in the range of 80-110°C in conjunction with a pressure of about 600 MPa have been used to inactivate spore-forming bacteria such as *Bacillus cereus* (Gao et al., 2006). Cycling treatments, where spores are exposed to alternating low and high pressures, or alternating cycles of pressurization and depressurization are also of interest for sterilization process. However, the mode of action of HPP on bacterial spores is still largely a matter of speculation.

2.3.3 Fungi

Fungi can be divided into two groups based on their vegetative structures; unicellular fungi (yeasts) and those producing hyphae (moulds, mushrooms, etc.). Vegetative bacterial cells, yeasts and moulds are, in general, more susceptible to pressure than bacterial spores; thus, they can be inactivated using relatively low pressures (Balasubramaniam, 2010).

Yeasts are simple single-celled fungi that reproduce by budding or fission. The group includes members of ascomycetes and imperfect fungi. Yeasts are an important group of spoilage microorganisms, but are generally not food pathogens, although toxic mould growth may be a safety concern in foods. Treatment at pressures less than 400 MPa for few mins is sufficient to inactivate most yeasts (Goh, Hocking, Stewart, Buckle, & Fleet, 2007). Goh et al. (2007) reported that at about 100 MPa the nuclear membrane of yeasts was affected and that at more than 400-600 MPa, further alteration occurred in the mitochondria and the cytoplasm.

Moulds are mycelial fungi and many of these organisms are important industrially, such as in food spoilage, food fermentations, and biodegradation processes. Pressures between 300-600 MPa can inactivate most moulds (Tokusoglu, Alpas, & Bozoglu, 2011). HP has been reported to be effective in the inactivation of *Penicillium roqueforti* spores in cheese systems (O'Reilly, Murphy, Kelly, Guinee, & Beresford, 2000). High pressures between 500 and 600 MPa, held for 1-5 min are known to reduce counts of yeasts and moulds in meat products.

2.3.4 Viruses

Among viruses there is a high degree of structural diversity and this is reflected in a wide range of pressure resistances. The most common human enteric virus are Norwalk-like viruses (SRSVs), hepatitis A, rotavirus and human astrovirus. Complete inactivation of suspensions of feline calicivirus (a Norwalk-like virus surrogate), and adenovirus and hepatitis A can be achieved by treatment at 275 MPa for 5 min (Kingsley, Hoover, Papfragkou, & Richards, 2002) , 450 MPa for 15 min (Wilkinson, Kurdziel, Langton, Needs, & Cook, 2001) and at 450 MPa for 5 mins (Kingsley et al., 2002) respectively. The mode of inactivation of viruses by HP has not been fully elucidated although the viral envelope when present appears to be a target for HP inactivation. Treatment at pressures above 300 MPa damages the envelopes of human immunodeficiency virus (HIV) and cytomegalovirus (CMV), preventing the binding of virus particles to cells (Nakagami, Shigehisa, & Ohmori, 1992). Pressure can also cause the dissociation of virus particles; depending on the virus and the treatment conditions, pressure-induced dissociation maybe fully reversible or irreversible (DaPoian, Johnson, & Silva, 1994).. HP can also induce minor changes in viral structures by disassembling the whole particle (Gaspar, Silva, & Gomes, 2002). The formation of non-infectious particle after HP treatment has been observed for many viruses including rotavirus (Pontes, Cordeiro, & Giongo, 2001), HIV (Nagakami, Ohno, & Shigehisa, 1996), lambda phage (Bradley, Hess, & Tao, 2000) and picornaviruses (Oliveira, Ishimaru, & Goncalves, 1999).

2.4 Factors influencing microbial sensitivity to high pressure

As discussed in previous sections, the pressure resistance of microorganisms varies considerably, depending on factors such as species, strain, stage of growth and food composition. Factors that can affect the response of microorganisms, including pathogens, must be considered so that treatments can be optimized and microbiological safety assured.

2.4.1 Intrinsic Factors

pH

The likelihood that a particular organism will grow in a particular food depends on the interaction between all the factors that influence the growth of microorganisms including competition between species. The pH of the food is one of the main factors affecting the growth and survival of microorganisms; all microorganisms have a pH in which they can grow and an optimum pH at which they grow best. The pH of a food, if not optimal for a particular species can thus not only enhance inactivation during treatment but also inhibit outgrowth of sub-lethally injured cells. Bacterial spores are generally more resistant to direct effects of pressure at neutral pH (Gao & Ju, 2008).

Extent of pressure induced inactivation will generally be enhanced and recovery of sub-lethally injured cells inhibited, for most species at acidic pH values. For example, the pressure resistance of *E. coli* O157:H7 in orange juice is dependent on the pH of the juice, the degree of inactivation increasing as pH decreases; survival of *E. coli* O157:H7 in orange juice during storage is also dependent on pH (Bull et al., 2004). Compression of foods during HPP may shift the pH of the food as a function of applied pressure; the direction of pH shift and its magnitude must be determined for each food process.

Water activity (a_w)

Water in a liquid state is essential for the existence of all living organisms. The amount of water available for microbial growth is generally expressed in terms of water activity (a_w) of that system. Lowering the water activity of food can significantly influence the growth of food spoilage or food poisoning organisms that may be present in the raw materials or introduced during processing; this is the principle of the very old method of food preservation by drying. Reducing the a_w appears to protect the microbes against inactivation by HPP; however, on the other hand recovery of sub-lethally injured cells can be inhibited by low a_w (Kouassi, Floros, Anantheswaran, Hayman, & Knabel, 2008). Consequently, the net effect of water activity on microbial inactivation by HPP may be difficult to predict.

2.4.3 Extrinsic factors

Temperature, pressure and holding time

Increasing treatment pressure, holding time or temperature will generally increase the number of microorganisms inactivated (with bacterial spores being the important exception). While many HP processes are performed at ambient temperatures, increasing or, to a lesser extent, decreasing temperature has been found to increase the inactivation rate of microorganisms during HPP (McArdle, Marcos, Kerry, & Mullen, 2011). Temperatures above 45-50°C increase the rate of inactivation of food pathogens and spoilage microorganisms (Roberts & Hoover, 1996a). The use of high temperatures for food processing, the advantages of which for inactivation of spores have been discussed earlier, is complicated by the fact that large steel cylinders in which the food is held during treatment are very slow to change in temperature and that the food itself can undergo a significant increase in temperature during processing due to adiabatic heating. Temperature increases due to adiabatic compression can be 3°C or more per 100 MPa and, while these increases in temperature are generally transient, in some processes use of sample insulation may retain this heat and add to the thermal dimension of the processing conditions (McArdle et al., 2011). The choice of processing temperature will also influence the selection of pressure-transmitting media. There is a minimum critical pressure below which microbial inactivation by HP will not take place regardless of process time.

Important processing parameters to be considered in HPP are the come-up times (period necessary to reach treatment pressure) and pressure-release times. Obviously, long come-up times will add appreciably to the total process time and affect the product throughput, but these periods will also affect inactivation kinetics of microorganisms. Therefore, consistency and control of these times are important in HPP development.

2.5 Effects of high pressure on food quality

While food safety and shelf-life are often closely related to microbial quality, other phenomena such as biochemical reactions, enzymatic reactions and structural changes can significantly influence consumers' perception of food quality (Nielsen et al., 2009). Conventional thermal sterilization processes involve extensive slow heat penetration to the core (cold point) of the product and subsequent slow cooling. The thermal process induces changes in product quality to an extent dependent on the product being treated and the temperatures reached; these may include off-flavour generation, textural softening and destruction of colors and vitamins. As stated already, unlike heating, HPP at moderate pressures generally does not change the odor, flavour, or other sensory characteristics of foods.

Therefore, HPP offers the food industry a technology that can achieve the food safety properties of heat-treated foods while meeting consumer demand for fresher-tasting foods. In order to select the most suitable processing conditions for a particular food product, sensory characteristics must be taken into account. Increasing treatment pressures will generally increase microbial inactivation in shorter times, but higher pressures may also cause greater levels of protein denaturation and other potentially detrimental changes in food quality that could affect the appearance and texture of the food, compared to the unprocessed product (Nielsen et al., 2009).

2.5.1 Effect of high pressure on food color

The importance of color in consumer acceptance is well-known. For some food products, depending on the pressure-time exposure, some degree of protein denaturation can take place during HPP. This can result in a change in physical functionality and/or changes in color relative to raw products. For fresh meat, poultry and related products, pressure-induced color modifications greatly depend on treatment conditions (pressure, temperature and time) and are due to changes in myoglobin, such as denaturation, heme displacement or release and ferrous atom oxidation (Omana, Plastow, & Betti, 2011). Therefore, the application of HP to fresh meat products can result in a cooked-like appearance and the products may sometimes develop a rubbery consisteny (Garriga et al., 2004). For semi-cooked or cooked products, this effect is not observed, since their proteins have already been denatured (Sorenson et al., 2011).

Due to the effects of HPP on the color of raw red meat, the final products have a cooked appearance and cannot be sold as fresh meat. Packaging of meat under vacuum with an oxygen scavenger partly protects the meat color (Balasubramaniam, 2010). However, in practice, HPP of fresh red meat cannot be envisaged unless subsequent (or simultaneous) cooking is done before the final product is presented for sale and consumption. This would be the case if pre-cooked ready meals were to be HP-treated. In contrast, HPP of cured meats or white meats (or fish) are unlikely to cause any serious color problems (Ramirez-Suarez & Morrissey, 2006).

2.5.2 Effect of high pressure on food texture

Knowledge of the rheological and/or textural properties of food products is essential for product development, quality control, sensory evaluation and design, and evaluation of process equipment. The physical structure of most high-moisture products remains unchanged after exposure to HP, since no shear forces are generated by pressure (Farkas & Hoover, 2000). For gas-containing products treated under HP, the color and texture may be changed due to gas displacement and liquid infilteration (Balasubramaniam, 2010). Physical shrinkage can occur due to mechanical collapse of air pockets and shape distortion may be related to anisotropic behavior. For foods not containing air voids, HP frequently results in minimal or no permanent change in textural characteristics (Hogan, Kelly, & Sun, 2005).

For certain food products, HP has an enormous potential as a technique to modify the texture. If required, HPP can induce desirable changes in product texture and structure, and, accordingly, can be used for the development of new products or to increase the functionality of some ingredients (Serrano, Velazquez, Lopetcharat, Ramírez, & Torres, 2004). For example, it was recently reported that HPP of Mozarella cheese significantly accelerated the development of desirable functional properties on melting (O'Reilly, Murphy, Kelly, Guinee, & Beresford, 2002).

HPP is increasingly being used in the production of surimi and kamaboko, traditional Japanese products made from fish mince, due to the superior quality of pressure-induced gels (Ashie & Simpson, 1996a, 1996b; Ohshima, Ushio, & Koizumi, 1993; Okamoto, Kawamura, & Hayashi, 1990). The differences in structure of heat- and HP-treated gels are attributed to the different mechanisms of gelation induced by the differential effects of pressure and heat on various bonds (Angsupanich, Edde, & Ledward, 1999; Angsupanich & Ledward, 1998; Gudmundsson & Hafesteinsson, 2002). HP-induced fish gels are described as glossy and soft, with a smoother and more uniform texture than gels produced by heat treatment; they also retain the color and flavor of the untreated fish. In addition, HP can induce the gelation of sarcoplasmic proteins, usually lost during the traditional production process for surimi, thereby providing the opportunity for the use of HPP in the preparation of surimi and related products (Ohshima et al., 1993).

2.5.3 Effect of high pressure on food sensory quality

One of the most frequently cited benefits of HPP relative to other preservation methods is the possibility of increasing shelf-life while still retaining the sensory characteristics of fresh food products. A study conducted by Hugas et al. (2002) reported that sensory panelists could not distinguish between meat products treated with and/or HPP and untreated controls. Table 2 shows results from a sensory analysis of heat-treated and pressure-treated sausages. Generally, HP-treated sausages were considered more cohesive and less firm than heat-treated sausages. In some cases, the sensory panel did not detect any differences between both types of sausages. However, when differences were detected, HP-treated samples were preferred with respect to appearance, taste and texture in particular.

Table 2: Comparative sensory study of heat-treated (80-85°C for 40 min) and high pressure-treated (500 MPa at 65°C) sausages using the triangle test

Preservation type	Correct judgements	Subject preferences
Heat-treated versus pressurized for 5 min	16	Pressurized= 8 No preference= 5 Heat-treated= 3
Heat-treated versus pressurized for 15 min	22	Pressurized= 11 No preference= 5 Heat-treated= 6

Source: (Mor-Mor & Yuste, 2003)

It has also been reported that HPP can preserve delicate sensory attributes of avocado and assure a reasonably safe and stable shelf-life (Palou et al., 2002). Avocados are used in the preparation of guacamole, which was one of the first HP-treated food products to be commercialized in the USA (Ramaswamy et al., 2011). Furthermore, high pressure has also been used to preserve fruit juices. Table 3 shows that the HP-treated orange juice at 500 MPa for 5 minutes apparently had an extended shelf-life when compared to products subjected to thermal pasteurization at 80°C for 30 seconds (Polydera, Galanou, Stoforos, & Taoukis, 2003).

Table 3: Shelf-life comparison of HP-treated and thermally processed orange juice based on sensory evaluation

Storage temperature (°C)	Shelf-life (days)	
	High-pressure treated (500 MPa/ 5min/ 35°C)	Thermally processed (80°C for 30s)
0	>90	60
5	>90	47
10	47	25
15	32	16

Source: Polydera et al.(2003)

HPP of meat and fish can result in increased oxidation, probably due to the release of free metal ions into the tissue (Ashie & Simpson, 1996a). Oxidation, if not controlled, can negatively affect the color and flavor of such products (Angsupanich & Ledward, 1998). Although HPP does not substantially alter the taste of the fish fillet tissue, pressures above 300 MPa can give the product an opacity similar to that obtained by a very light cooking (Hoover, Metrick, Papineau, Farkas, & Knorr, 1989).

2.5.4 Effect of high pressure on enzyme activity in meats

Loss of color in meat is undesirable as it affects the salability of the products. The mechanisms leading to loss of meat color are not clearly understood. Some researchers attribute it to certain enzyme systems in the meat (Grauwet et al., 2011), while others attribute this change to denaturation of globin proteins at higher pressures (Hayakawa et al., 1994). Color changes typically occur above 150 MPa, resulting in products that resemble cooked meats (Hugas et al., 2002). In fresh beef, formation of metmyoglobin is responsible for loss of red color and a decrease in perceived quality. Fresh beef processed for up to 2 days with pressures between 80 and 120 MPa for 20 min showed a reduction in formation of this pigment. However, according to Cheah and Ledward (1996), the effect of pressure processing was quickly lost if the time between slaughter and processing increased to more than 7 days.

Color changes are most significant in raw products. In sausages containing various levels of mechanically recovered poultry meat and minced pork meat, and cooked with high pressure (500 MPa for 30 min) at 50, 60, 70 or 75°C, a significant loss of redness and an increase in lightness were observed (Yuste, Mor-Mur, Capellas, Guamis, & Pla, 1999) . In products that have already been cooked, the application of high pressure showed no additional effect.

Secondary pasteurization is routinely performed for meat products where casings have been removed to destroy any microbes that have been transferred during post-process handling. High pressure treatment of cooked sausages at 500 MPa for 5 or 15 min at 65°C did not significantly affect the color and therefore high pressure has been suggested as a substitute for pasteurization for this product (Mor-Mor & Yuste, 2003). Absence of color changes after HPP have also been observed in cooked ham (Yuste et al., 2001).

2.5.5 Effect of high pressure on food yield and cost of processing

HPP may affect the yield of certain products, a very important economic issue for food manufacturers (Mor-Mor & Yuste, 2003). While effects on yield depend on the type of product and the intensity of treatment, HPP can give higher yield in food products than heat treatment. Mor-Mor and Yuste (2003) reported that weight loss was significantly higher in heat-treated sausages than in HP-treated control samples. Apparently, HPP also contributed to the prevention of any sour taste and off-flavour development, ropiness and color changes (Hugas et al., 2002).

HPP also has an important additional advantage for oyster processors. A large proportion of oysters are sold ‘on the half-shell’ or ready shelled, traditionally requiring the shucking of oysters by hand. This process is hazardous and inexpert handling can damage the oyster meat, reducing the quality and appearance of the finished product. During HPP, the adductor muscle of oysters detaches from the shell. It has been reported that HPP at

241 MPa for 2 min caused detachment of adductor muscle in 88 percent of oysters, while treatment at 310 MPa with immediate pressure release, resulted in 100 percent efficiency of shucking (He, Adams, Farkas, & Morrissey, 2002). The fitting of oysters with heat-shrinkable plastic bands before treatment holds the shells together and reduces the loss of intervalval fluid. Oysters treated in this way do not gape and have shown to be an attractive alternative to traditional live oysters. In addition to reduced labor cost and risks and increased safety and shelf-life of oysters, yield increases of 25-50% using HPP are not uncommon (Hogan, Kelly, & Sun, 2005).

Both heat treatment and HPP inactivate microorganisms, denature proteins and extend shelf-life of food products. However, heat treatment often achieves this at the expense of quality attributes, such as color, flavour or nutrient levels (Barbanti & Pasquini, 2005). In contrast, HPP maintains the quality of fresh foods, with few direct effects on the flavour and little effect on nutritional quality (Mor-Mor & Yuste, 2003). HPP can also potentially modify the functional properties of food constituents (eg. proteins) and even increase the yield of certain food products (Choi, Hong, Ko, & Min, 2008).

On a commercial scale, high-pressure vessels cost between $500,000 to $2.5 million US dollars depending upon the capacity and extend of automation required by companies (T. Richter, personal communication, 26 August 2011). As a new processing technology with a growing market, pressure-processed products may cost 3 to 10 cents per pound (about 2.2 kg) more to produce than thermally processed products (Balasubramaniam, 2010). With two 215-litre HPP units operating under typical food processing conditions, a throughput of approximately 20 million pounds (about 44 million kg) per year is achievable (Avure Technologies, 2011a). High throughput can be accomplished by using multiple pressure vessels. Factory production rates beyond 40 million pounds (about 88 million kg) per year are now in operation (T. Richter, personal communication, 26 August 2011). As demand for HPP equipment grows, capital cost and operating cost is expected to decrease. Consumers will benefit from the increased shelf-life, quality, and availability of value-added products and new types of foods that cannot be produced with thermal processing methods.

2.6 High pressure regulations

Today, in most countries, food safety is tightly controlled by company, national and international regulations. While many of the factors and microorganisms that can present hazards to the consumer are known and have been intensely studied, new and emerging pathogens not previously regarded as problematic continue to be identified (Hogan, Kelly, & Sun, 2005). As previously discussed, processors must also continue to balance the need for assurance of food safety against consumer demand for minimally-processed products. For these reasons, emerging technologies such as HPP are of considerable interest and potential benefit to the food industry.

However, before the implementation of new preservation technologies, several issues need to be addressed, such as the mechanisms of microbial and enzyme inactivation, the identification of the most resistant and relevant microorganisms in every food habitat, the role of bacterial stress, the effectiveness of the technologies, the increased safety relative to the existing technologies as well as the legislation required to implement them.

Two regulatory attitudes towards commercialization of food products manufactured using HP have emerged, i.e., within the EU or outside. In countries outside the EU, there is currently no specific legislation applicable to HP treatment. In the USA, for example, the traditional health regulations are applied and products treated by HP, such as guacamole and oysters, have already been introduced to the market without any specific regulation (Hugas et al., 2002).

In EU countries, however, national regulations for new products have been replaced, in the application of the precautionary principle, using a community regulation for novel foods and ingredients (Regulation 258/97/EC), which has been in force since 1997 (Hugas et al., 2002). This ‘Novel Foods’ legislation establishes an evaluation and licensing system that is compulsory for new foods and new processes. HPP food products are novel foods since they fulfill two conditions; their history of human consumption has so far been negligible and, secondly, a new manufacturing process has produced them.

In July 2001, the EU commission responsible for 'novel foods' took several decisions to simplify the regulations (Rastogi et al., 2007). Specifically, if it is possible to show that the new product (eg. HP-treated food) is substantially equivalent to a product existing in the market, then the product can be treated at a national regulation level and will not need to comply with the 'novel food' regulation (Jones, 2010).

All new pressure vessels to be used in the EU have to comply with the 'Pressure Equipment Directive' (PED) regulation, which came into force in 2002 (Jones, 2010). This regulation is an extension of the 'CE' safety standard already employed in the EU and now recognized world-wide. As pressure vessels of all types utilize potentially hazardous energy, the PED regulation seeks to identify good design, good manufacturing practices and detailed safety assessment for safe operation and maintenance of the vessels and auxiliary parts (Rastogi et al., 2007).

2.7 HPP in combination with other processing technologies

With respect to HPP, a hurdle approach (Leistner & Gorris, 1995) is evident for significant widespread use in commercial food processing. The inherent high resistances of bacterial endospores and food enzymes are the major challenges to the broad application of HPP.

A preservative method employing HPP (at significantly reduced pressures) is the processing of food under pressure and carbon dioxide (Haas et al., 1989).This method is often referred to as high pressure carbon dioxide processing, even though the pressure levels are normally <15 MPa. Hong et al. (1997) evaluated a CO_2.pressure process for the inactivation of *Lactobacillus* species at pH 4.2 in kimchi (fermented Korean vegetables, pH-4.2). The optimal process parameters that decreased microbial cell populations by 5-log cycles were a 200 min treatment of kimchi samples at 30°C under a CO_2 pressure of 6.9 MPa. Ballestra et al. (1996) examined pressures of 1.2, 2.5, and 5 MPa at 25, 35 and 45°C for the inactivation of *E. coli*. The higher treatment temperatures permitted a shorter processing time (~ 20 min) for the elimination of a cell suspension between 10^9 and 10^{10}

cfu/mL in Ringer's solution when pressure was 1.2 MPa. At higher pressures, temperature had no effect on efficacy. Although the pressures were modest by HPP standards, the effectiveness was high due to the antimicrobial effect of carbon dioxide. The suggested lethal mechanism was attributed to a lowered intracellular pH caused by penetration of elevated levels of carbon dioxide into the cell, not by physical rupture of the cell walls or membrane due to the pressure of CO_2. Results were not as conclusive in studies by Wei et al. (1991). These researchers used 13.7 MPa for 2 h at 35 °C to kill inoculated *Salmonella typhimurium* in chicken and egg yolk, and inoculated *L. monocytogenes* in shrimp, orange juice, and egg yolk. Levels of microbial reduction varied considerably depending on the nature of the food and treatment conditions. Bacterial reductions ranged from limited effect to 9-log cycles. Results were poor for whole egg formulations. Enomoto et al. (1997) reduced spores of *Bacillus megaterium* by 10^7cfu/mL when treated for 30 hours and exposed to 5.9 MPa pressure at 60 °C. Above this pressure spore inactivation was lessened.

Based on these studies, an obvious commercial limitation for pressurized carbon dioxide is the lengthy processing times necessary to allow for diffusion of carbon dioxide into microbial cells. Carlez et al. (1992) investigated the effect of supercritical carbon dioxide on the inactivation rate of *Citrobacter freundii* at pressures of 230 MPa at 35 °C. The treatment did not affect the rate of inactivation. The pH of the meat did not drop below 5.7 and the concentration of carbon dioxide in the meat was 6.5 g/kg. *C. freundii* was recommended as a surrogate for *Salmonella* spp.

Combination treatments of HPP and irradiation have been investigated by several laboratories. Paul et al. (1997) targeted *Staphylococci* in lamb meat. A population of approximately 10^4cfu/g was reduced by only 1-log cycle using either treatment with gamma irradiation (1.0 kGy) or HPP (200 MPa for 30 min). When used in combination, no *Staphylococci* were found immediately after completion of the process. After 3 weeks of storage at 0 to 3°C, mannitol-negative *Staphylococci* (presumably coagulase-negative as well) were detected ($<10^3$ cfu/mL). Crawford et al. (1996) were able to eliminate *C. sporogenes* in chicken breast using combinations of HPP and irradiation.

Raso and co-workers have combined heat, pressure and ultrasound. The pressures used in such combinations are significantly lower than the magnitudes traditionally used in HPP (instead of MPa, kPa levels are used). Raso et al. (1998a) found heat and ultrasound to act independently under pressure. To a large extent, it appeared that the individual contributions of heat and ultrasound under pressure depended upon the temperature used. Above 58°C, any added inactivation caused by pressure was not observed. These results suggested that microbial inactivation was not a simple additive reaction of the three treatment types. D-values recorded for *Y. enterocolitica* ATCC 9610 were 1.39 min at 59°C, 1.5 min for the highest ultrasound setting (150 db at 20 kHz), and 0.28 min for a treatment of 300 kPa and 150 db (ultrasound) at 30°C. In this study by Raso et al. (1998a), *Y. enterocolitica* was suspended in citrate-phosphate buffer (pH 7.0) and the treatment chamber volume was 23 mL.

Raso et al. (1998b) found that a 12-min treatment of 500 kPa and 117 db at 20 kHz was able to kill approximately 99% of a spore suspension of *B. subtilis* ATCC 9372 in McIlvaine citrate-phosphate buffer (pH 7.0). The sporicidal effect depended upon the static pressure, amplitude of ultrasonic waves, and the treatment temperature. Above 500 kPa, additional increments of pressure did not increase the amount of spore inactivation. In the range of 70 to 90°C, a combination with 20 kHz, 300 kPa, 117 db for 6 min had a synergistic effect on spore inactivation.

Many different antimicrobial compounds have been used in combination with HPP in a hurdle approach. Examples include HPP and lytic enzymes (Popper & Knorr, 1990), HPP and antimicrobial chitosans (Papineau, Hoover, Knorr, & Farkas, 1991), and HPP and bacteriocins. Use of nisin with pressure has been addressed by several laboratories. Roberts and Hoover (1996a) examined the concurrent use of nisin with pressure treatment on *B. coagulans* 7050. While pressure alone (up to 400 MPa) had no effect in reducing the number of viable spores when treated at neutral pH and ambient temperature, the use of a 400 MPa/ 70°C/ 30 min pressure treatment at pH 4.0 and 0.8 IU/mL nisin resulted in the sterilization of spore crops containing 2.5 x 10^6 cfu/mL.

Kalchayanand et al. (1998) examined the effectiveness of the pediocin AcH in combination with HPP. The goal of this work was to identify those HPP/AcH treatments capable of inactivating within 5 min 10^7 to 10^8 cfu/mL of *S. aureus, L. monocytogenes, S. tryphimurium, E. coli* O157:H7, *Lactobacillus sake, Leuconostoc mesenteroides, Serratia liquefaciens* and *Pseudomonas fluorescens* in 0.1%-peptone water (Kalchayanand et al., 1998). This could not be accomplished using HPP treatments of 345 MPa/ 50°C/ 5 min, unless 3,000 AU/mL of pediocin AcH were included in the peptone water. Of the Gram-negative bacteria, *E. coli* O157:H7 strain 932 was the most pressure resistant, while for the Gram-positive bacteria in the study, *L. sake* FM1 and *L. mesenteroides* Ly were the most barotolerant. In earlier work, Kalchayandand et al. (1994) had evaluated the hurdle combination of electroporation with HPP and bacteriocins against various Gram-negative and Gram-positive bacteria.

The monoterpenes were investigated by Adegoke et al. (1997) in combination with HPP against *S. cerevisiae*. Alone, *S. cerevisiae* IFO 10149 was found to be resistant when exposed to 300 and 600 mg/L of α-terpinene, but sensitive to a concentration of 1,250 mg/L. When 150 mg/L of α-terpinene was combined with exposure to 177 MPa for 1 h at 25°C, a reduction of 6-log cycles was reported. A 3-log cycle reduction was reported with similar pressure parameters but on replacement of the α-terpinene with 200-mg/L (+)-limonene.

Ishiguro et al. (1993) examined the inactivation of *B. coagulans* in tomato juice with the addition of the antimicrobial compounds polylysine, protamine, and an extract of etiolated seedlings of adlay. Polylysine and protamine were ineffective processing aids; in fact, these compounds conferred protection to *B. coagulans* in the tomato juice treated at 400 MPa. The adlay extract did demonstrate enhanced destruction of *B. coagulans*, improving inactivation by approximately 1 log cfu/mL after 100 min. The treatment temperature was not specified; regardless, treatment times of 100 min are not commercially practical.

2.7.1 Ethyl lauroyl arginate

Introduction

Ethyl N$^{\alpha}$-lauroyl-L-arginate hydrochloride (hereinafter ethyl lauroyl arginate or LAE) is a novel antimicrobial compound derivative of lauric acid, L-arginine and ethanol, which are all naturally occurring substances. The molecule was first synthesized by the CSIC (Higher Council of Scientific Research) in Barcelona in 1984. In 2007, the European Food Safety Authority (EFSA) established an acceptable daily intake (ADI) for LAE of 0.5 mg/kg body weight per day (European Food Safety Authority, 2007). On 1 September 2005, the United States Food and Drug Administration issued a notice that LAE is GRAS (Generally Recognized As Safe- GRAS Notice no. GRN 000164) for use as an antimicrobial agent at concentrations of upto 225 mg/kg in specified categories of commodities (FSANZ, 2009b).

The molecule was synthesized by first esterifying L-arginine with ethanol to obtain ethyl arginate HCl, which was reacted with lauroyl chloride to form the active ingredient ethyl-N$^{\alpha}$-lauroyl-L-arginate hydrochloride (Martin, Griffis, Vaughn, Bryan, Friedly, Marcy, Ricke, Crandall, & Lary, 2009). The preservative is a cationic surfactant, and as a food preservative, is effective against a wide array of Gram-positive and Gram-negative bacteria, yeasts and moulds. LAE in combination with cinnamic acid and sodium benzoate was reported to effectively inhibit *Saccharomyces cerevisiae, Zygosaccharomyces bailii, Brettanomyces bruxellensis,* and *Brettanomyces naardenensis* at pH 3 (Dai, Normand, Peleg, & Weiss, 2010).

LAE has exhibited the ability to inhibit *L. monocytogenes* effectively in meat and poultry products. In a study conducted by Luckansky et al. (2005), the effect of LAE against *L. monocytogenes* on 1.36 kg hams that had been spot inoculated to achieve 7 log cfu per ham was evaluated. For hams inoculated with 7 log cfu, the levels of *L. monocytogenes* decreased by 5.1, 5.4, and 5.5 log cfu/ham within 24 hours at 4°C in samples treated with 4,6, and 8 mL of 5% LAE solutions (Luchansky et al., 2005). The same study also reported that LAE controlled the outgrowth of *L. monocytogenes* for 60 days if the initial inoculum was 3 log cfu per ham, and growth was inhibited for 28 days if the initial inoculum was 7 log cfu per ham.

Sprayed solutions containing LAE have also demonstrated bacterial efficacy against *L. monocytogenes* in frankfurters and sausages. Taormina and Dorsa (2009b) reported that spray treatment with 2.5 mL of LAE solutions at 5000 ppm and 8000 ppm reduced counts of *L. monocytogenes* from 7.69 log cfu/package to 6.80 and 6.13 log cfu/package, respectively.

Seemeen and Mutukumira (2010) evaluated both the lethality and inhibition of *Listeria monocytogenes* 15E03 and aerobic plate counts (APCs) by ethyl lauroyl arginte (LAE) when applied as an ingredient of the formulation and as a post-cook spray in samples of chicken breast roast stored at 4°C for 49 days. In samples inoculated with *Listeria monocygenes* 15E03, there was no significant (P>0.05) difference observed in cell counts and APCs in the treatments with LAE (200 ppm) as an ingredient and LAE (700 ppm) as a post-cook spray. The results suggested that the performance of LAE was not affected by the method of application.

Stability of LAE when combined with other food components

The interaction of the active ingredient ethyl-N$^{\alpha}$-lauroyl-L-arginate hydrochloride with other food components such as hydrocolloids, food preservatives, antioxidants, enzymes, colour additives and proteins was evaluated and out of 33 samples, nine samples showed interaction between the active ingredient and the compounds that constituted the sample (European Food Safety Authority, 2007). Ethyl-N$^{\alpha}$-lauroyl-L-arginate hydrochloride was shown to decrease over time due to its hydrolysis to LAS in four of the samples. The remaining five samples showed interaction with other components including meat, soy proteins, ovo-albumin and lacto-albumin, resulting in the degradation of ethyl-N$^{\alpha}$-lauroyl-L-arginate hydrochloride to ethanol, lauric acid and arginine. Interaction between ethyl-N^{α}-lauroyl-L-arginate hydrochloride and nitrite has been reported, however no nitrosamines were detected (European Food Safety Authority, 2007).

Ethyl-N^{α}-lauroyl-L-arginate hydrochloride is also stable through the duration of shelf life of all the processed foods but a decrease was observed in uncooked foods (Codex Alimentarius Commission, 2007). This can be attributed to the inherent enzymatic action on ethyl-N^{α}-lauroyl-L-arginate hydrochloride in foods such as chickpeas and marinated meats. A higher level of ethyl-N^{α}-lauroyl-L-arginate hydrochloride is suggested as a potential solution to achieve the required shelf life (Seemeen & Mutukumira, 2010).

2.7.2 Mode of action of Ethyl Lauroyl Arginate (LAE)

Ethyl Lauroyl Arginate is a cationic surfactant. The antimicrobial properties of the molecule include the reduction of surface tension and the formation of ionic aggregates leading to changes in conductivity and solubility of cell membranes (Rodríguez, Seguer, Rocabayera, & Manresa, 2004). The disruption of proteins in the cellular membrane can lead to the leaking of ions and other cellular constituents resulting in permanent alterations in cell permeability and subsequent inhibition or inactivation of the microorganism. LAE is reported to have a broad spectrum of activity against Gram-positive, Gram- negative bacteria, yeasts and moulds (Bakal & Diaz, 2005).

The level of action of cationic surfactants against specific microorganisms is influenced by the cell structure and physiology. An increased quantity of phospholipids, fatty acids, and neutral lipids in cell membranes inhibits the penetration of cationic surfactants. Another mechanism that has been suggested with increased sensitivity is the increased activity of efflux pumps which act by reducing intracellular surfactant concentrations (Ishikawa, Matsumura, Tsuchido, & Yoshizako, 2002).

2.7.3 Metabolism of Ethyl Lauroyl Arginate (LAE) in humans

LAE interacts with the cell membranes of microorganisms causing denaturation of the proteins resulting in increased cell permeability and growth inhibition or cell death (Rodríguez et al., 2004). As part of safety evaluation studies, the metabolic fate of LAE in rats, which are the main animals in toxicity studies, has been greatly investigated (Hawkins, Rocabayera, Ruckman, Segret, & Shaw, 2009). In a study conducted by Hawkins et al. (2009), it was concluded that LAE was readily hydrolysed to the corresponding lauroyl arginine, LAS, which is the corresponding acid formed by hydrolysis of the ester, on incubation with human plasma samples to the extent of about 50% during 4 hours (Hawkins et al., 2009). LAE is stable in simulated gastric fluid but in simulated intestinal fluid it is rapidly hydrolysed to LAS and arginine, with more than 90% conversion to arginine after 1 hour (Hawkins et al., 2009).

Metabolism of LAE in mammalian cells

Based on research studies on biotransformation of LAE in humans and rats, a pathway (Figure 4) was proposed for mammalian metabolism of LAE (Hawkins et al., 2009).

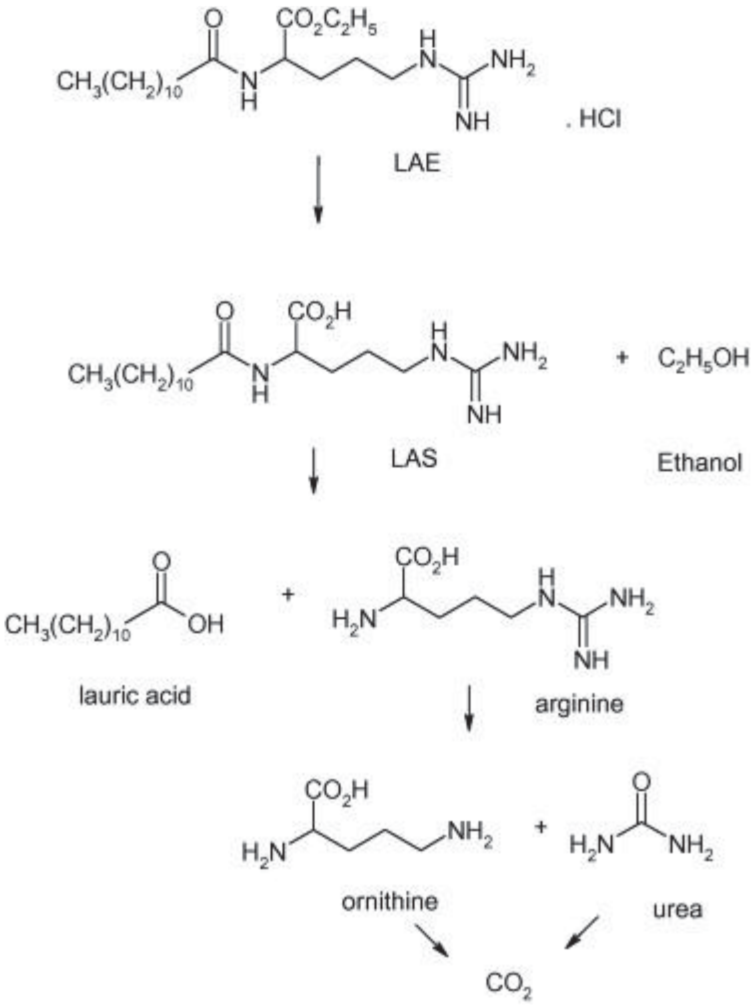

Figure 4: Proposed pathway for the *in vivo* mammalian metabolism of LAE (Hawkins et al., 2009).

The pathway shown in Figure 4 was established from *in vitro* and *in vivo* metabolism studies conducted by the Huntington Life Sciences Limited (2001), Ruckman et al. (2004); European Food Safety Authority (2007) and Laboratories Miret S.A. (2008). In humans, LAE is rapidly hydrolysed by loss of the lauroyl side-chain to form L- arginine ethyl ester and/or by cleavage of the ethyl ester to form N^{α}-lauroyl-L-arginine (Hawkins et al., 2009). Further hydrolysis of either intermediate results in the production of amino acid L-arginine. Once formed, the L-arginine, as a normal dietary constituent, is metabolized to ornithine, urea and carbon dioxide by well established pathways (Guoyao & Morris, 1998). Ornithine can be incorporated into endogenous compounds via the urea and citric acid cycles and finally degraded to carbon dioxide. EFSA concluded that, on ingestion in humans, LAE is broken down to products of normal metabolism (European Food Safety Authority, 2007). Based on the data collected from human studies, which show a higher plasma concentration of arginine as compared to N^{α}-lauroyl-L-arginine, Hawkins et al. (2009) suggested that the majority of LAE is metabolized prior to absorption.

2.7.4 International Regulatory Status of Ethyl Lauroyl Arginate (LAE)

In September 2005, the United States Food and Drug Administration (USFDA) issued a letter of No objection regarding a submission that LAE is Generally Recognised as Safe (GRAS) for use as an antimicrobial at level of upto 200 mg/kg of ethyl- N^{α}-lauroyl-L-arginate HCl in meat and poultry products.

In 2006, the Food Safety Inspection Service (FSIS) Directive 7120.1 summarized the approval of LAE as a safe and suitable ingredient for comminuted meat products, fresh cuts and RTE meat and poultry products.

In April 2007, the European Food Safety Authority (EFSA) issued the opinion of the Scientific Committee on LAE as a new food preservative for use in a range of food categories. EFSA established an ADI of 0.5mg LAE/kg body weight/day (European Food Safety Authority, 2007).

In June 2008, during the 69th session of the Joint FAO/WHO Expert Committee on Food Additives (JECFA), the proposal of LAMIRSA on LAE as a food additive was reviewed and an ADI (Allowable Daily Intake) of 4 mg/kg of body weight/day was allocated for LAE (JECFA, 2008).

Food Safety Authority of Australia and New Zealand (FSANZ)

In August 2008, an application from Laboratories Miret SA (LAMIRSA) was submitted to FSANZ to approve LAE as a preservative in a number of food groups to control food spoilage microorganisms. The assessment commenced in September 2008 and the preservative was approved for use in food products in 2009 (FSANZ, 2009b).

3.0 MATERIALS AND METHODS

High pressure processing parameters (pressure, hold time) were selected for this study based on supporting literature published with regard to HPP of RTE meat products (Garriga, Aymerich, Costa, Monfort, & Hugas, *In Press;* Garriga et al., 2004; Hayman et al., 2004; Jo et al., 2011; Kilpatrick & Patterson, 1998; Linton, McClements, & Patterson, 2004). Concentrations of LAE were selected based on the limits outlined by the Food Safety Authority of Australia and New Zealand (FSANZ, 2009a).

The experiment was conducted in two phases. In the first phase (screening), 14 treatments were subjected to microbial, color and texture tests. Selection of treatments was done in the second phase, where treatments that significantly ($P<0.05$) inhibited the growth of microorganisms for atleast 16 weeks, with insignificant ($P>0.05$) changes in color and texture were selected for sensory evaluation. Alongside the sensory analysis, the samples from the selected treatments were also subjected to microbial, color and texture analysis.

The preparation of the samples and methods for the analysis (microbial, color and texture) of the samples were consistent for both phases of the study.

3.1 Experimental Plan

Phase 1: Screening of treatments

A 2×2 multifactor nested block design was designed for this study (Montgomery, 2008; Shah & Sinha, 2006). In this design, two types of treatments were investigated, comprising samples treated with (i) High Pressure Processing (HPP) and (ii) Lauric Arginate (LAE). The levels of the LAE treatment were similar but not identical for different levels of each HPP treatment; therefore making it unique to that particular treatment. The HPP treatments comprised of two factors: pressure tested at two levels (450 MPa and 600 MPa) and process time tested at three levels: 1 min, 3.5 min and 5 min, giving a 2×3 factorial design (nested) within the main experimental plan. The factor structure in this 2×3 factorial experiment within a 2×2 factorial experiment is shown in Table 4.

Table 4: Block design of the nested experiment showing the multiple factors within the treatments.

		HPP		
		Yes		**No**
LAE (200 ppm)	**Yes**	**450 MPa**	**600 MPa**	**LAE alone (200 ppm)**
		1 min	1 min	
		3.5 min	3.5 min	
		5 min	5 min	
	No	**450 MPa**	**600 MPa**	**Control**
		1 min	1 min	
		3.5 min	3.5 min	
		5 min	5 min	

A control with no preservatives was included in the study and combination treatments of LAE (200 ppm) with HPP were investigated as shown in Table 4. RTE sliced chicken breast roast samples were prepared at the factory of Tegel Foods Limited, Christchurch, while HPP of the samples was done at the University of Auckland, Auckland City.

Fourteen treatments were coded as shown in Table 5.

Table 5: Factorial design of treatments for the shelf-life extension of RTE cooked chicken breast roast

No.	Treatment code	Treatment type	Treatment pressure (MPa)	Process time (min)
1	HPP/450/1	HPP alone	450	1
2	HPP+LAE/450/1	HPP+LAE	450	1
3	HPP/600/1	HPP alone	600	1
4	HPP+LAE/600/1	HPP+LAE	600	1
5	HPP/450/3.5	HPP alone	450	3.5
6	HPP+LAE/450/3.5	HPP+LAE	450	3.5
7	HPP/600/3.5	HPP alone	600	3.5
8	HPP+LAE/600/3.5	HPP+LAE	600	3.5
9	HPP/450/5	HPP alone	450	5
10	HPP+LAE/450/5	HPP+LAE	450	5
11	HPP/600/5	HPP alone	600	5
12	HPP+LAE/600/5	HPP+LAE	600	5
13	LAE (200)	LAE alone	-	-
14	CONTROL	Control	-	-

Phase 2: Selection of treatments

After screening the 14 treatments in Phase 1, the treatments that significantly ($P<0.05$) inhibited the growth of microorganisms for atleast 12 weeks, with insignificant ($P>0.05$) changes in color and texture were selected for sensory evaluation. Methods and analysis are explained in detail in further sections.

3.1.1 Description of process factors

Description of the Treatments

The shelf-life of poultry products can be significantly extended by treating the product with high pressure (HPP) (Diez et al., 2009; Pal, Labuza, & Diez-Gonzalez, 2008; Ramirez-Suarez & Morrissey, 2006) and with a novel preservative, ethyl lauroyl arginate (LAE) (Seemeen & Mutukumira, 2010). Treatments investigated in this study, were HPP alone, LAE alone at 200 ppm concentration and HPP in combination with LAE (200 ppm).

Process pressure

Process pressure refers to the pressure held during the sample treatment. The treatments HPP alone and HPP+LAE (200 ppm) were subjected to two levels of pressures, 450 MPa and 600 MPa. The pressures were adjusted on the HPP equipment at the time of processing.

Process time

The following parameters are included in process time:

- *Come-up time:* the time required to increase the pressure of the sample from atmospheric pressure to the target process pressure. The come-up time likely depends upon the rate of compression of the sample and pressure-transmitting fluid and is proportional to the power of the pump used and the target process pressure.

- *Processing time:* the constant pressure holding time is the interval between the end of compression to the beginning of decompression. For successful industrial applications, short-processing times must be targeted since process time has a significant effect on the commercial economics of HPP. A processing time <5 min is preferred to maximize productivity and economically justify use of the technology (Robbins et al., 2004).

The sample treatments were subjected to three processing times which were 1 min, 3.5 min and 5 min for 450 MPa and 600 MPa respectively.

- *Decompression time:* The time to bring a food sample from process pressure to near atmospheric pressure.

3.2 Industrial preparation of RTE chicken breast roast

Preparation and processing of the chicken breast roast were done according to report by Seemeen and Mutukumira (2010). Pilot-scale production of the chicken breast roast was carried out at Tegel Foods Limited, Christchurch. Fresh donor (raw chicken breast) was obtained from the primary processing plant which was maintained at a temperature of 4°C. The donor contained about 4% fat, 8% sodium and 47% proteins.

The donor was weighed out in two portions of 40 kg each. One batch served as the control with no preservative added and the other batch contained 200 ppm of LAE as a preservative. The two portions of donor were transferred to two stainless steel bins in the chill room (0-2°C). A marinade was prepared for the two batches of donor by mixing the ingredients with water using a commercial formulation (Appendix 1). Each mix was blended separately using an electric blender for approximately 2 min until the ingredients were uniformly suspended in the marinade mix. The donor was loaded into a Lutecia vacuum tumbler (Figure 5) with the marinade mix and tumbled for 90 minutes. The consistency of the meat was checked visually after 90 min to ensure that the marinade had been absorbed by the meat. Marinating is a traditional culinary technique that is used to tenderize and to improve flavour and juiciness of poultry meats (Volpato, Zandonai Michielin, Salvador Ferreira, & Cunha Petrus, 2008). Sodium chloride, polyphosphates and sugars are considered important ingredients of marinades, as they improve meat tenderness and flavour (Lemos, Nunes, & Viana, 1999). Marinating also increases water binding capacity of meats, thus reducing cooking losses and improving meat juiciness (Brotsky, 1976; Sackett & Froning, 1985).

Figure 5: Donor and marinade loaded into the tumbler prior to tumbling

The two batches of marinated meat were transferred from the tumbler to two separate stainless steel bins and filled into collagen casings (Globus Food Packaging and Equipment, Australia) using a Risco filling machine (RS 3005, Risco SpA, Thiene, Italy). Six logs of chicken, each weighing approximately 6 kg were obtained after filling. Colored netting was used on the casings to distinguish between the two treatments. The treatments were lined on a trolley with the LAE treatment on the top shelf and the control treatments with no preservative on the lower shelf to prevent the effect of any vapors that may be released from the LAE-treated meat on the control. The trolley was then moved into an oven to begin the cooking process. A temperature probe (Part number-16002, Sensing Devices Limited, UK) was inserted into a probe log to record the internal temperature of the product during the cooking process. An oven (HR 1 Wg 256-1000/01, Schröter®, Germany) equipped with forced convection air-steam was used to cook the chicken logs. Steam in the air –steam forced convection oven was supplied by a water boiler maintaining humidity at 95% during cooking. The chicken logs were cooked to an internal temperature of 75°C for 5 min.

After cooking, the all the products were transferred to the chill room (0-2°C) for further-processing. Each log of chicken was sliced using a Weber® slicer (SKC 304, Weber Columbit, Germany) to uniform slices of 3mm thickness. The slices were cut into two equal halves and packed into SP21 retail packs (Sealed Air, Wellington, NZ) (Figure 6) to give an approximate weight of 30 grams per package followed by vacuum-packaging (R240, Multivac ®, Wolfertschwenden, Germany) (Figure 7).

Figure 6: Cutting slices of cooked chicken for vacuum-packaging.

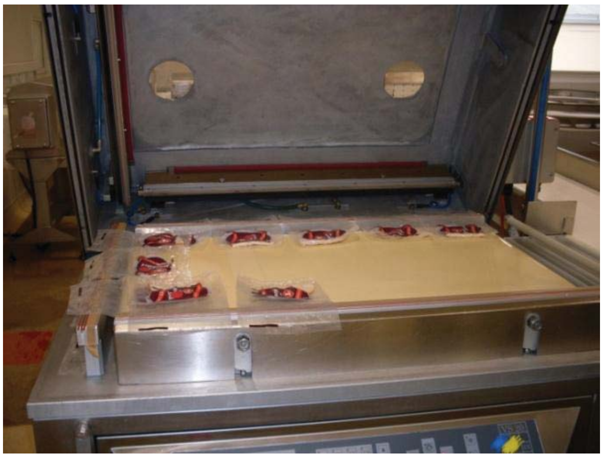

Figure 7: Sample layout in the vacuum-packaging machine (R240, Multivac ®, Wolfertschwenden, Germany).

The retail packs were packed into 5 kg capacity cartons, followed by Danband Premium 12.00mm×0.6 mm blue taping (ITW Plastic Strapping Operations, NZ), and coded. The cartons were transferred to the warehouse chiller for approximately 3 hours before loading into a temperature-controlled truck (1-2°C), and then dispatched to Massey University, Auckland. A TempRecord™ temperature recorder (Argus Real Cold Limited, NZ) was placed in one of the cardboard boxes to monitor the temperature of the samples during transportation (Appendix 2).

A detailed flowchart of the industrial production process of RTE chicken breast roast is shown in Figure 8.

The samples packed into cardboard boxes were delivered to Massey University, 4 days after preparation at Tegel Foods Ltd, Christchurch.

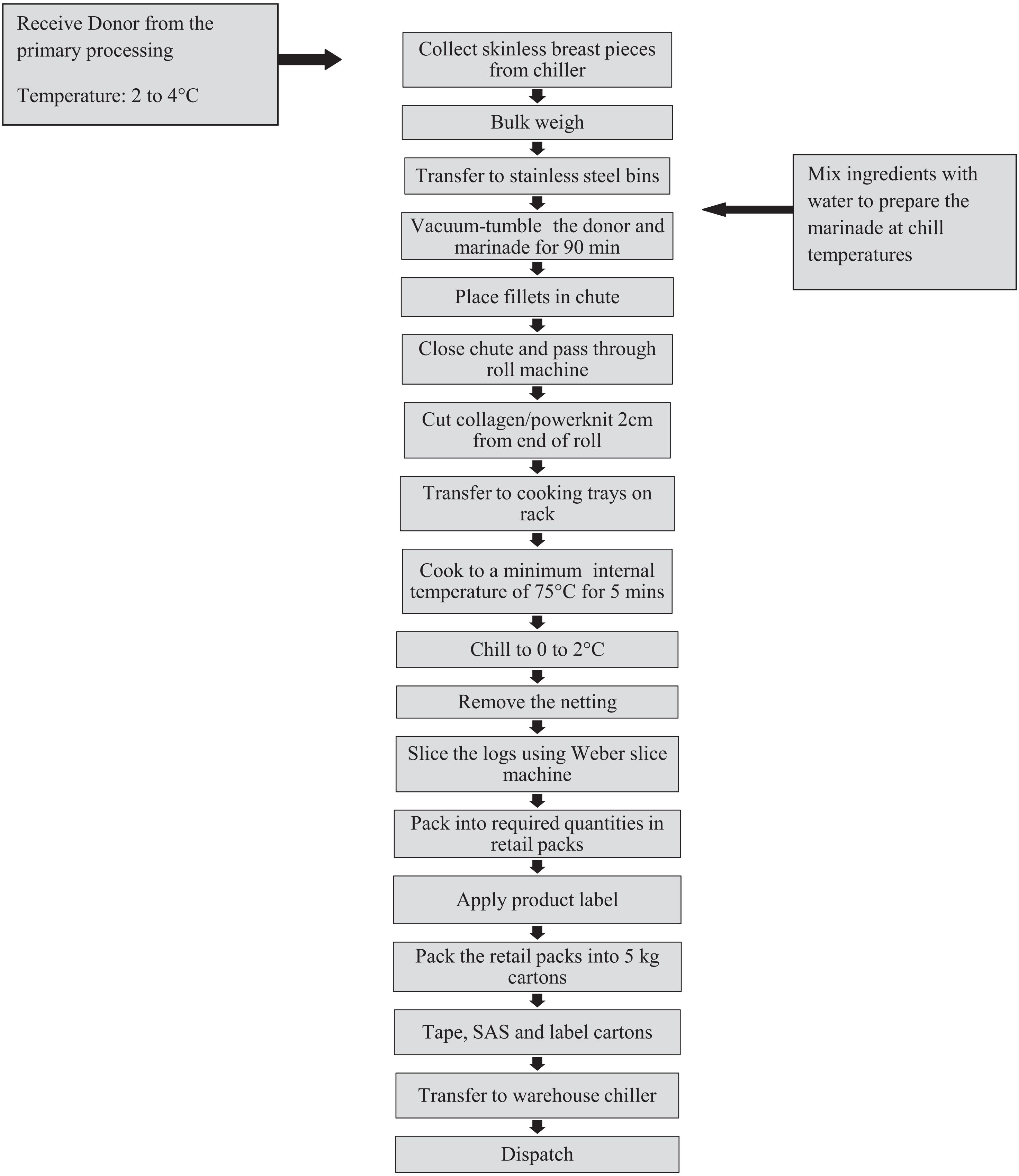

Figure 8: Industrial production process of RTE sliced chicken breast roast

3.3 High Pressure Processing

3.3.1 Description of the Avure™ High Pressure Processing Unit

The cooked and packaged samples were subjected to 'further-processing' in the 2 L Avure High Pressure Press (Avure™ Technologies, USA), with a 100 mm diameter cylinder and 254 mm height at maximum pressure. The process module comprised of a main cabinet which contained the following pressure containment system: vessel, frame and closure lifting device. The dual-wall high pressure vessel had an inbuilt 'leak-before-break' channel on linear and non-linear threaded closures. A rail mounted yoke frame contained the axial force of closures. Non-threaded closures were equipped with high pressure seals. The connection for pressure medium was located in the upper closure. The upper closure was equipped with an automatic vent for de-aeration of the vessel prior to a pressure cycle, and a thermocouple fed through with two Type K thermocouples, that were immersed in the pressurization fluid during treatment, to monitor the temperature of the samples. The pneumatic closure lifting device inserted and removed the top cover during loading and unloading of the press.

3.3.2 Sample loading and processing

The high pressure system used an intensifier-based electro-hydraulic pump to pressurize the vessel. Pressure, temperature and process time set-points were entered into the automated control system. About 15 samples were loaded into the cylinder of the HPP equipment in a single pressure cycle (Figure 9) and the equipment was closed for the pressure transmission (Figures 10 and 11 a, b). After the vessel was closed, the system automatically filled with potable water, and the pressure cycle was initiated. The initial temperature of the water was 23°C and the temperature increase due to adiabatic heating was approximately 3°C per 100 MPa. The pressure come-up time was approximately 20-25 seconds per 100 MPa. The decompression or pressure-release time was approximately between 15 and 30 seconds for 450 MPa and 600 MPa respectively. The duration of the pressure treatment did not include the come-up or pressure release time. After depressurization, the vessel was opened and the pressurized samples were removed and wiped dry using paper towels, before being transferred to an icy chilly bin maintained between 6-7°C.

Figure 9: Samples being packed into the 2L HPP equipment cylinder

Figure 10: Securing the 2L cylinder by screwing to the upper closure of the pneumatic lifting device.

(a) (b)

Figure 11: 2L cylinder being lowered to close the frame of the vessel for processing. (a) lowering of the cylinder into the pressure vessel (b) Rail mounted yoke frame showing closure of the equipment prior to pressurization.

The samples were subjected to pressure treatment in two batches. One batch of products was treated with HP alone at 450MPa and 600 MPa with process times of 1 min, 3.5 mins and 5 mins respectively. The other batch of products was HP treated in combination with LAE (200 ppm) as an ingredient of the product. Similar times and pressures were employed for the second batch of products. A flow schematic diagram of the Avure HPP unit used is shown in Figure 12.

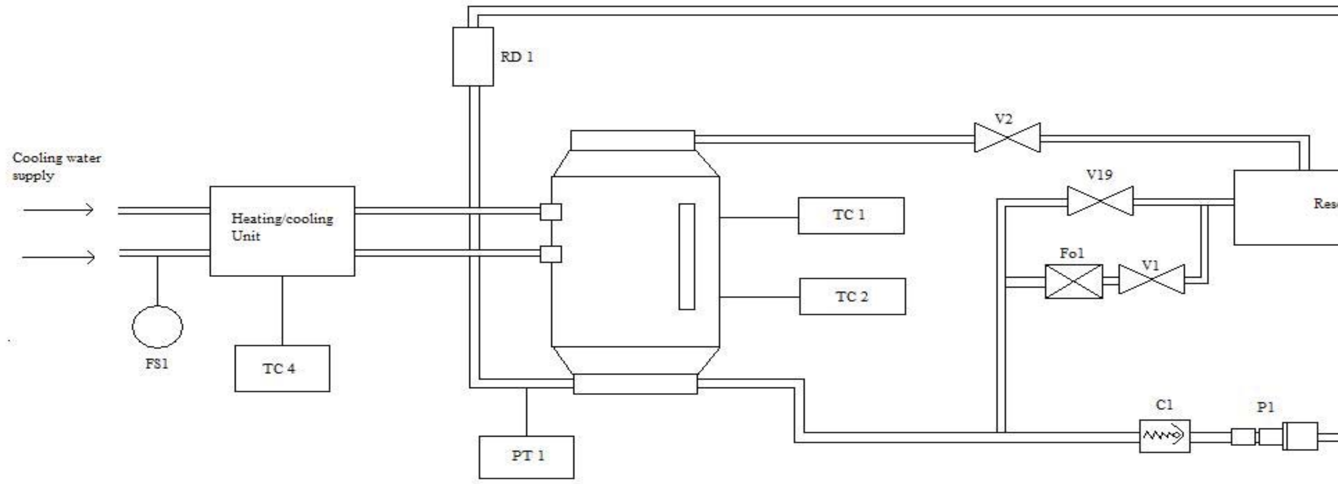

Figure 12: Flow schematic diagram of the Avure$^{\text{TM}}$ High Pressur

FS-Electromagnetic Interference Filter

TC- Thermo couple

PT- Pressure Transducer

V- Valves

C- Circuit breaker

RD1- Rupture disc that protects the vessel from over pressurizing

P-Pressure pump

F- Electromagnetic Interference power filter

LLS1- Supply reservoir

Fo1- Fixed Orifice Assembly

3.4 Analyses of the samples

After HPP treatment, the samples were stored in a Coldmaster NLT 700 (Refrigeration Engineering Company Ltd., NZ) cool store maintained at 4°C at Massey University, Albany. The samples were stored in the cool store for 16 weeks. During storage, samples were withdrawn for microbiological, color and texture analyses fortnightly up to 8 weeks. Thereafter, the samples were analysed weekly. The current shelf-life of the RTE chicken breast roast is six weeks when sodium metabisulphite is used as a preservative (Seemeen and Mutukumira, 2010).

3.4.1 Microbiological analysis

The samples were analyzed for aerobic plate counts (APCs), lactic acid bacteria (LAB) and yeasts and moulds (Y&M) during storage at 4°C for 16 weeks as described by the AOAC International (2002a) and Chung, Kim & Ha (2010). Five packaged samples were withdrawn from each treatment at every sampling interval. Random sections of each slice of chicken were cut using a sterile knife, weighing approximately 25 g, and transferred to a sterile stomacher bag using a pair of sterile forceps. Maximum recovery diluent (MRD) (Oxoid, NZ) at 0.1% (w/w) was prepared in the laboratory and added to the stomacher bag to give a total weight of 250 g. The contents of the bag (mixture) were homogenized in a stomacher for 2 min. One mL of the stomached sample was aseptically withdrawn using a pipette and sterile tips, and serial dilutions prepared in 9 mL of MRD (Downes & Itoq, 2001).

i) Enumeration of APCs and Y&M

The 3M^{TM} Petrifims^{TM} (USA) were used for the enumeration of APCs and yeasts and moulds. The petrifilms were coded and the manufacturer's directions for use were followed. One mL of each dilution was aseptically transferred to the film surface using a pipette and sterile tips. A spreader, provided with the petrifilm kit, was used to spread the surface of each film to distribute the sample adequately over the film surface and allowed to gel. The Petrifilms^{TM} for the enumeration of APCs and Y&M were transferred to an incubator maintained at 30°C for 48 h and 5 days respectively. The results were expressed as logarithm of colony forming units (cfu) per gram of sample.

ii) Enumeration of LAB

The pre-prepared MRS-agar (Fort Richard, NZ) plates were coded and 0.1 mL of each sample dilution was aseptically transferred onto the surface of the agar plate using a micro-pipette and sterile tips.

A sterile glass spreader was used to spread the surface of each plate to adequately distribute the sample over the plate surface. The petri-plates were allowed to dry and transferred to an air-tight re-sealable container. An Oxoid AnaeroGen 3.5 L sachet (Oxoid, NZ) was placed in the container to absorb the atmospheric oxygen in the container and create an anaerobic environment required for the growth of LAB. The containers were transferred to an incubator maintained at 30°C for 72 hours. The results were expressed as logarithm of colony forming units (cfu) per gram of sample.

3.4.2 Color Measurement

A Minolta CR-300 Colorimeter (Minolta Company, Tokyo, Japan) using illuminant D65 as the light source was used to measure the color of the samples. The measuring head of the CR-300 uses diffuse illumination/0° viewing geometry to measure the color of the samples as seen under diffuse lighting conditions. The Hunter's L*, a*, b* values were recorded for the samples. The L* value on a 0 to 100 scale denotes the color from black (0) to white (100). The a* value denotes redness (+) or greenness (-), and the 'b' value denotes yellowness (+) or blueness (-) (Omana et al., 2011). The instrument was calibrated using a white ceramic calibration plate with L* value 94.01, a* value +0.29

and b* value +1.77. An average of three values obtained from color measurements at three different points on the sample surface was recorded for every time point in the study period.

3.4.3 Texture measurement

i) Description of the TA-XT^{TM} plus texture analyzer

Texture measurement of samples was done as described by Bourne (1978), Peleg (1980), Sherman (1989) and Brabanti & Pasquini (2005), using a TA-XT™ *plus* texture analyzer (Stable Micro Systems, UK). The texture analyzer was connected to a PC for data logging and analysis via Texture Exponent® software, working at a crosshead speed of 10 mm s^{-1}. A Warner-Bratzel™ blade (TA-42, Stable Micro Systems, UK) was used to determine the shear force as a function of meat tenderness as described by (Barbanti & Pasquini, 2005). The probe was attached to the analyzer's base or the horizontal arm. The texture analyzer was calibrated before use. The force was calibrated at a maximum of 50 kg load cell capacity and the probe height was calibrated at 4 mm. Samples were placed on the base of the instrument and the arm of the texture analyzer which contained the load cell (50 kg) moved down, when the test was run, to compress the sample and return to its initial position (calibrated height value).

ii) Sampling for texture measurement

Each sample slice was cut to 5 cm by 5 cm pieces for the texture measurement. The shear force (g), distance (mm) and time (s) values were quantified for each sample by the Texture Expert® software. The calculated values were copied and pasted on Microsoft® Office Excel 2007 (Microsoft®, USA) for statistical data analysis. An average of six values (from six different sample slices) was recorded.

3.4.4 Consumer sensory evaluation

Consumer panels were conducted fortnightly for 16 weeks. About 20-30 panelists were recruited fortnightly with an advertising sign, to evaluate the different treatments. The evaluation was conducted between 11am and 3pm. The samples from each treatment were coded with random three-digit numbers and were served to the panelists at 6-7°C. Before tasting, panelists were familiarized with the assessment criteria, the attributes to be rated and the prescribed method for completion of the sensory evaluation form (see Appendix 6). Panelists were presented the samples in sensory booths (Massey University, Albany) under white light. The consumers were required to score the acceptability of the sample color, texture, flavour, freshness and overall liking on a 9-point hedonic scale as described by Jo et al. (2011) and Kim & Marshall (1999). The category definitions were: 9-Like extremely; 8- Like very much; 7- Like moderately; 6- Like slightly; 5- Neither like nor dislike; 4- Dislike slightly; 3- Dislike moderately; 2- Dislike very much; 1- Dislike extremely (Meilgaard, Civille, & Carr, 2007). Additionally, there was space provided for further description and comments. Panelists were asked to rinse their palate with still bottled water between samples. Participants were presented one sample at a time from each of the five treatments resulting in five total samples. All panelists signed a standard informed consent form as provided by the Researcher.

4.0 STATISTICAL ANALYSIS

Microbiological data were used to generate graphs using Microsoft® Excel 2007 (Microsoft®, USA).

Multiple comparisons of data using ANOVA

Data obtained was normally distributed, as determined by the Kolmogorov-Smirnov normality test (Appendix 8). The APCs obtained in all the treatments were analyzed using the ANOVA function of Minitab, version 15.0 (Minitab Inc., USA, 2009), to determine any significant difference between the treatments. Following the ANOVA, a Tukey's post hoc test was carried out to determine the treatments that significantly differed from each other. Similar procedures were employed to test for color and texture.

Sensory shelf-life estimation using survival analysis

Determining food shelf-life of food using survival analysis, samples at different storage times are presented to consumers. The key concept of this methodology is to focus the sensory shelf life hazard on the consumer rejecting the product (Hough, Langohr, Gomez, & Curia, 2003). In this study, overall acceptability was used to model the rejection time of the product/ sample. For samples that were rated 5 (5=neither like nor dislike) or below 5 (<5=dislike) by the consumers, the score was considered as a rejection and samples rated between 6-9 on the 9-point hedonic scale were interpreted as accepted. The storage time at which the consumer rejects the sample can be defined by a random variable T. The rejection function, $F(t)$ can be defined as the probability of a consumer rejecting a product before time t, i.e., $F(t)=P(T \leq t)$.

Storage times for the RTE sliced chicken breast roast samples were between 1 and 16 weeks. If, for example, a consumer evaluated a sample stored for 16 weeks and rated the sample 5 or less than 5, it was concluded that the consumer's rejection time was less than

16 weeks. The data for this consumer would be left-censored. If the consumer accepted the sample stored for 16 weeks, the rejection time was considered to be greater than 16 weeks. The data for this consumer would be right-censored.

The likelihood function, which is used to estimate the rejection function *F(t)*, is the joint probability of the given observation of the *n* consumers (Klein & Moeschberger, 1997)

Equation 1:

$$L = \prod_{\mathrm{ifR}} \left(1 - F(r_i)\right) \prod_{\mathrm{iff}} F(l_i)$$

where r is the set of right-censored observations and l is the set of left-censored observations. Equation 1 shows how each type of censoring contributes differently to the likelihood function.

Generally, data on rejection times are not normally distributed; instead their distribution is often right-skewed. For the evaluation of rejection times, a log-linear model is commonly chosen:

$$Y = \ln(T) = \mu + \sigma W$$

where W is the error term distribution. That is, instead of the rejection time *T*, its logarithmic transformation is modeled. In Klein and Moeschberger (1997), different possible distributions for *T* are presented, for example, the log-normal or the Weibull distribution. Choosing the Weibull distribution, the rejection function is equal to:

Equation 2:

$$F(t) = F_{sev}\left(\frac{\ln(t) - \mu}{\sigma}\right)$$

where $F_{sev}(.)$ is the rejection function of the smallest extreme value distribution: $F_{sev}(w) = 1 - \exp(-e^{w})$, and μ and σ are the model's parameters.

The likelihood function is a mathematical expression that describes the joint probability of obtaining the data actually observed on the subjects in the study as a function of the unknown parameters of the model being considered. To estimate μ and σ in the Weibull distribution, the likelihood function is maximized by substituting *F(t)* in Equation (1) by the expression given in Equation (2).

To estimate sensory shelf-life, the probability of a consumer rejecting a product, i.e., *F(t)* must be chosen. The most commonly used consumer rejection percentages are 25% and 50% (Ares, Parentelli, Gambaro, Lareo, & Lema, 2006; Cruz et al., 2010; Hough, 2006). In the present study, both the rejection percentages (25 and 50%) were chosen.

Calculations were performed using procedures of the R statistical software, version 2.13.1 (R Development Core Team, Vienna, Austria, 2011).

5.0 RESUTS

5.1 Screening of treatments

5.1.1 Microbiological Results

Microbiological quality of high pressure-treated cooked RTE sliced chicken breast roast

The initial microbial counts (APCs and Y&M) in the control and pressure-treated (450 MPa and 600 MPa) cooked chicken were at, or below, the level of detection (<1 $\log_{10}$ cfu g^{-1}) on APC Petrifilms^{TM} and Y&M Petrifilms^{TM}. The initial microbial counts for lactic acid bacteria (LAB) were also at, or below the level of detection (<2.3 $\log_{10}$ cfu g^{-1}) on MRS-agar plates incubated in anaerobic environment. For the purposes of statistical analyses, these counts were taken to be 1 $\log_{10}$ cfu g^{-1} and 2.3 $\log_{10}$ cfu g^{-1} respectively.

The LAB and Y&M counts were the same as the initial counts throughout the storage period of 16 weeks. The LAB counts were below the level of detection (<2.3 $\log_{10}$ cfu g^{-1}) for all the treatments including the control, when stored at 4°C for 16 weeks. The Y&M counts were also below the level of detection (<1 $\log_{10}$ cfu g^{-1}) for all the treatments including the control, when stored at 4°C for 16 weeks. However, variations in the growth of APCs were observed in all the HPP treatments. Viable APCs were enumerated ($\log_{10}$ cfu g^{-1}) for all the treatments (Table 6). The counts obtained were plotted on a graph against storage time (in weeks) to determine the effect of high pressure on the microbiological quality of the cooked RTE sliced chicken breast roast, when stored at 4°C (Figure 13).

The control was not HP-treated and did not contain preservatives added to it. Initial counts on the control were <1 $\log_{10}$ cfu g^{-1}. However, with increase in storage period, the counts had increased to >4 $\log_{10}$ cfu g^{-1} by week 10 and continued to increase until the end of storage at week 16. The APCs recorded at the end of storage were (5 ± 0.89) $\log_{10}$ cfu g^{-1}. HP-treatment at 450 MPa when pressurized for 1 min was able to significantly inhibit the growth of APCs in the samples until week 12 (APCs <1 $\log_{10}$ cfu g^{-1}). However, a steady increase to the magnitude of 1 $\log_{10}$ cfu g^{-1} was observed every week after 12 weeks of storage at 4°C. Similar trends were observed in the samples treated with HP at 450 MPa for 3.5 min. An increase in APCs (>1 $\log_{10}$ cfu g^{-1}) was observed from week 6, and (3.46 ± 0.28) $\log_{10}$ cfu g^{-1} was recorded at week 12 and the counts continued to increase up to (4.9 ± 0.45) $\log_{10}$ cfu g^{-1} at week 16. Samples treated with HP at 450 MPa for 5 min recorded APCs < 1 $\log_{10}$ cfu g^{-1} until week 8 and thereafter, an increase in counts was observed, with a final count of (3.24 ± 0.78) $\log_{10}$ cfu g^{-1} at week 16.

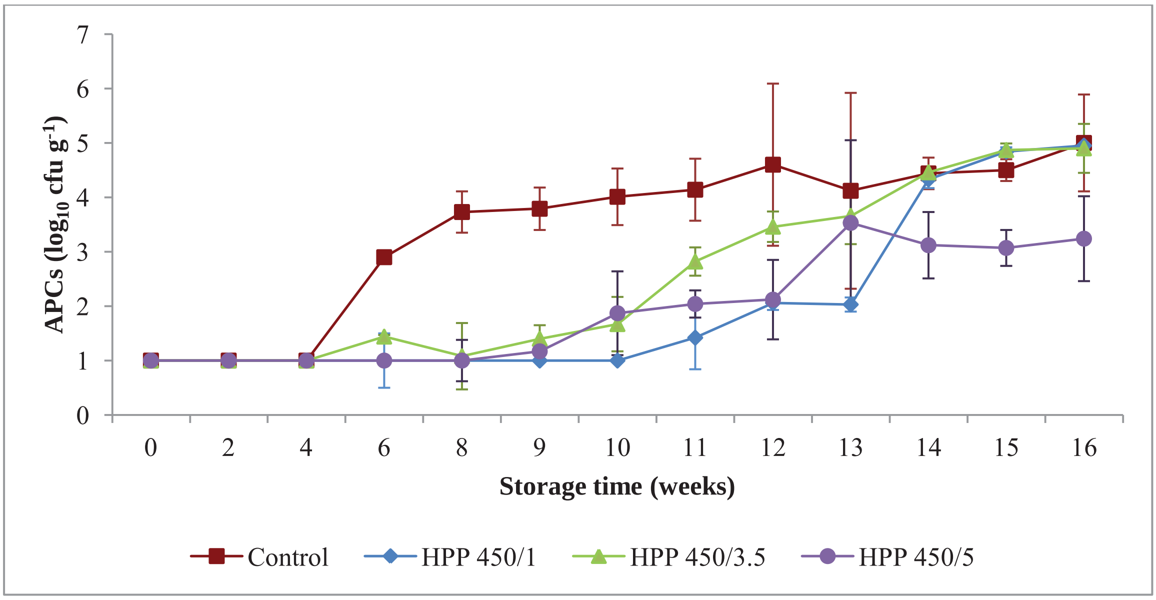

Figure 13: Changes in log APCs of RTE sliced chicken breast roast samples treated with HP at 450 MPa during storage at 4°C for 16 weeks.

The most significant effect in microbial reduction was observed in samples treated with 600 MPa which inhibited the growth of APCs for at least 9 weeks (to below detectable limits). Samples treated with 600 MPa for 1 min, 3.5 min and 5 min inhibited the growth of APCs (to below detectable limits) for 12 weeks, 9 weeks and 11 weeks, respectively (Figure 14).

It is noteworthy to mention that samples treated with 600 MPa pressure for 3.5 and 5 min showed higher APCs compared to samples treated with 600 MPa for 1 min.

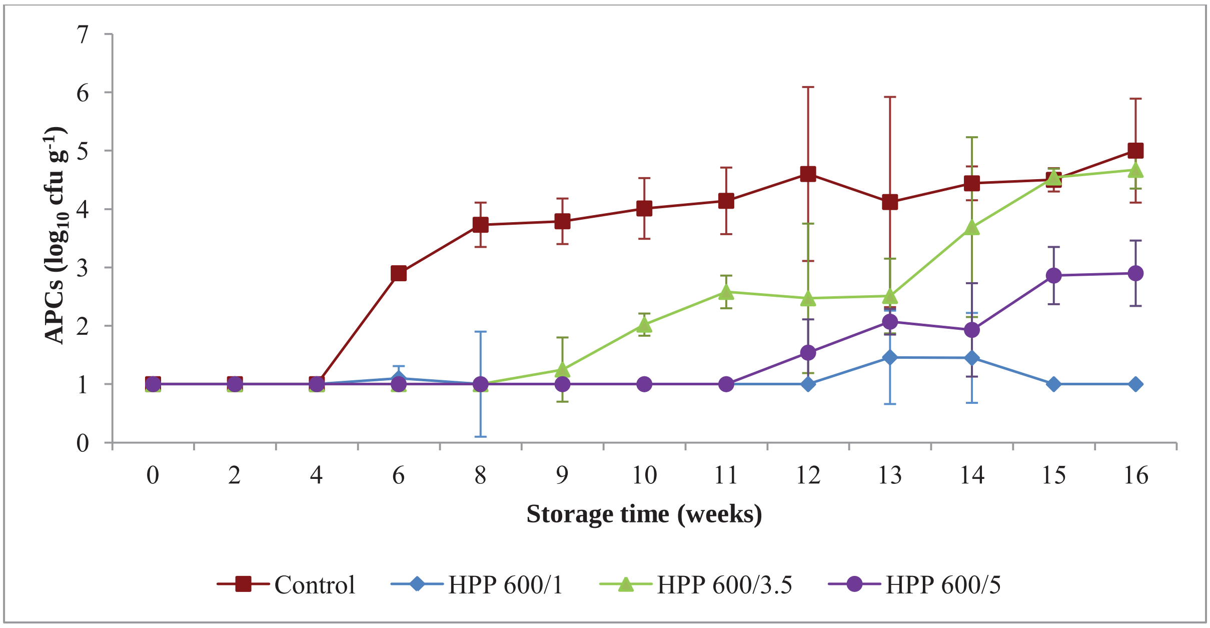

Figure 14: Changes in log APCs of RTE sliced chicken breast roast samples treated with HP at 600 MPa during storage at 4°C for 16 weeks.

Multiple comparison of the treatments revealed that there were significant ($P<0.05$) differences between the samples treated with HP at 450 MPa and samples treated with HP at 600 MPa. Significant ($P<0.05$) differences were also observed between the samples treated for 1 min, 3.5 min and 5 min at the prescribed high pressures.

Table 6: Levels of APCs* (log$_{10}$ cfu g^{-1}) in samples treated with HPP at 450 MPa and 600 MPa for 1 min respectively during storage, respectively at 4°C for 16 weeks.

Storage time (weeks)	Control	HPP 450/1	HPP/450/3.5	HPP 450/5	HPP 600/1	HPP
0	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00
2	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00
4	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00
6	^{ab}2.90 ± 0.11^{b}	1.00 ± 0.00^{a}	^{ab}1.44 ± 0.03^{c}	1.00 ± 0.00^{a}	1.10 ± 0.21^{c}	1.00
8	^{bc}3.73 ± 0.38^{c}	1.00 ± 0.00^{a}	1.08 ± 0.61^{b}	1.00 ± 0.38^{a}	1.00 ± 0.90^{a}	1.00
9	^{bc}3.79 ± 0.39^{c}	1.00 ± 0.00^{a}	^{ab}1.40 ± 0.25^{b}	^{ab}1.17 ± 0.01^{b}	1.00 ± 0.00^{a}	1.25
10	^{bc}4.01 ± 0.52^{c}	1.00 ± 0.00^{a}	^{ab}1.67 ± 0.50^{b}	^{ab}1.87 ± 0.77^{b}	1.00 ± 0.00^{a}	^{ab}2.0
11	^{bc}4.14 ± 0.57^{c}	1.42 ± 0.58^{b}	^{bc}2.82 ± 0.26 ^{b}	^{bc}2.04 ± 0.25^{b}	1.00 ± 0.00^{a}	^{ab}2.5
12	^{abc}4.60 ± 1.49^{c}	^{ab}2.06 ± 0.13^{b}	^{bc}3.46 ± 0.28^{c}	^{bc}2.12 ± 0.73^{b}	1.00 ± 0.00^{a}	^{ab}2.4
13	^{bc}4.12 ± 1.80^{c}	^{ab}2.03 ± 0.13^{b}	^{bc}3.66 ± 0.52	^{bc}3.53 ± 1.52^{c}	1.46 ± 0.80^{a}	^{ab}2.5
14	^{abc}4.44 ± 0.29^{c}	^{bc}4.33 ± 0.16^{c}	^{abc}4.46 ± 0.01^{c}	^{bc}3.12 ± 0.61^{b}	1.45 ± 0.77^{a}	^{bc}3.6
15	^{abc}4.50 ± 0.20^{c}	^{abc}4.84 ± 0.08^{c}	^{abc}4.87 ± 0.12^{c}	^{bc}3.07 ± 0.33^{b}	1.00 ± 0.00^{a}	^{bc}4.5
16	^{abc}5.00 ± 0.89^{c}	^{abc}4.95 ± 0.16^{c}	^{abc}4.90 ± 0.45^{c}	^{bc}3.24 ± 0.78^{b}	1.00 ± 0.00^{a}	^{bc}4.6

* All values are means of log$_{10}$ cfu g^{-1} ± standard deviation (n= 5 samples per sampling interval). Within rows, mea different letters are significantly different (*P* <0.05). Within columns, means preceded by different letters are signi <0.05).

Microbiological quality of combination treatment (LAE+HPP) on RTE sliced chicken breast roast

The RTE chicken breast roast samples were treated with a combination treatment of LAE (200 ppm), which was included in the marinade prior to cooking, followed by HPP after the packaging of the product. The samples were HP-treated at 450 MPa for 1 min, 3.5 min and 5 min, and at 600 MPa for 1 min, 3.5 min and 5 min. APCs were enumerated (in $\log_{10}$ cfu g^{-1}) for all the treatments (Table 7). The initial cell counts on all the sample treatments were below the level of detection (<1 $\log_{10}$ cfu g^{-1}). Samples treated with HP at 450 MPa for 1 min, 3.5 min and 5 min were able to inhibit the growth of microorganisms to below levels of detection (<1 $\log_{10}$ cfu g^{-1}) for 11 weeks (Figure 15). APCs were recorded on samples from week 12, with levels reaching >3 $\log_{10}$ cfu g^{-1} by week 14 in all the treated samples. Samples treated with HPP and LAE at 450 MPa pressure for 5 min recorded counts similar to the control from week 14 onwards. Samples treated with HPP and LAE at 450 MPa pressure for 1 min showed the lowest cell counts among the samples treated with 450 MPa pressure, recorded at (2.33±0.34) $\log_{10}$ cfu g^{-1} by week 16. LAB and Y&M counts were below detectable limits throughout the storage period for all treatments including the control when stored at 4°C for 16 weeks.

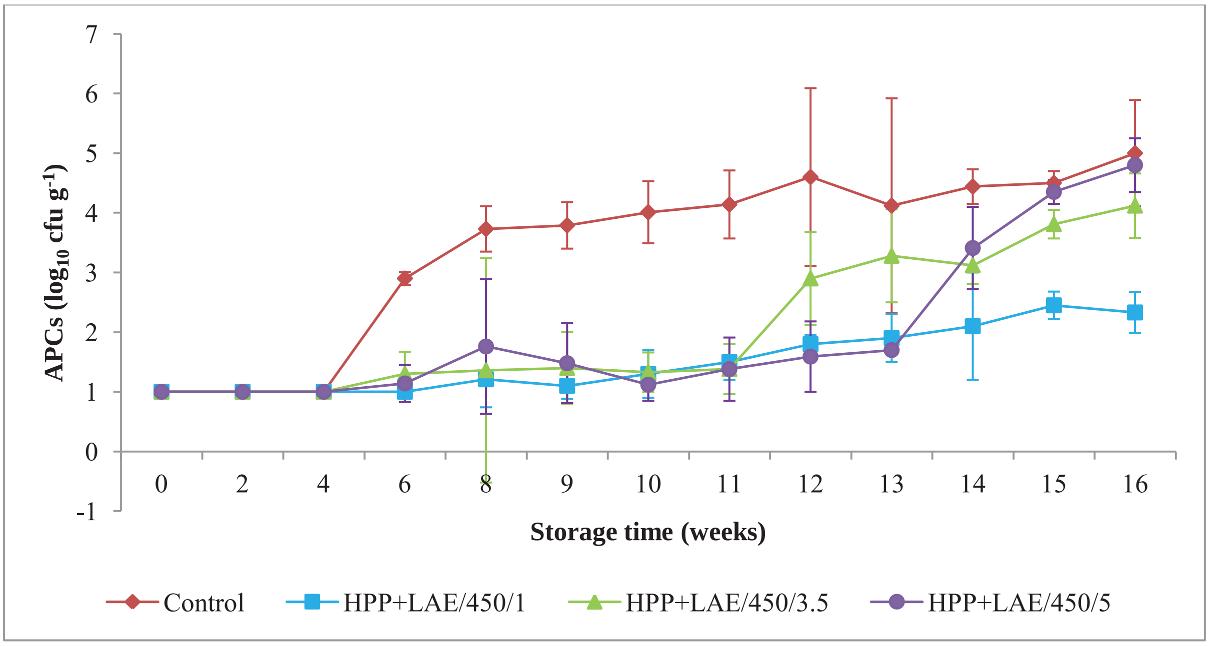

Figure 15: Changes in log APCs of RTE sliced chicken breast roast samples treated with HP at 450 MPa in combination with LAE (200 ppm) when stored at 4°C for 16 weeks.

The results of the combination treatment of LAE (200 ppm) and HP at 600 MPa are shown in Figure 16. The samples were treated at 600 MPa pressure for 1 min, 3.5 min and 5 min respectively. Multiple comparison of the treatments at 600 MPa for the three cycle times revealed no significant (*P*>0.05) differences between the cell counts obtained. Samples treated with HPP+LAE at 600 MPa pressure for 1 min, 3.5 min and 5 min recorded APCs (3.4 ± 0.85) $\log_{10}$ cfu g^{-1}, (2.8 ± 0.34) $\log_{10}$ cfu g^{-1} and (3 ± 0.41) $\log_{10}$ cfu g^{-1} respectively at the end of storage at 4°C.

Table 7: Levels of APCs* ($\log_{10}$ cfu g^{-1}) in samples treated with HPP + LAE (200 ppm) at 450 MPa min, 3.5 min and 5 min, respectively during storage at 4°C for 16 weeks.

Storage time (weeks)	Control	HPP+LAE/450/1	HPP+LAE/450/3.5	HPP+LAE/450/5	HPP+LAE/600/1	HPP+L
0	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00
2	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00
4	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00
6	^{ab}2.90 ± 0.11^{b}	1.00 ± 0.00^{a}	^{ab}1.30 ± 0.37^{a}	1.14 ± 0.31^{a}	1.00 ± 0.00^{a}	1.00
8	^{bc}3.73 ± 0.38^{c}	1.21 ± 0.47^{a}	^{ab}1.36 ± 1.88^{b}	^{ab}1.76 ± 1.13^{b}	1.00 ± 0.00^{a}	1.00
9	^{bc}3.79 ± 0.39^{c}	1.10 ± 0.22^{a}	^{ab}1.40 ± 0.60^{b}	^{ab}1.48 ± 0.67^{b}	1.00 ± 0.00^{a}	1.00
10	^{bc}4.01 ± 0.52^{c}	1.30 ± 0.40^{b}	^{ab}1.33 ± 0.33^{b}	^{ab}1.12 ± 0.27^{a}	1.00 ± 0.00^{a}	^{ab}1.20
11	^{bc}4.14 ± 0.57^{c}	^{ab}1.50 ± 0.30^{b}	^{ab}1.38 ± 0.42^{b}	^{ab}1.38 ± 0.53^{b}	1.00 ± 0.00^{a}	^{ab}1.64
12	^{abc}4.60 ± 1.49^{d}	^{ab}1.80 ± 0.15^{b}	^{bc}2.90 ± 0.78^{c}	^{ab}1.59 ± 0.59^{b}	1.00 ± 0.00^{a}	^{bc}2.94
13	^{bc}4.12 ± 1.80^{c}	^{ab}1.90 ± 0.40^{b}	^{bc}3.28 ± 0.78^{c}	^{ab}1.70 ± 0.01^{b}	1.00 ± 0.00^{a}	^{bc}2.02
14	^{abc}4.44 ± 0.29^{d}	^{ab}2.10 ± 0.90^{b}	^{bc}3.12 ± 0.31^{c}	^{bc}3.41 ± 0.69^{c}	^{ab}1.73 ± 0.69^{a}	^{ab}1.78
15	^{abc}4.50 ± 0.20^{c}	^{abc}2.45 ± 0.23^{c}	^{abc}3.81 ± 0.24^{c}	^{bc}4.35 ± 0.20^{b}	^{bc}3.26 ± 0.20^{a}	^{bc}2.51
16	^{abc}5.00 ± 0.89^{c}	^{abc}2.33 ± 0.34^{a}	^{abc}4.12 ± 0.54^{c}	^{bc}4.80 ± 0.45^{c}	^{bc}3.40 ± 0.85^{b}	^{bc}2.80

* All values are means of $\log_{10}$ cfu g^{-1} ± standard deviation (n= 5 samples per sampling interval). Within rows, mea different letters are significantly different (*P* <0.05). Within columns, means preceded by different letters are signif <0.05).

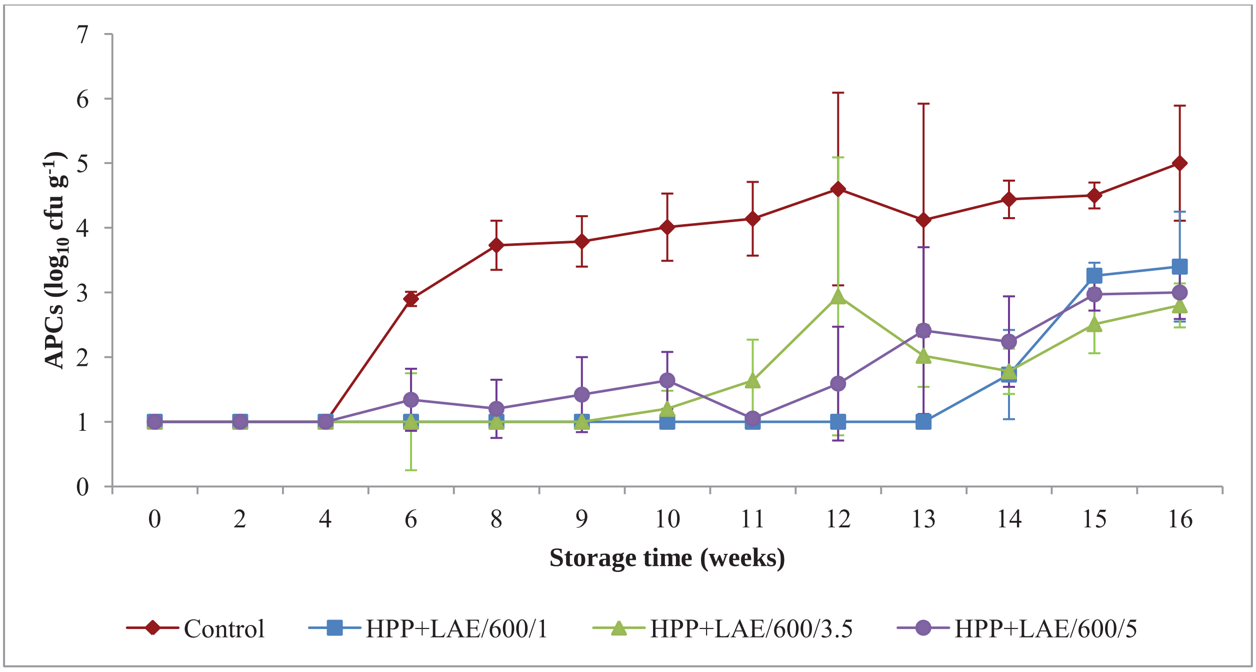

Figure 165: Changes in log APCs of RTE sliced chicken breast roast samples treated with HP at 600 MPa in combination with LAE (200 ppm) during storage at 4°C for 16 weeks.

Microbiological quality of LAE-treated RTE sliced chicken breast roast

The cooked RTE chicken breast roast samples were treated with LAE alone at 200 ppm, as an ingredient in the marinade of the product prior to cooking. The results of the APCs obtained for the LAE-treated samples are shown in Figure 17 and Table 8. The APCs were inhibited by LAE up to 9 weeks, after which a linear increase in the cell counts was observed. The initial level of APCs in the LAE-treated samples was below detectable limits, at <1 $\log_{10}$ cfu g^{-1}. However, with an increase in the storage time, the cell counts increased significantly (*P*<0.05), with >2.7 $\log_{10}$ cfu g^{-1} recorded at week 12 and >4.5 $\log_{10}$ cfu g^{-1} recorded at the end of the storage period. The counts obtained at the end of storage for the LAE-treated samples were similar to the counts obtained for the control.

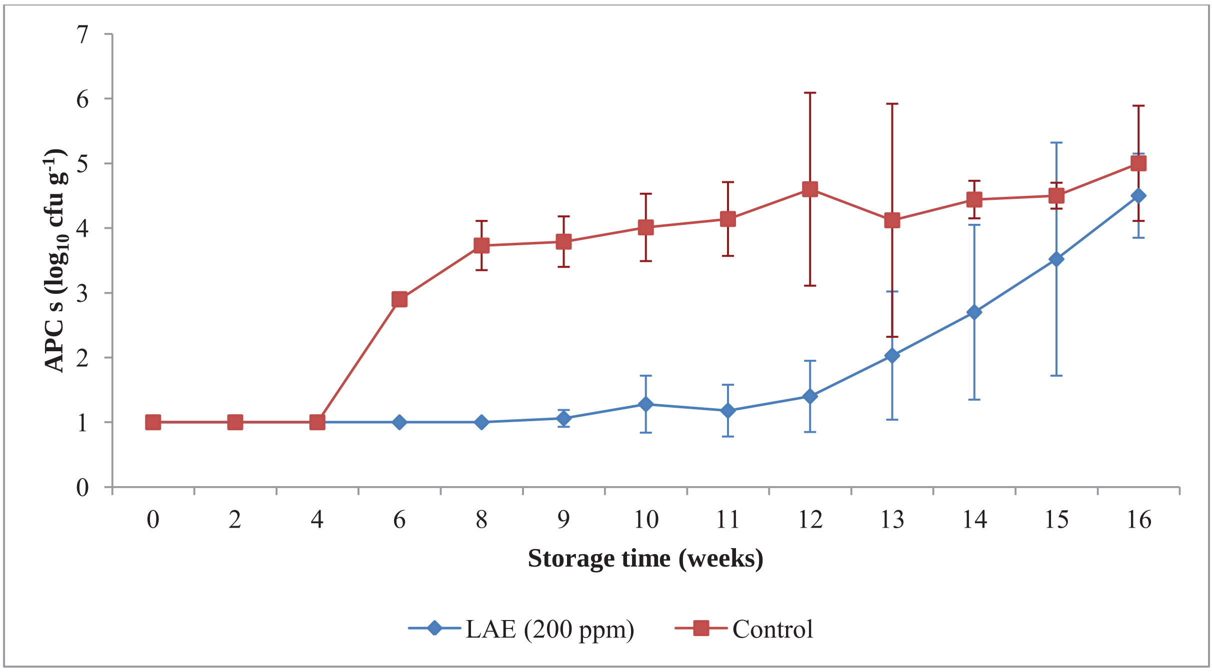

Figure 67: Changes in log APCs of RTE sliced chicken breast roast samples treated with LAE (200 ppm) during storage at 4°C for 16 weeks.

Table 8: Levels of APCs* ($\log_{10}$ cfu g^{-1}) in LAE (200 ppm)-treated RTE chicken breast roast against during storage at 4°C for 16 weeks.

Storage time (weeks)	Control	LAE (200 ppm)
0	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}
2	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}
4	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}
6	$^{ab}2.90 \pm 0.11^{b}$	1.00 ± 0.00^{a}
8	$^{bc}3.73 \pm 0.38^{b}$	1.00 ± 0.00^{a}
9	$^{bc}3.79 \pm 0.39^{b}$	1.06 ± 0.22^{a}
10	$^{bc}4.01 \pm 0.52^{b}$	$^{ab}1.28 \pm 0.40^{a}$
11	$^{bc}4.14 \pm 0.57^{b}$	$^{ab}1.18 \pm 0.30^{a}$
12	$^{abc}4.60 \pm 1.49^{b}$	$^{ab}1.40 \pm 0.15^{a}$
13	$^{bc}4.12 \pm 1.80^{b}$	$^{ab}2.03 \pm 0.40^{a}$
14	$^{abc}4.44 \pm 0.29^{b}$	$^{bc}2.70 \pm 0.90^{a}$
15	$^{abc}4.50 \pm 0.20^{a}$	$^{abc}3.52 \pm 0.23^{a}$
16	$^{abc}5.00 \pm 0.89^{a}$	$^{abc}4.50 \pm 0.34^{a}$

*All values are means of $\log_{10}$cfu g^{-1}± standard deviation (n= 5 samples per sampling interval). Within rows, mean values followed by different letters are significantly different (*P*<0.05). Within columns, mean values preceded by different letters are significantly different (*P*<0.05).

5.1.2 Instrumental color

L values of the processed samples*

Lightness, as indicated by L* value was not significantly (*P*>0.05) different in the samples treated with HP alone, irrespective of the processing pressure or holding time (Figure 18a). The control sample had L* values ranging from 66.0-72.0 during the entire storage. The lightness in control samples increased with storage time. A slight increase in the L* value was observed for the samples treated with HP and the increase was directly proportional to the applied pressure and the pressure holding time. However, the lightness of the samples did not significantly (*P*>0.05) differ between the samples. The L* value of the samples did not significantly (*P*>0.05) change over storage time. There was no significant (*P*>0.05) difference observed in the samples treated with the combination treatment of HPP and LAE (200 ppm) (Figure 18b). The samples treated with LAE (200 ppm) alone showed no significant (*P* >0.05) difference in the lightness of samples when stored at 4°C for 16 weeks (Figure 18c). The L* values for the LAE-treated samples ranged from 70-71.

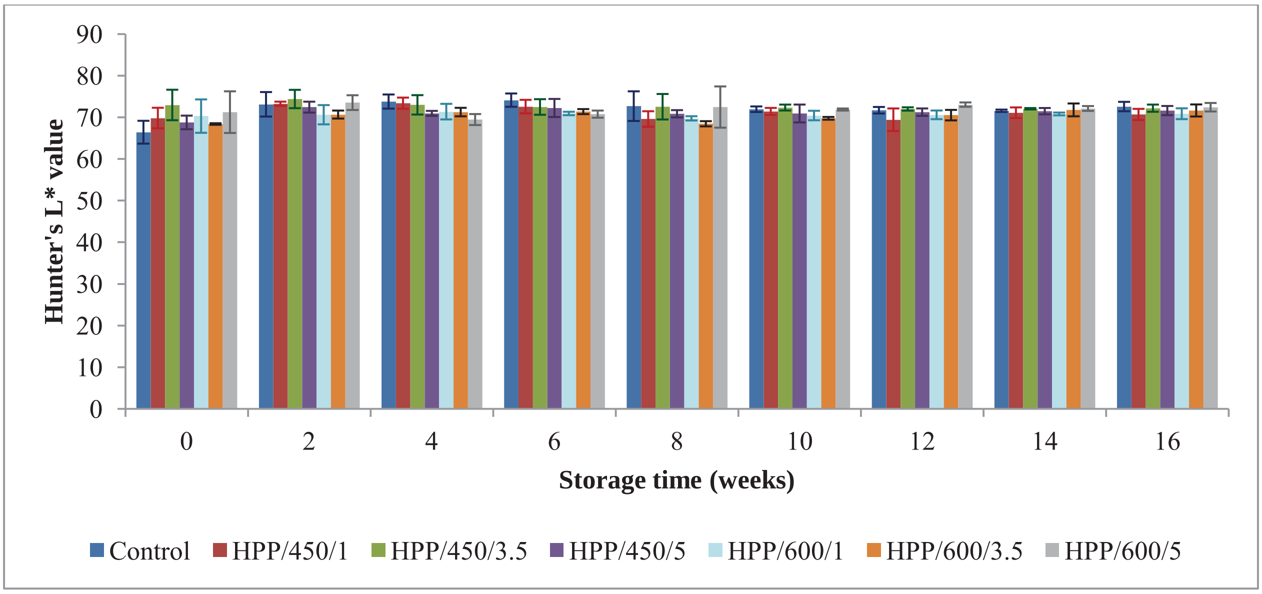

Figure 18a: Changes in Hunter's L* values of the HP-treated RTE sliced chicken breast roast during storage at 4°C for 16 weeks.

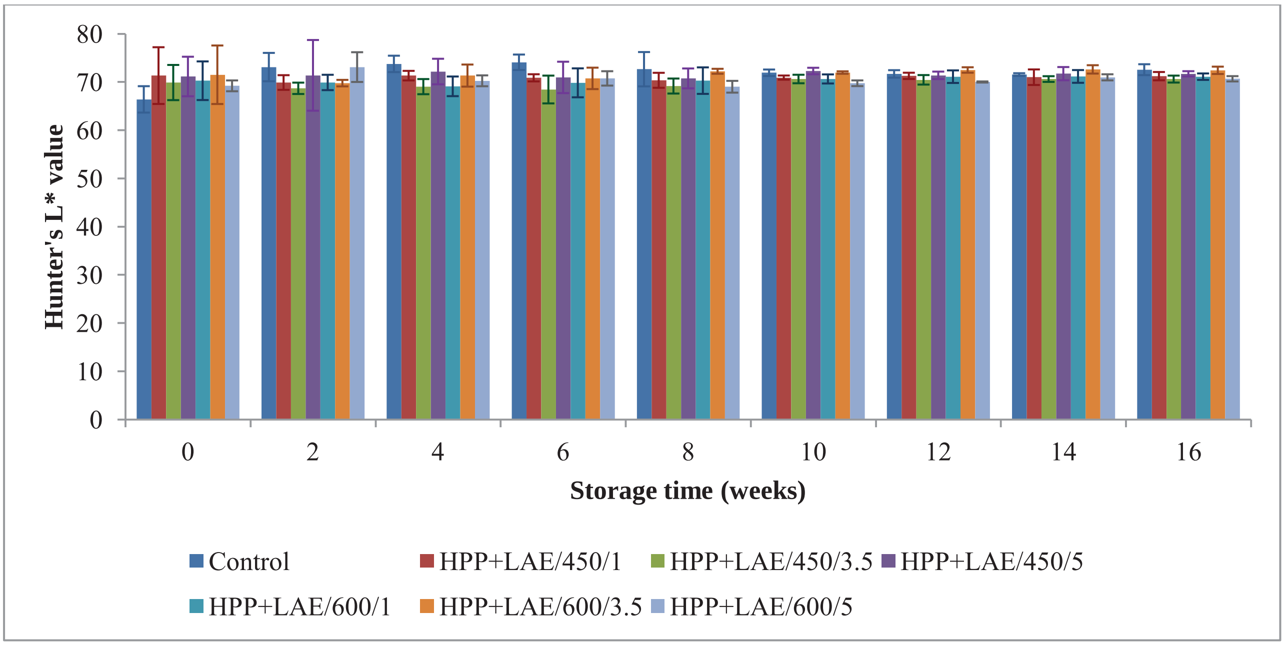

Figure 18b: Changes in Hunter’s L* values of HPP+LAE (200 ppm) treated RTE sliced chicken breast roast during storage at 4°C for 16 weeks.

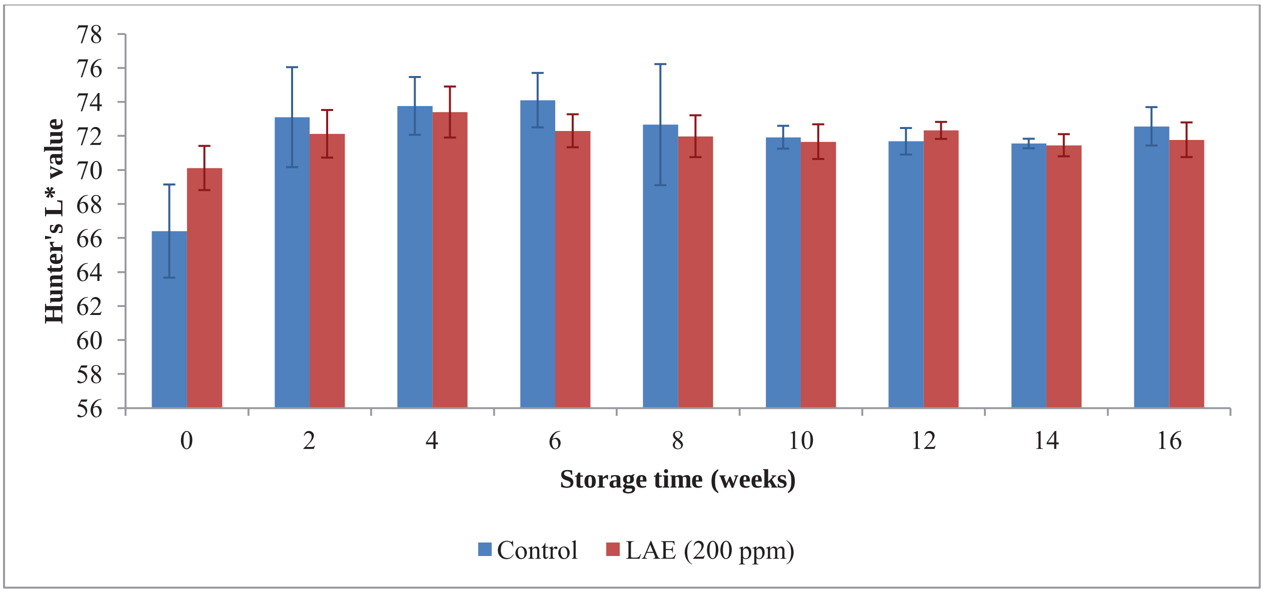

Figure 18c: Changes in Hunter’s L* values of the LAE-treated RTE sliced chicken breast roast during storage at 4°C for 16 weeks.

a values of the processed samples*

The redness, indicated by Hunter's +a* values, were variable during storage of the sample treatments including the control (Figure 19a). The initial a* value in the control samples was 2.37 which decreased with increase in storage time at 4°C. However, the decrease was not statistically significant (*P*>0.05). There was a significant (*P*<0.05) difference in the redness of samples treated with HP at 450 MPa and 600 MPa. The reduction in the a* values of the samples was directly proportional to the applied pressure and the pressure holding time. Maximum decrease in redness value was observed in the samples treated with HP for 600 MPa for 5 min. However, by the end of 16 weeks, all the samples treated with HP showed a* values in the range of 0.51-0.96. The initial a* value of the control samples was 2.37 and declined during storage time to 1.30 by week 16 (end of storage period).

The +a* values of samples treated with HP in combination with LAE (200 ppm) significantly decreased with increase in pressure and holding time (Figure 19b). The maximum reduction in the a* values was observed in samples treated with pressure at 600 MPa for 3.5 min and 600 MPa for 5 min. Samples treated with 600 MPa for 1 min showed significant (*P*<0.05) reduction in the a* values from week 8. Samples treated with the combination treatment of HP at 450 MPa and LAE (200 ppm) showed significant (*P*<0.05) reduction in a* values when treated for 1 min, 3.5 min and 5 min. However, a* values of samples treated with LAE (200 ppm) remained stable after 16 weeks of storage (Figure 19c).

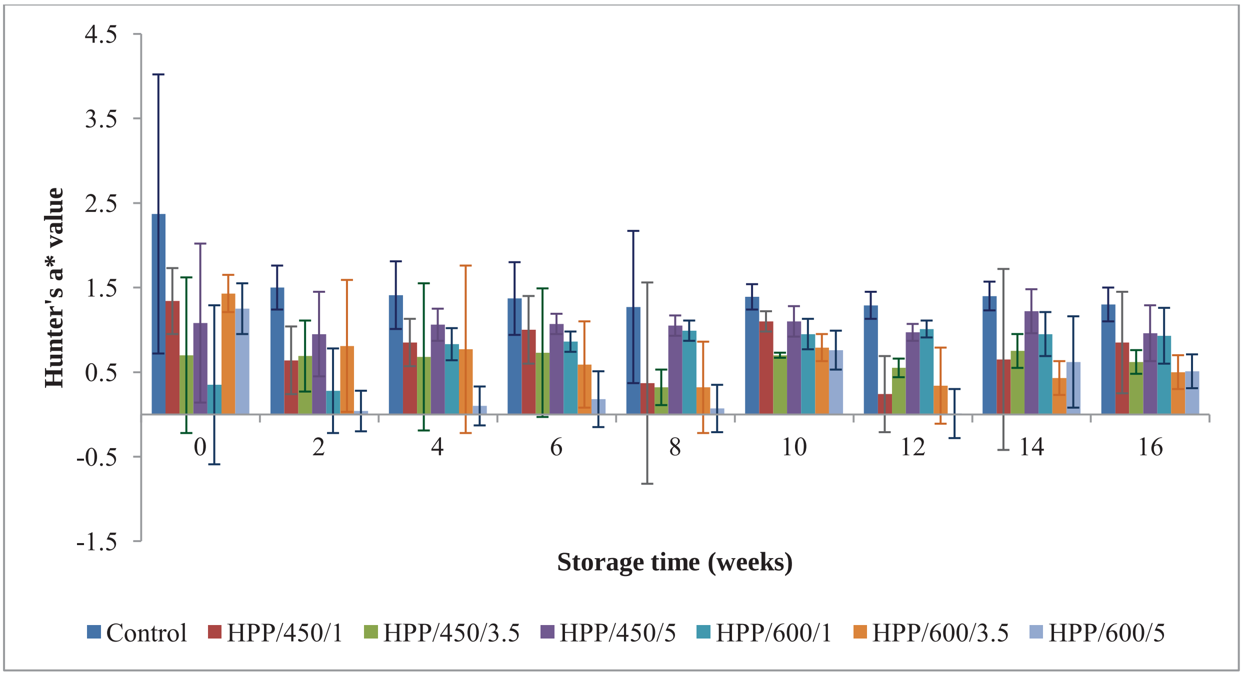

Figure 19a: Changes in Hunter's a* values of the HP-treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

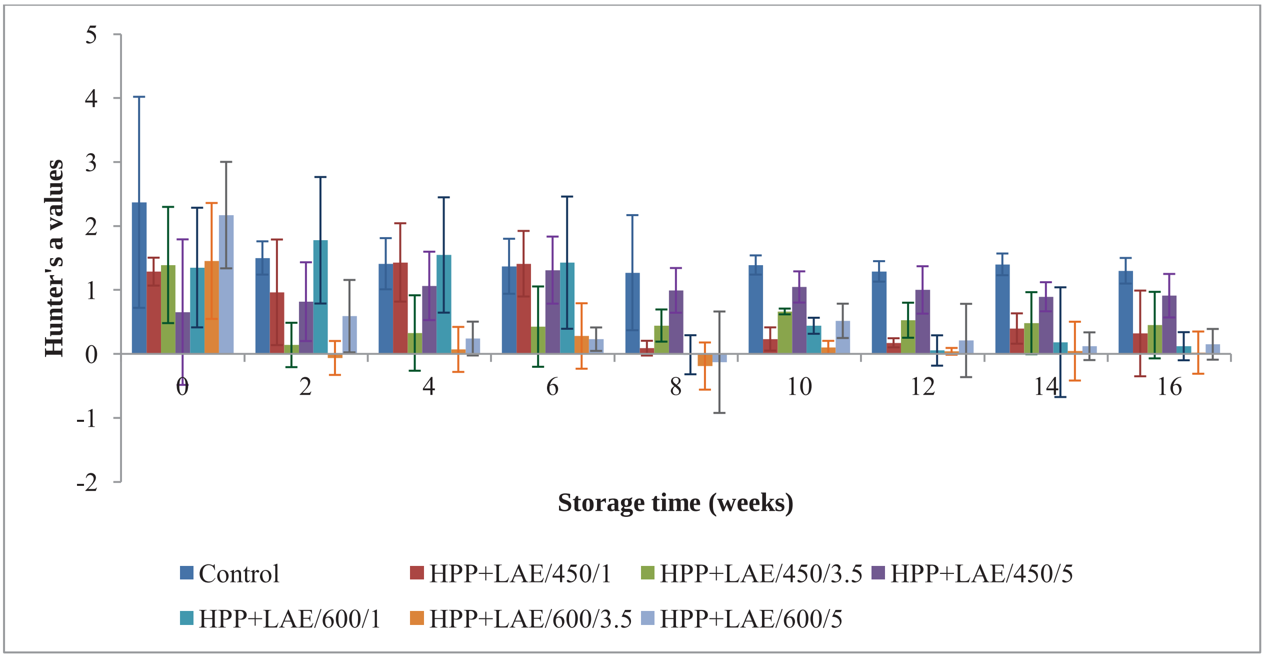

Figure 19b: Changes in Hunter's a* values of HPP+LAE (200 ppm) treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

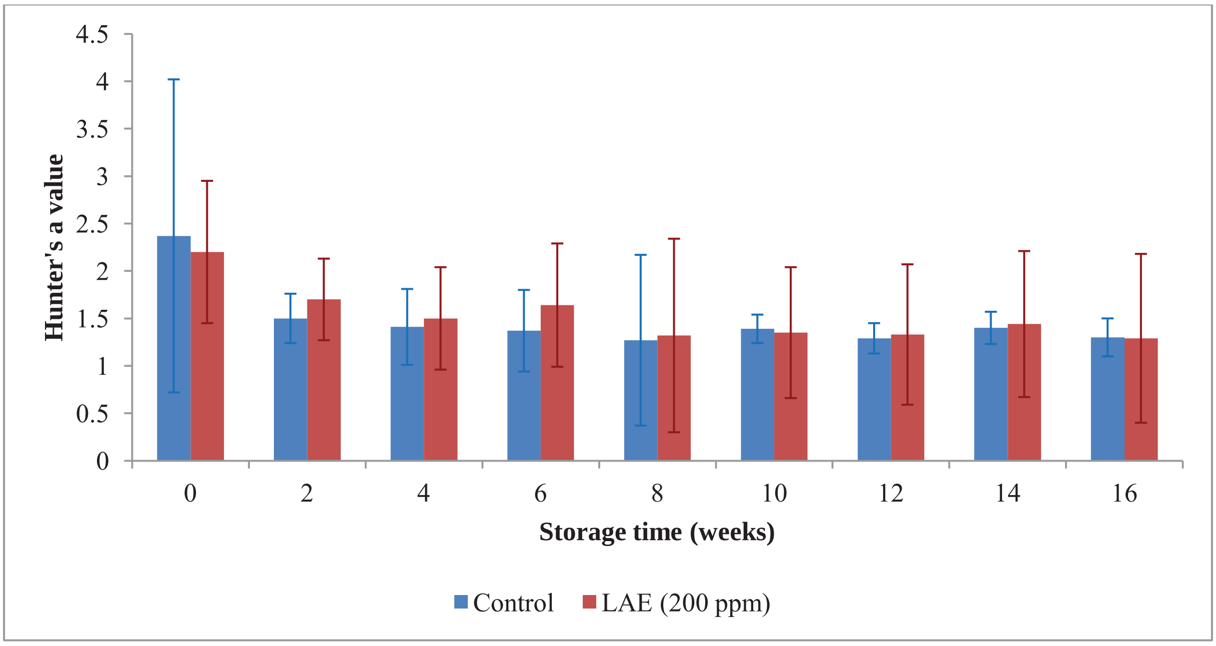

Figure 19c: Changes in Hunter's a* values of the LAE-treated RTE sliced chicken breast roast during storage at 4°C for 16 weeks.

b values of the processed samples*

Hunter's +b* value represents the yellowness of the samples as detected by the colorimeter. The b* value in the control samples at time 0 was 5.99 and increased during storage (Figure 20a). However, the change was not significant (P>0.05). The final b* value on the control samples was 6.33. A slight increase, although not significant (P>0.05) in the b* values of samples treated with HP was observed and this change was proportional to the pressure applied and the cycle time/pressure holding time. Generally, the b* values ranged from 6.00 to 7.00 for all the samples including the control. Therefore, the b* value was stable for all the samples when stored at 4°C for 16 weeks.

No significant (P >0.05) changes were observed in the yellowness of the samples treated with HP in combination with LAE (200 ppm) when stored at 4°C for 16 weeks (Figure 20b). The b* values ranged between 6.89-7.12 for samples treated with HP at 450 MPa in combination with LAE (200 ppm), during storage at 4°C for 16 weeks. No significant (P>0.05) change in the yellowness of samples was observed when samples were treated with HP at 600 MPa in combination with LAE (200 ppm). The HPP treatment alone and in combination with LAE (200 ppm) did not have a significant (*P*>0.05) effect on the b* values of the cooked RTE sliced chicken breast roast samples.

The b* values of the LAE-treated samples did not change significantly (*P*>0.05) during storage at 4°C (Figure 20c). The b* values ranged between 6 and 6.43 for samples treated with LAE (200 ppm).

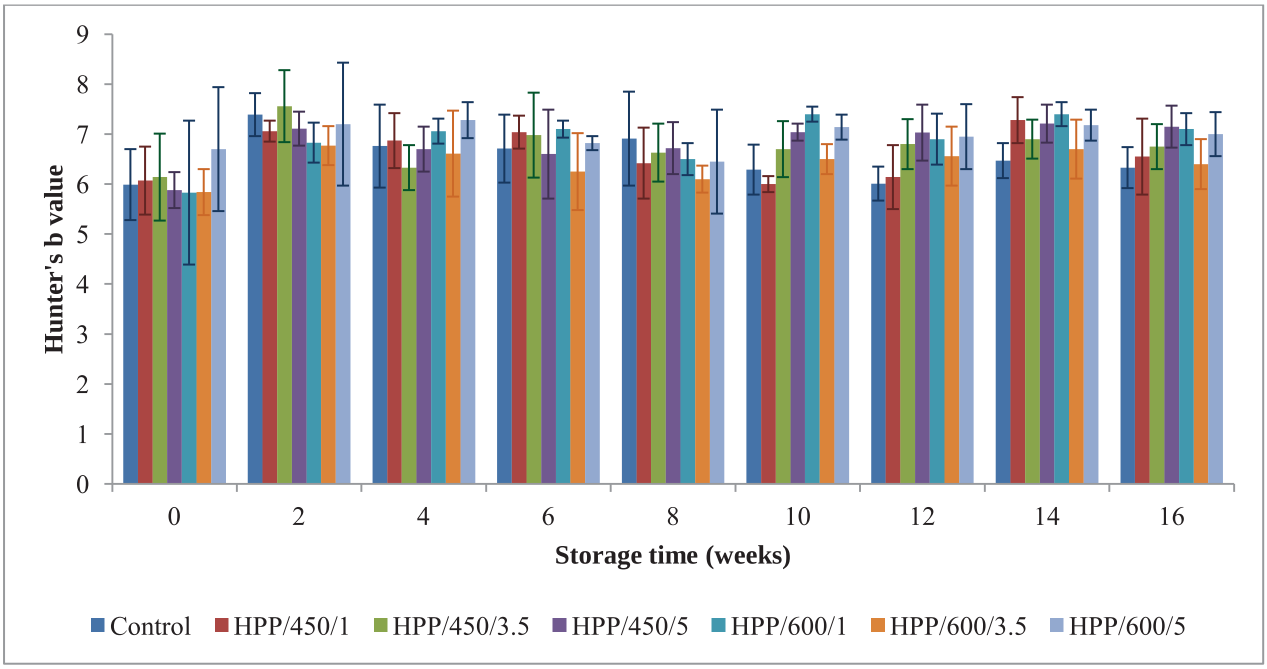

Figure 20a: Changes in Hunter's b* values of the HP-treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

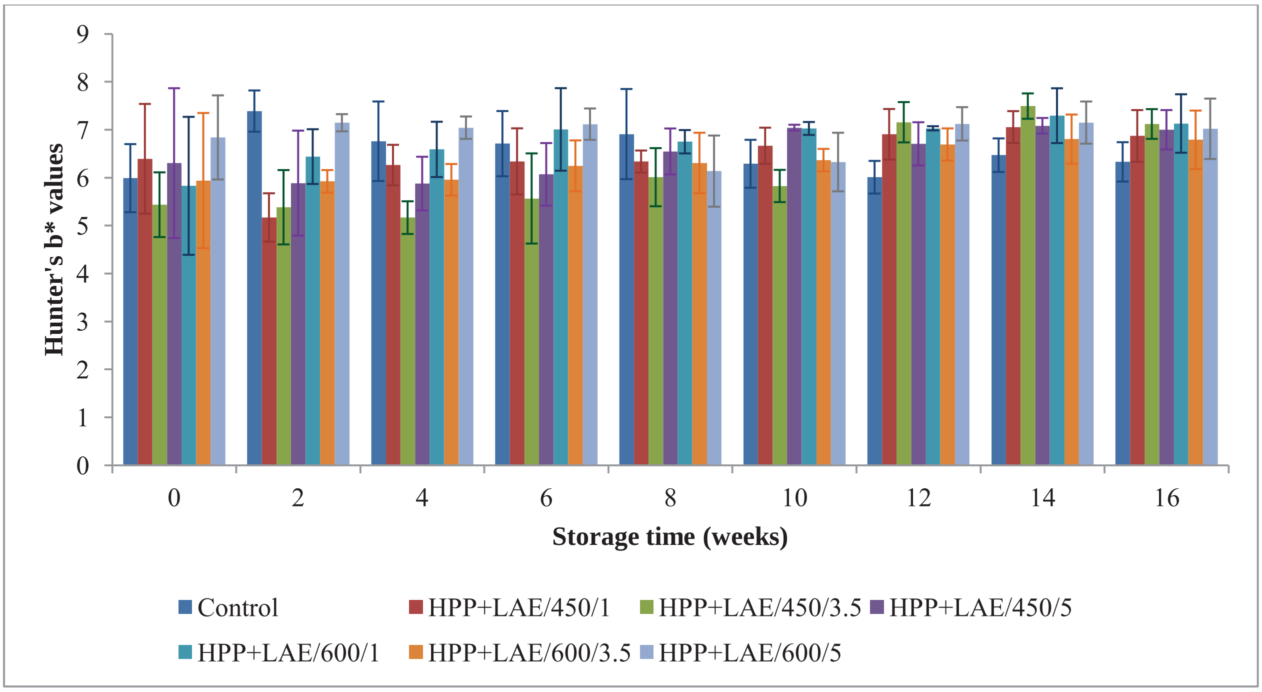

Figure 20b: Changes in Hunter's b* values of HPP+LAE (200 ppm) treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

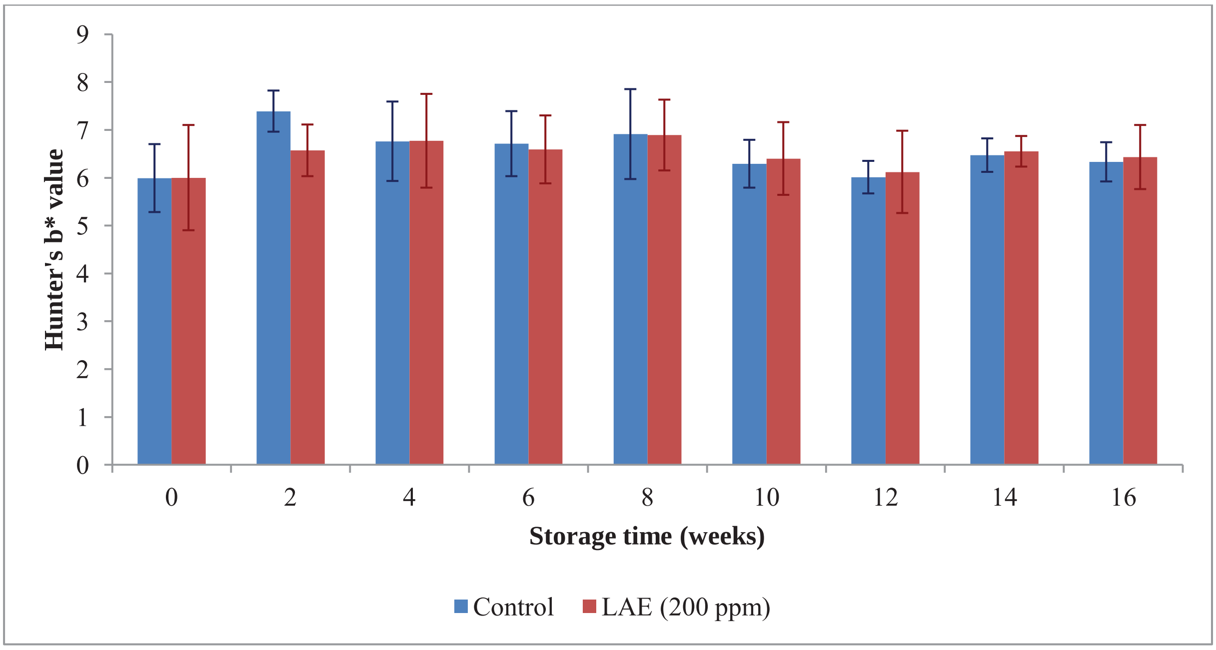

Figure 20c: Changes in Hunter's b* values of LAE (200 ppm)-treated RTE sliced chicken breast roast during storage at 4°C for 16 weeks.

5.1.3 Instrumental texture

Shear force as an index for firmness was used to measure the texture quality of the chicken samples. The sliced samples were approximately 3 mm thick. The samples treated with HP at 450 MPa and 600 MPa showed similar shear force to the control. The shear force decreased in the control samples during storage time, indicating a slight reduction in the firmness of the samples. However, the samples treated with HP at 450 MPa for 1 min, 3.5 min and 5 min were not significantly (*P* >0.05) different from the samples treated with HP at 600 MPa; the firmness in the samples was stable when stored at 4°C for 16 weeks (Figure 21a).

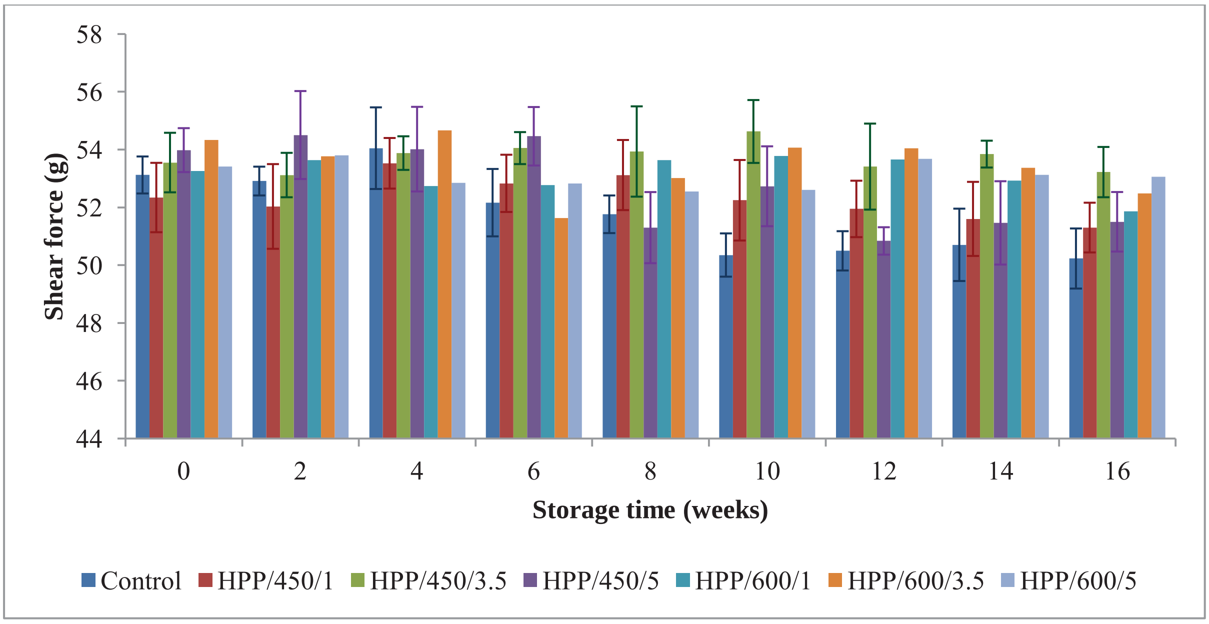

Figure 21a: Changes in shear force of HPP-treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

The samples treated with HPP in combination with LAE (200 ppm) showed no significant (*P*>0.05) change in the shear force with increase in storage time (Figure 21b). The initial shear force on LAE-treated samples was 53 g which remained stable throughout the storage time (Figure 21c). Thus, HPP or LAE, alone and in combination did not have a significant (*P*>0.05) on the texture of the cooked RTE chicken breast roast samples when stored at 4°C for 16 weeks.

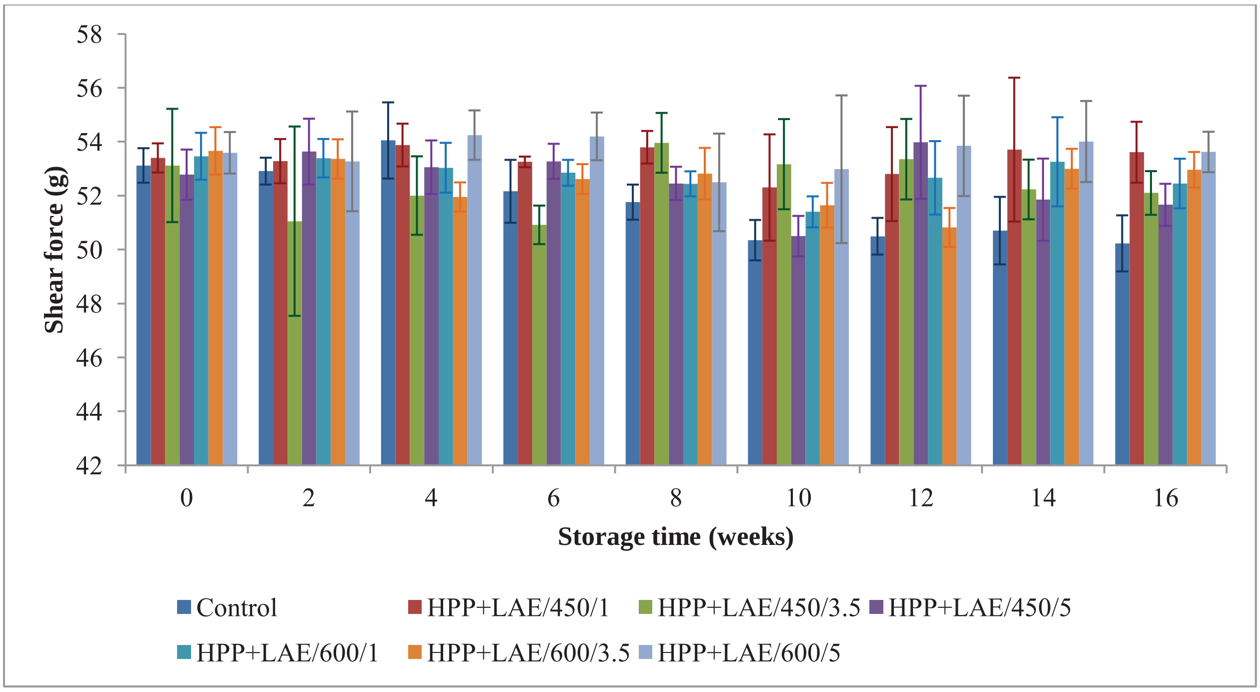

Figure 21b: Changes in shear force of HPP+LAE-treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

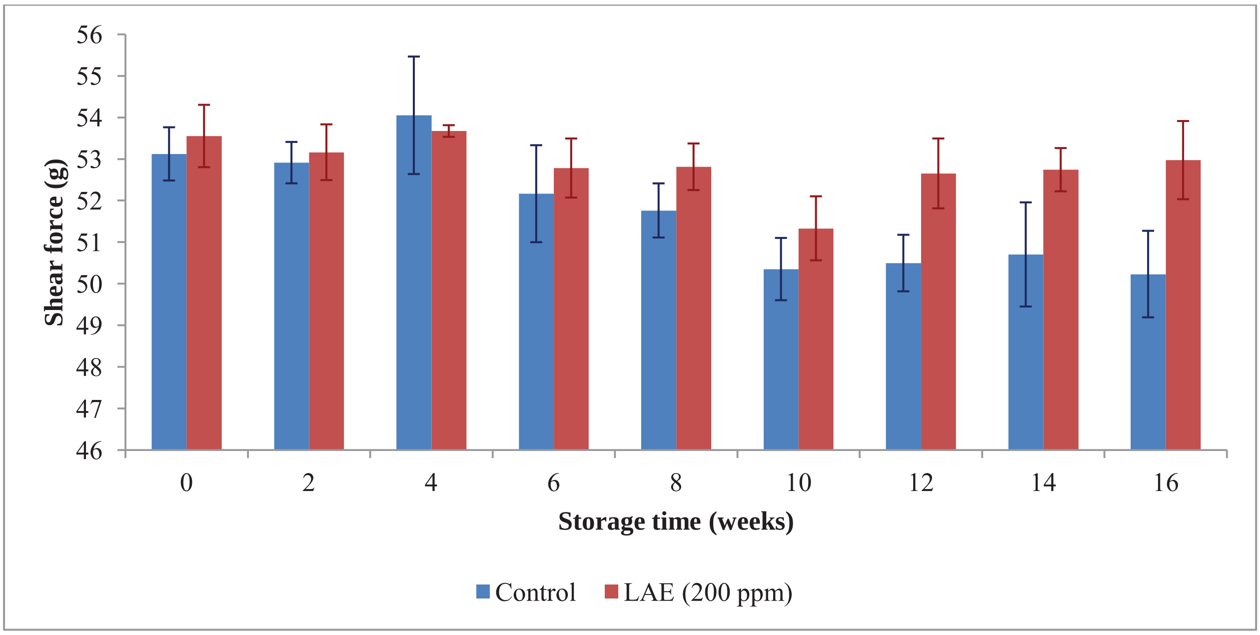

Figure 21c- Changes in shear force of LAE (200 ppm)-treated RTE sliced chicken breast roast samples during storage at 4°C for 16 weeks.

5.2 Selection of treatments for processing in Phase 2

Fourteen treatments (Table 4) were screened in the previous phase based on specific criteria. Criteria for selection of treatments for sensory evaluation was that (i) the treatments must be capable of inhibiting the growth of microorganisms (APCs, LAB and Y&M) for 16 weeks and (ii) treatments must not significantly alter the color and texture properties of the product. Based on the above selection criteria, the following treatments were selected, also keeping in mind the economic feasibility of the treatments (in terms of suitability of the selected high pressure and the pressure holding time for industrial application):

(i) HPP at 600 MPa/1min,
(ii) HPP at 600 MPa/5min,
(iii) HPP+LAE (200 ppm) at 600 MPa/1min,
(iv) LAE (200 ppm), and
(v) LAE (315 ppm).

LAE at 200 ppm was tested in the screening phase and was capable of inhibiting microorganisms for upto 11 weeks, with no significant (*P* >0.05) changes in color and texture of the cooked RTE sliced chicken samples. Therefore, a higher concentration of LAE, at 315 ppm was also tested in this phase, and its microbiological, color, texture and sensory quality were determined. An untreated (No HPP/No LAE) control was also studied in this phase.

5.2.1 Microbiological quality of the selected samples processed in Phase 2

The initial counts in the control and all the HPP treatments were below the level of detection (<1 $\log_{10}$ cfu g^{-1}). However, the counts gradually increased in the control samples that had no LAE and was not HP-treated. The counts in the control sample increased to (2.8±0.15) $\log_{10}$ cfu g^{-1} by week 2 and had reached (5.0±0.05) $\log_{10}$ cfu g^{-1} by week 10. The counts recorded in the control samples at week 16 were (5.79±0.43) $\log_{10}$ cfu g^{-1}. The viable APCs were enumerated in all the selected treatments during storage at 4°C for 16 weeks (Table 9). The APCs obtained in all the treatments are shown in Figure 22. The initial cell counts in the LAE-treated (200 and 315 ppm) samples were (1.65±0.99) $\log_{10}$ cfu g^{-1} and (1.89±0.89) $\log_{10}$ cfu g^{-1} respectively. The APCs in samples treated with HPP alone and HPP in combination with LAE were below the level of detection (<1 $\log_{10}$ cfu g^{-1}) for 16 weeks during storage at 4°C. However, the samples treated with 200 ppm LAE, showed an increase in the log counts of APCs from (2.37±1.26) $\log_{10}$ cfu g^{-1} at week 2 to (5.6±1.2) $\log_{10}$ cfu g^{-1} at week 16. Samples that were treated with 315 ppm of LAE showed an increase of >1.5 $\log_{10}$ cfu g^{-1} from the initial count by week 4 and continued to increase with a final count of (5.15±0.45) $\log_{10}$ cfu g^{-1} recorded at week 16.

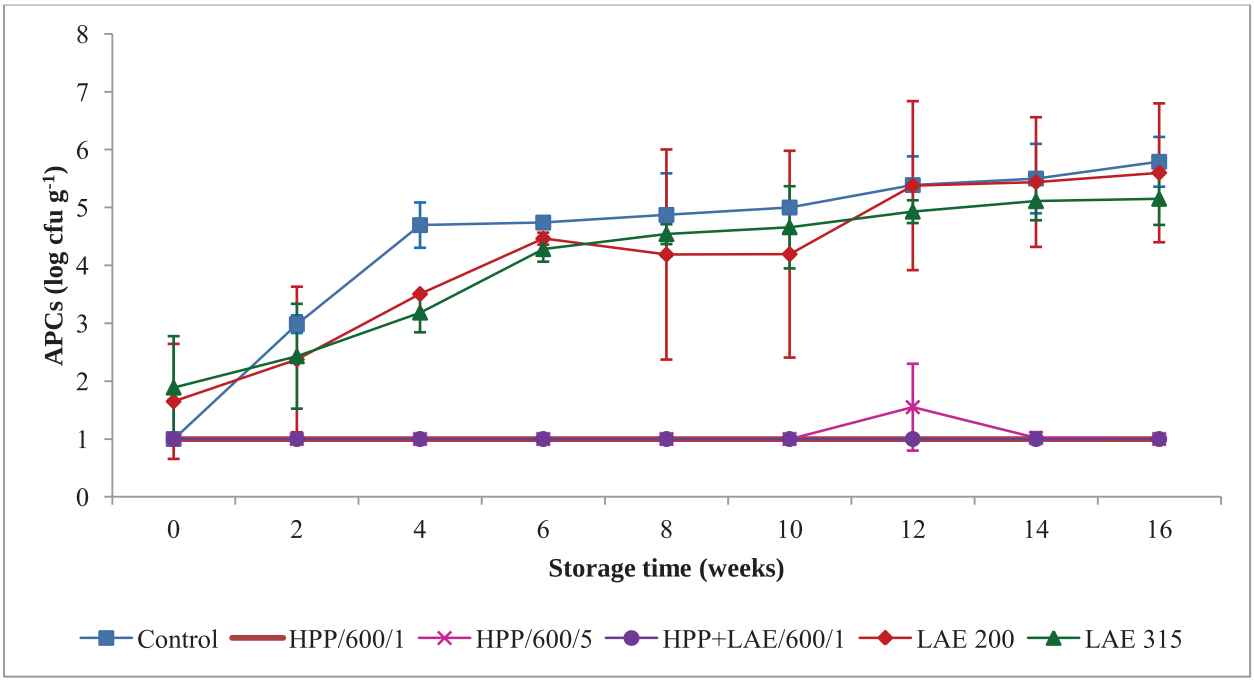

Figure 22: Changes in the APCs of the selected treatments during storage of RTE sliced chicken breast roast at 4°C.

Table 9: Change in the level of APCs*(log cfu g^{-1}) of selected treatments during storage at 4°C for 1

Storage time (weeks)	Control	HPP 600/1	HPP 600/5	HPP+LAE 600/1	LAE 200
0	$^{a}1.00 \pm 0.00^{a}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{a}1.65 \pm 0.99^{b}$
2	$^{b}2.98 \pm 0.15^{b}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{b}2.37 \pm 1.26^{b}$
4	$^{c}4.70 \pm 0.39^{c}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{c}3.51 \pm 0.02^{b}$
6	$^{c}4.74 \pm 0.08^{b}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{d}4.46 \pm 0.10^{b}$
8	$^{c}4.87 \pm 0.72^{b}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{d}4.19 \pm 1.82^{b}$
10	$^{d}5.00 \pm 0.05^{b}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{d}4.19 \pm 1.79^{b}$
12	$^{d}5.39 \pm 0.50^{b}$	1.00 ± 0.00^{a}	1.55 ± 0.75^{a}	1.00 ± 0.00^{a}	$^{abc}5.38 \pm 1.4^{b}$
14	$^{d}5.50 \pm 0.60^{b}$	1.00 ± 0.00^{a}	1.02 ± 0.10^{a}	1.00 ± 0.00^{a}	$^{abc}5.44 \pm 1.12^{b}$
16	$^{d}5.79 \pm 0.43^{b}$	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	$^{abc}5.60 \pm 1.20^{b}$

*All values are means of $\log_{10}$cfu g^{-1}± standard deviation (n= 5 samples per sampling interval). Within rows, m different letters are significantly different (P <0.05). Within columns, mean values preceded by different letters a (P <0.05).

The data shows that HPP alone and in combination with LAE (200 ppm) were most effective in increasing the microbiological shelf-life of the cooked RTE sliced chicken breast roast.

5.2.2 Instrumental color of selected treatments

L values of the selected treatments*

The lightness (Hunter's L* values) measured on the surface of the RTE sliced chicken breast roast decreased with increase in storage time in the control samples (Figure 23). The L* value initially was at 74.09 ± 1.55 in the control samples and then decreased to 66.79 ± 1.89 by week 16. However, this change was not statistically significant (*P*>0.05). The samples treated with HPP alone and in combination with LAE, showed stable L* values throughout the storage period, indicating that the lightness of the samples was stable. LAE (200 ppm)-treated samples had an initial L* value of 72.47 ± 1.17 and decreased to 70.44 ± 0.58 by the end of week 16. However, this did not have a significant (*P*>0.05) effect on the lightness of the samples. Samples treated with 315 ppm LAE recorded an initial L* value of 72.75 ± 1.55 and a 3 unit decrease (*P*>0.05) in the L value was observed at the end of the storage period.

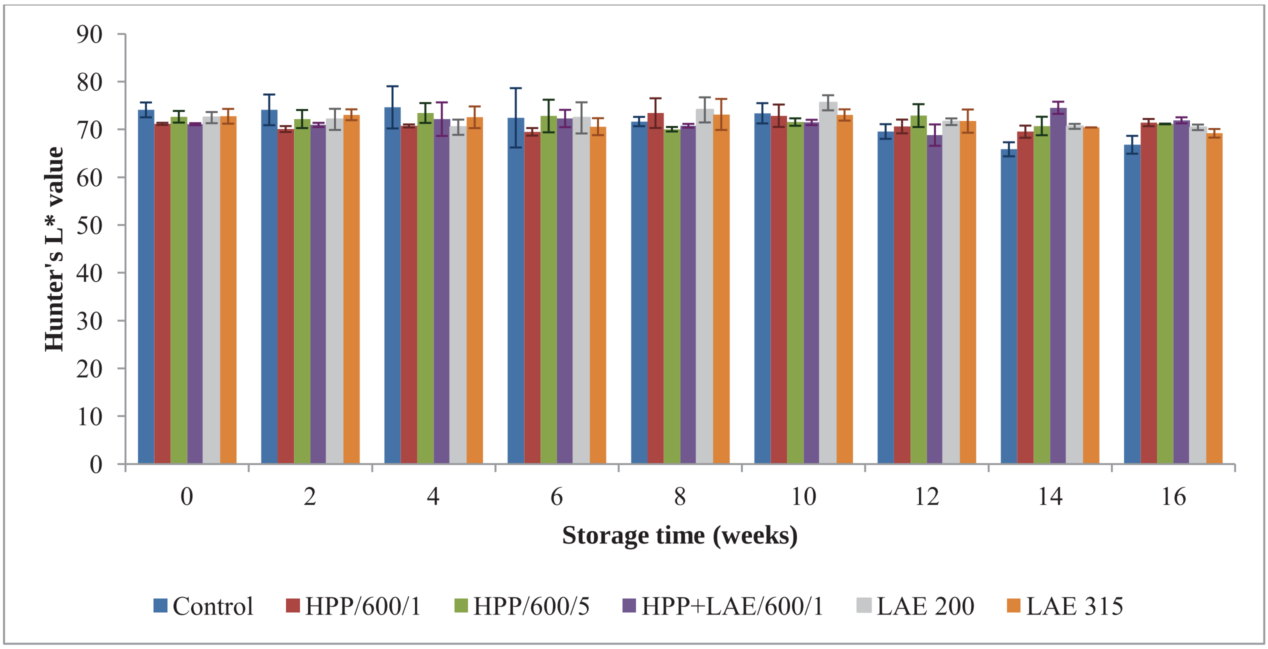

Figure 23: Changes in Hunter's L* values of the selected treatments over storage period.

a values of the selectively processed samples*

The redness (Hunter's a* value) of all samples increased during storage (Figure 24). A significant (*P*<0.05) increase from the initial a* value was observed in the control samples by week 8. The redness also significantly (*P*<0.05) increased in the HPP/600/1 treatment from an initial value of 0.52 ± 0.17 to 2.30 ± 0.17 by week 16. In samples treated with HP alone at 600 MPa for 5 min, the redness increased slightly, from an initial value of 0.96 to ± 0.06 to 1.33 ± 0.13 at the end of week 16. This change in the a* values was however not significant (*P*>0.05) in these samples. A similar trend was observed in samples treated with HP in combination with LAE, where the a* values ranged between 1.21 ± 0.11 at week 0 and 1.73 ± 0.35 at week 16. Samples treated with 200 ppm LAE had an initial a* value of 1.02 ± 0.04 and increased to 1.83 ± 0.19 at week 16. The increase in a* values of the 200 ppm LAE-treated samples was similar to the HPP+LAE/600/1 treatment. However, the maximum increase in a* values was observed in the samples treated with 315 ppm LAE. It was observed that samples treated with 315 ppm LAE had significantly (*P*<0.05) increased in redness after 6 weeks. The initial a* value of the LAE (315 ppm)-treated samples was 1.66 ± 0.39 and increased to 3.14 ± 0.49 at the end of week 16.

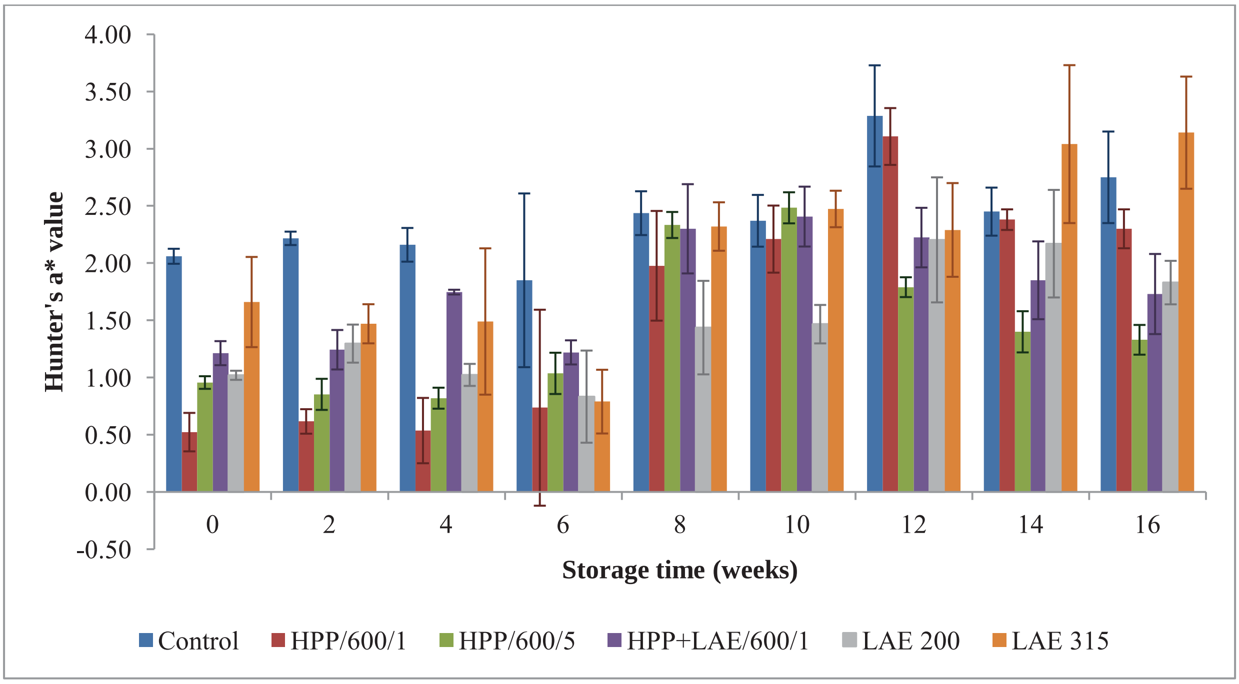

Figure 24: Changes in Hunter's a* values for the selected treatments over storage time.

b values of the selectively processed samples*

The yellowness of the samples, also expressed by the +b* values on the Hunter's lab color scale, was 6.73 ± 0.26 at week 0; an insignificant (*P*>0.05) increase to 7.14 ± 0.17 was observed by week 16 (Figure 25). The samples treated with HP at 600 MPa for 1 min showed a slight decrease in yellowness (compared to the untreated control) soon after processing. The b* value for HPP/600/1 treated samples after processing was 5.30 ± 0.17. However, the b* values increased during storage and a final b* value of 6.41 ± 0.33 was recorded at week 16. Samples treated with HPP/600/5 showed an initial b* value of 7.12 ± 0.10 and decreased (*P*>0.05) with increase in storage time. Samples treated with HP and LAE (200 ppm) in combination had stable b* values during the entire storage period. Similar trends were observed in the samples treated with 200 ppm and 315 ppm of LAE.

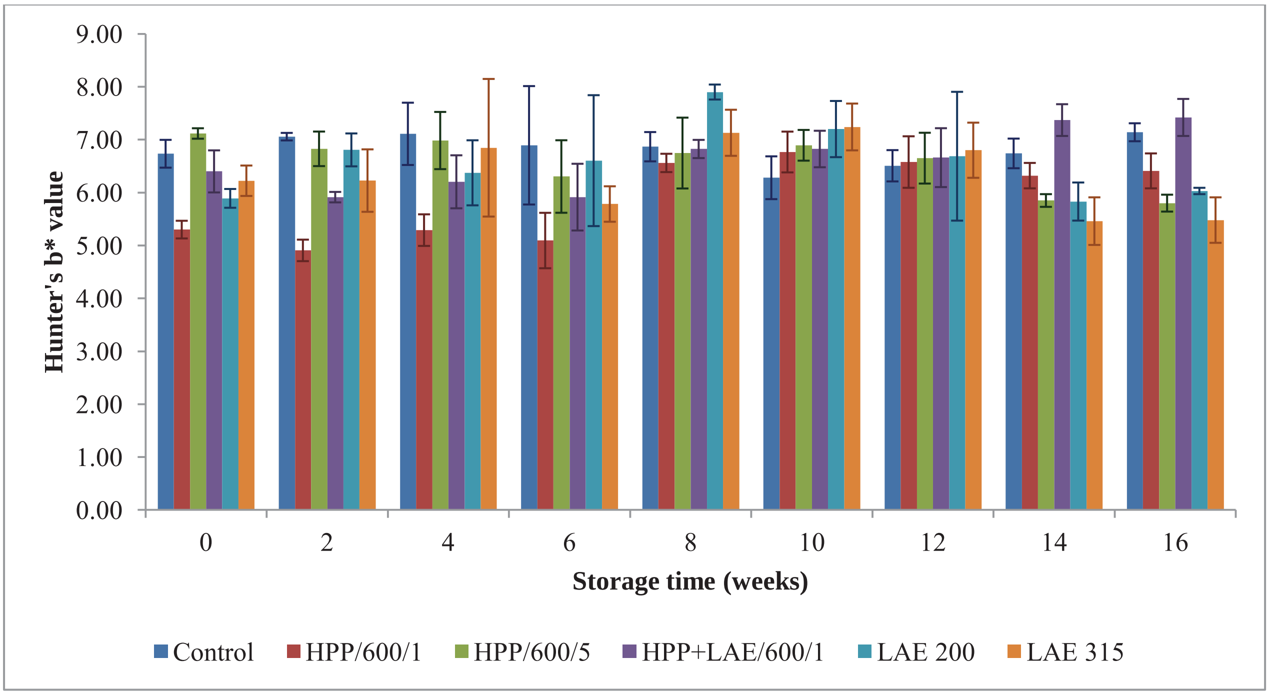

Figure 75: Changes in Hunter's b* values of the selected treatments over storage period.

5.2.3 Instrumental texture of the selected treatments

The shear force to compress the sample slices was used to study the effect of the various treatments on the firmness of the samples (Figure 26). About 2 gram force decrease was observed in the control samples at the end of the storage period. Samples treated with HPP/600/1 had a 3 gram force increase in firmness soon after processing as compared to the control; however the firmness/hardness decreased with increase in storage time to aboat the same magnitude as the control. A minimal change in the firmness of HPP/600/5-treated samples was observed throughout the storage period. Similar trend was observed in the samples treated with HPP+LAE/600/1. A 1 gram force increase was observed in the two LAE-treated samples at week 0 and a decrease in the gram force required to compress the samples increased during storage. The final shear force values of the 200 and 315 ppm LAE were 51.04 ± 1.10 g and 51.31 ± 0.92 g, respectively. Therefore, no significant (*P*>0.05) changes were observed in the texture of the samples treated with the selective treatments.

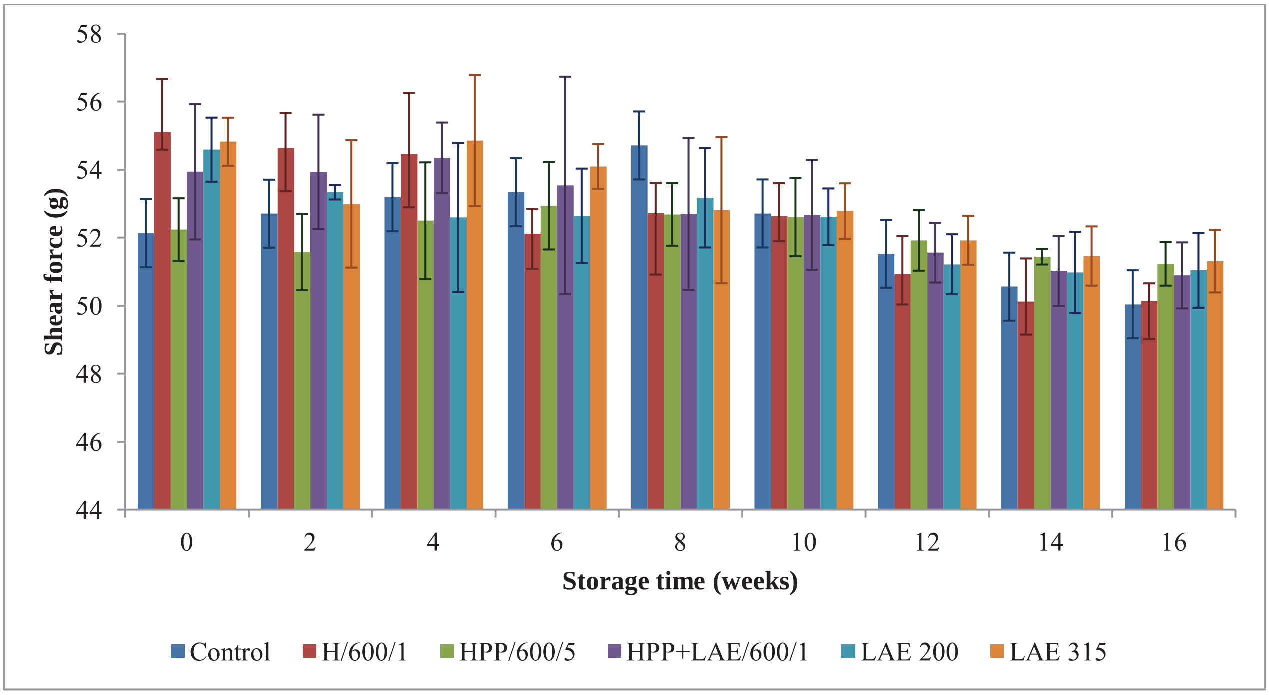

Figure 26: Changes in the shear force of selected treatments over storage time.

5.2.4 Sensory analysis of selected treatments processed in Phase 2

The sensory analysis of the selected samples was carried out fortnightly for 16 weeks. Between 20-30 panelists were recruited to participate in the sensory test from Massey University.

The samples were evaluated for color, texture, flavour, freshness and overall liking using a 9-point hedonic scale. Texture, flavour, freshness and overall liking were significantly ($P<0.05$) affected by storage time. However, color as a sensory attribute was not significantly ($P>0.05$) affected by storage time in all the treatments. Mean values for the sensory attributes are shown in Table 10.

Color was rated to be stable by the consumers in all the treatments throughout the storage period. The mean scores for texture ranged between 6.80-5.50 for samples treated with HP at 600 MPa for 1 min and 6.40-5.50 for samples treated with HP at 600 MPa for 5 min respectively. Sensory scores for samples treated with HP (at 600 MPa for 1 min) and

LAE (200 ppm) in combination ranged between 6.60 at week 2 to 5.79 by week 16 on the hedonic scale during storage. Consumer ratings for LAE at 200 ppm were rated 6.35 at week 2 to 4.00 by week 16. A significant (*P* <0.05) difference in the texture scores was observed between samples treated with 200 ppm LAE and the HPP treatments. The texture of samples treated with 200 ppm LAE was least liked by consumers after 10 weeks of storage at 4°C. Scores for samples treated with 315 ppm LAE was rated 6.40 at week 2 to 4.71 by week 16 and a significant (*P* <0.05) difference was observed between all the treatments. The most liked samples in terms of texture were those that were treated with HP at 600 MPa for 1 min and 5 min respectively and samples treated with HP + LAE (200 ppm) at 600 MPa for 1 min. No significant (*P* >0.05) differences were observed in the texture scores between these treatments. Samples treated with 200 ppm LAE had the least liked texture scores.

Flavour was not significantly (*P*>0.05) different between samples treated with HP at 600 MPa for 1 min and 5 min respectively and samples treated with HP + LAE (200 ppm) at 600 MPa for 1 min, during the initial period (up to 6 weeks) of storage. However, after 6 weeks of storage, a significant (*P*<0.05) difference was observed in the flavour scores of samples treated with HP at 600 MPa for 1 min and 5 min. Samples treated with HP at 600 MPa for 1 min had a higher flavour score when compared to the samples treated with HP at 600 MPa for 5 min. However, samples treated with HP at 600 MPa for 1 min and samples treated with HP + LAE (200 ppm) were correlated in their flavour scores and were the most liked samples for their flavour profile. The least liked samples with respect to flavour were those that were treated with 315 ppm LAE. Consumers rated samples treated with 315 ppm LAE with flavour scores like 4.43 by week 8 and as low as 2.90 on the 9 point hedonic scale by week 14. Samples treated with 200 ppm LAE were also disliked slightly after week 10 and were disliked very much by week 14, as rated by the consumer panel.

No significant (*P*>0.05) difference for the attribute ‘freshness’ was observed between the HPP treatments (HPP at 600 MPa for 1 min and 5 min) and the HPP + LAE (200 ppm) treatment. Therefore, suggesting that the HPP treatments and HPP+LAE (200 ppm) treatment had consistent scores for freshness. Consumers rated the LAE (200 ppm and 315 ppm) treatments as the least fresh amongst all treatments by week 10.

Overall liking for all the treatments was not significantly (*P*>0.05) different between all the treatments for the first 6 weeks. However, after 6 weeks a significant (*P*<0.05) decrease in the overall liking scores was observed for samples treated with 200 ppm and 315 ppm LAE as compared to the other treatments. Overall liking scores for the samples treated with HP at 600 MPa for 1 min and 5 min and HPP+LAE (200 ppm) at 600 MPa for 1 min were similar and these were the most liked samples by the consumers throughout the storage period.

Table 10: Comparison of the mean consumer sensory scores* of the selected treatments

Storage time (in weeks)	Treatment	Overall liking	Color	Texture	Fla
2	HPP/600/1	6.48 (1.44)^a	5.91 (1.73)^a	6.43 (1.56)^a	6.70 (
	HPP/600/5	6.17 (1.66)^a	5.61 (1.77) ^{a}	6.30 (1.66)^a	6.17 (
	HPP+LAE/600/1	6.04 (1.72)^a	5.70 (1.79) ^{a}	6.35 (1.80)^a	6.13 (
	LAE 200	6.30 (1.69)^a	6.13 (1.60) ^{a}	6.35 (1.50)^a	6.26 (
	LAE 315	5.83 (1.75)^a	5.48 (2.13) ^{a}	6.09 (1.81)^a	6.09 (
4	HPP/600/1	6.11 (1.36)^a	5.78 (1.39) ^{a}	6.22 (1.20)^a	7.00 (
	HPP/600/5	6.11 (1.36)^a	6.00 (1.80) ^{a}	6.22 (1.72)^a	5.67 (
	HPP+LAE/600/1	6.44 (1.33)^a	5.56 (1.74) ^{a}	6.22 (1.86)^a	6.22 (
	LAE 200	6.22 (1.20)^a	5.67 (1.66) ^{a}	6.22 (1.56)^a	6.56 (
	LAE 315	6.22 (1.79)^a	6.33 (1.50) ^{a}	6.00 (1.87)^a	5.78 (
6	HPP/600/1	6.04 (1.72)^a	5.74 (1.66) ^{a}	5.78 (1.81)^a	6.13 (
	HPP/600/5	6.00 (1.51)^a	5.87 (1.60) ^{a}	6.00 (2.00)^a	6.04 (
	HPP+LAE/600/1	5.52 (1.73)^a	5.74 (1.84) ^{a}	5.43 (1.70)^a	5.43 (
	LAE 200	5.22 (2.00)^a	5.78 (1.65) ^{a}	5.96 (1.46)^a	5.26 (
	LAE 315	5.91 (1.47)^a	5.52 (1.27) ^{a}	5.65 (1.53)^a	6.09 (
8	HPP/600/1	5.93 ± 1.69^a	5.43 (1.91) ^{a}	5.93 (1.64)^a	6.29 (
	HPP/600/5	5.21 ± 1.72^a	4.07 (2.13) ^{a}	5.71 (1.68)^a	5.50 (
	HPP+LAE/600/1	6.07 ±1.59^a	6.00 (1.71) ^{a}	5.79 (2.01)^a	6.21 (
	LAE 200	5.43 ± 1.40^a	6.14 (1.41) ^{a}	5.79 (1.19)^a	5.43 (
	LAE 315	4.50 ± 1.74^b	4.86 (2.03) ^{a}	4.71 (1.94)^b	4.43 (

Table 10$_{\text{contd.}}$: Comparison of the mean consumer sensory scores* of the selected treatments

Storage time (in weeks)	Treatments	Overall liking	Color	Texture	Fla
10	HPP/600/1	5.60 (1.34)^{a}	5.60 (1.52) ^{a}	6.00 (1.58)^{a}	6.40 (
	HPP/600/5	6.20 (1.79)^{a}	5.40 (1.95) ^{a}	6.20 (1.92)^{a}	5.80 (
	HPP+LAE/600/1	6.40 (1.14)^{a}	6.00 (1.58) ^{a}	6.60 (1.52)^{a}	6.60 (
	LAE 200	4.60 (2.70)^{b}	4.80 (1.64) ^{a}	4.00 (1.87)^{b}	4.40 (
	LAE 315	4.60 (2.70)^{b}	6.60 (2.70) ^{a}	5.00 (1.87)^{b}	4.20 (
12	HPP/600/1	6.60 (0.84)^{a}	6.00 (0.94) ^{a}	6.60 (0.84)^{a}	7.30 (
	HPP/600/5	6.30 (1.06)^{a}	6.10 (1.10) ^{a}	6.40 (0.84)^{a}	5.90 (
	HPP+LAE/600/1	5.90 (0.74)^{a}	5.70 (1.06) ^{a}	6.00 (0.67)^{a}	6.00 (
	LAE 200	5.40 (1.43)^{a}	5.80 (1.14) ^{a}	4.10 (1.37)^{c}	4.90 (
	LAE 315	4.30 (0.95)^{b}	5.60 (1.26) ^{a}	5.40 (1.17)^{c}	3.90 (
14	HPP/600/1	6.50 (0.53)^{a}	5.90 (0.99) ^{a}	6.80 (0.92)^{a}	7. 00
	HPP/600/5	6.30 (0.67)^{a}	5.80 (1.03) ^{a}	6.30 (0.95)^{a}	5.70 (
	HPP+LAE/600/1	5.60 (0.52)^{b}	5.30 (0.82) ^{a}	5.50 (0.53)^{b}	5.50 (
	LAE 200	4.80 (0.92)^{c}	5.40 (1.17) ^{a}	4.70 (1.25)^{c}	3.90 (
	LAE 315	4.00 (1.05)^{d}	6.00 (1.25) ^{a}	5.10 (0.74)^{b}	2.90 (
16	HPP/600/1	6.50 (0.51)^{a}	5.90 (0.97) ^{a}	6.75 (0.85)^{a}	7.00 (
	HPP/600/5	6.45 (0.60)^{a}	5.65 (0.75) ^{a}	6.20 (0.77)^{a}	5.75 (
	HPP+LAE/600/1	5.70 (0.47)^{a}	5.45 (0.69) ^{a}	5.60 (0.50)^{b}	5.65 (
	LAE 200	3.90 (0.79)^{b}	5.30 (1.03) ^{a}	4.80 (1.11)^{c}	3.20 (
	LAE 315	3.65 (0.75)^{b}	5.60 (0.88) ^{a}	4.60 (0.50)^{c}	3.00 (

* All values are means of consumer sensory scores followed by standard deviation in ( ). Within columns, all mean val
letters are significantly (*P*<0.05) different.

5.2.5 Sensory shelf life estimation using survival analysis

The shelf life of the food products is not exclusively related to deterioration, but to a complex phenomenon dependent on the interaction of the consumer with the actual food (Hough et al., 2003). In this context, the key concept of survival analysis methodology to focus the shelf-life hazard on the consumer rejecting the product rather than on the product deterioration (Hough et al., 2003). The Weibull distribution was chosen to model rejection times for the data. Figure 27 shows the trend of the overall acceptability score for the selected treatments with increase in storage time. As expected, a continuous decrease of the acceptability scores for the samples treated with 200 ppm and 315 LAE respectively and samples treated with HPP+LAE (200 ppm) was observed with increase in storage time. However, the overall acceptability of the samples treated with HPP at 600 MPa for 1 min and 5 min remained stable throughout the storage period.

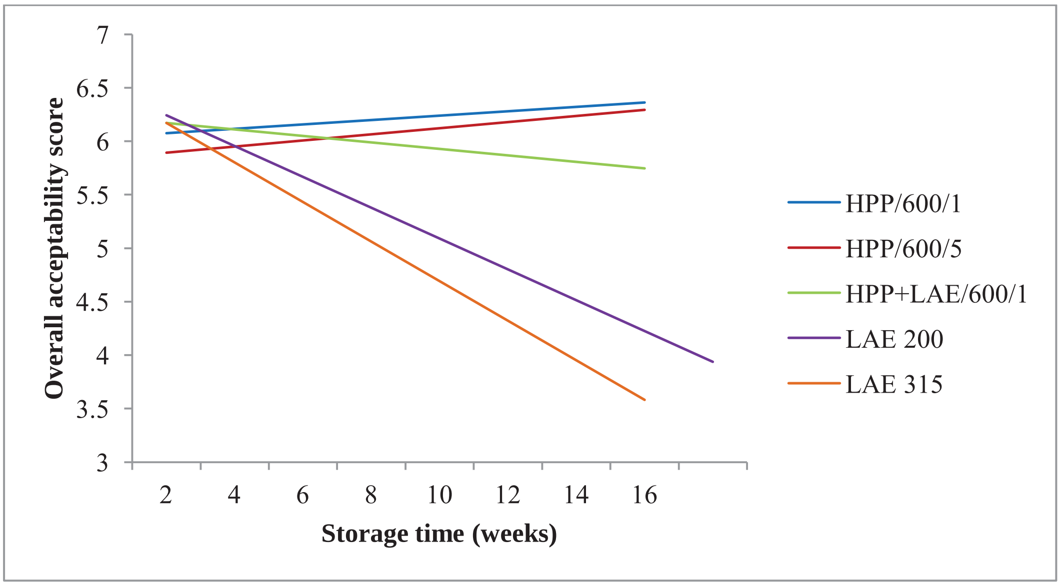

Figure 27: Overall acceptability score of selected treatments for RTE sliced chicken breast roast during storage at 4°C for 16 weeks.

Weibull models were fitted separately for the data obtained for the different treatments. The parameters of the Weibull model for the selected treatments are shown in Table 11.

These parameters were used to plot consumer rejection probability versus storage time for each treatment, as shown in Figure 28. These curves were used to estimate the shelf-life (*x*-axis), by entering with a 25 and 50% consumer rejection (*y*-axis). The estimated shelf lives of the samples are shown in Table 12.

Table 11: μ and σ values of the Weibull distribution.

Treatment	μ	σ
HPP/600/1	0.92	49.91
HPP/600/5	1.17	28.14
HPP+LAE/600/1	1.38	20.10
LAE 200	2.33	11.91
LAE 315	2.01	11.10

Note: μ=shape parameter; σ= scale parameter

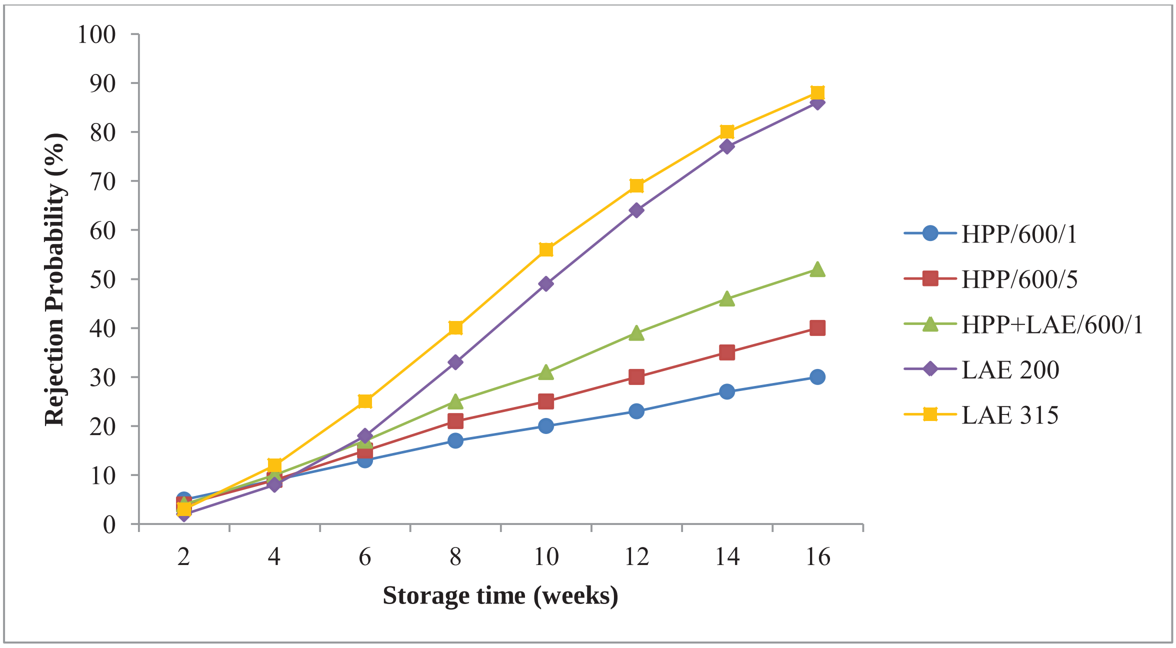

Figure 28: Consumer rejection probability of the samples as a function of storage time.

Table 12: Estimated shelf lives of RTE sliced chicken breast roast based on selected treatments.

Treatment	Shelf-life time (weeks)	
	For 25% rejection	For 50% rejection
HPP/600/1	13.8	>16
HPP/600/5	10.5	>16
HPP+LAE/600/1	8.3	14.3
LAE 200	7.2	10.1
LAE 315	6	9.2

For a 25% consumer rejection, estimated shelf lives were 13.8 weeks for samples treated with HP at 600 MPa for 1 min, 10.5 weeks for samples treated with HP at 600 MPa for 5 min, 8.3 weeks for samples treated with HPP+LAE at 600 MPa for 1 min, 7.2 for samples treated with LAE at 200 ppm and 6 weeks for samples treated with LAE at 315 ppm. Therefore, samples treated with HP alone at 600 MPa for 1 min and 5 min had a significantly higher sensory shelf-life as compared to the other treatments.

For 50% consumer rejection, estimated shelf lives of the samples were >16 weeks for the treatments HPP/600/1 and HPP/600/5, 14.3 for treatment HPP+LAE/600/1, 10.1 for LAE 200 and 9.2 for LAE 315. The shelf lives of all the HPP treatments were significantly higher than the LAE treatments. No significant difference was found between the sensory shelf lives of the two LAE treatments.

6.0 DISCUSSION

6.1 Microbiological quality

6.1.1 Effect of HPP (450 MPa and 600 MPa) on the microbiological quality of RTE sliced chicken breast roast

The efficacy of high-pressure treatments for the inactivation of vegetative bacteria in foods has been reported previously (Cheftel, 1995a; Farkas & Hoover, 2000; Smelt, 1998; Yuste et al., 2001). However, there are limited studies on the evaluation of microbial safety and quality of pressurized products during chilled storage (Capri et al., 1999; Lopez-Caballero, Carballo, & Jimenez-Colmenero, 1999; Yuste, Pla, Capellas, Ponce, & Mor-Mur, 2000). Often, food composition can have a protective effect during pressurization, and it is therefore, important to evaluate microbial resistance to pressure in foods rather than in traditional buffer solutions (Yuste et al., 2001).

Marketing of convenience foods is increasing due to consumer demand. Slicing and packaging operations take place after cooking, and cross-contamination at these points is critical regarding the shelf-life and safety of the products. In this study, the initial microbial cell numbers of the HPP-treated (450 MPa and 600 MPa) cooked and vacuum-packaged chicken breast roast were at or below the level of detection for APCs (1 $\log_{10}$ cfu g^{-1}) and LAB and Y&M (2.3 $\log_{10}$ cfu g^{-1}), suggesting that the good manufacturing practices observed in the production area were satisfactory. Results of the current study are similar to those reported by Patterson et al. (2010), where the initial microbial counts in the untreated and pressure-treated cooked chicken were reported to be <1 $\log_{10}$ cfu g^{-1}. In many cases, these products later became spoiled during storage (Bjorkroth & Korkeala, 1996; Chenoll, Macian, Elizaquivel, & Aznar, 2007; Hamasaki, Ayaki, Fuchu, Sugitama, & Morita, 2003). However, in this study, none of the pressure-treated poultry meat samples showed any sign of obvious spoilage, irrespective of the pressure level or hold time used. Patterson et al. (2010) also reported similar results, although in some cases, microbial numbers were at 7-8 $\log_{10}$ cfu g^{-1} after 35 days of storage at 4°C.

In another study by Garriga et al. (2004), samples pressurized at 600 MPa for 6 min showed a significant delay in the growth of spoilage associated microorganisms compared to untreated samples, contributing to the maintenance of the sensory freshness of the products for at least 60 days after treatment. Even after storage of samples for 90 days at 4°C, the cell counts in the treated samples did not exceed $>10^7$ cfu g^{-1}. The use of HPP process retained product quality in terms of off-odors, ropiness, and color changes. Capri et al. (1999) reported an extended shelf-life of sliced cooked ham treated at 600 MPa for 5 min up to 75 days stored at 4°C. The results achieved by Lopez-Caballero et al. (1999) with the same type of product but treated at lower pressures (200 MPa and 400 MPa) did not achieve the same degree of inactivation and the maximum shelf-life obtained was up to 21 days when stored at at 3°C, for sliced and pressurized (400 MPa for 20 min) cooked ham. The results of these previous studies agree with the generally accepted fact that the degree of inactivation is directly related to the level of pressure applied (Garriga et al., 2004).

Gola et al. (2000) demonstrated that use of pressures between 400 to 700 MPa caused significant reductions of eight mixed *E.coli* strains. Malicki, Sysak, and Bruzewicz (2005) showed that pressures between 100 and 400 MPa efficiently reduced strains of *Salmonella.* Styles, Hoover, and Farkas (1991) reported at least 7-log reductions in *L. monocytogenes* at a pressure of approximately 340 MPa and Patterson, Quinn, Simpson and Gilmour (1995) reported a similar reduction of *L. monocytogenes* at 400 MPa. Pressures of 450 MPa and 600 MPa were also reported to be very effective in increasing shelf-life of chicken breast fillet up to 14 days of storage at 4°C (Jo et al., 2011). Pressure treatments between 400 and 700 MPa were reported to increase shelf-life of minced meat under refrigeration conditions (Gola et al., 2000). The results obtained in this study clearly demonstrated that increased hydrostatic pressure (600 MPa) was able to inactivate microbial populations and extend the shelf-life of RTE sliced chicken breast roast to 16 weeks.

The cell populations of LAB and Y&M in the HPP-treated samples were below the detection limit (<1 $\log_{10}$ cfu g^{-1}) throughout the storage period in this study. The results were however in contrast with studies conducted by Garriga et al. (2004), where the LAB counts in samples pressurized at 600 MPa for 6 min were 2.65 ± 1.14 $\log_{10}$ cfu g^{-1} at 60 days of storage and increased to 7.62 ± 0.97 $\log_{10}$ cfu g^{-1} by the end of storage period (120 days).

High pressure processing conditions used in this study were effective against the growth of yeasts and Enterobacteriaceae, which can produce off-flavours and gas. Pressures between 300-600 MPa are known to inactivate most moulds (Tokusoglu et al., 2011). For microorganisms, the primary site of pressure damage is the cell membrane (Balasubramaniam, 2010). Pressures of 200-400 atmospheres can disrupt the stressed cell wall, and this may be a primary factor for yeasts (Hoover et al., 1989). Eukaryotic microorganisms are generally more sensitive to pressure than prokaryotic microorganisms. In general, Gram-negative bacteria are more sensitive to pressure than Gram-positive bacteria (Carlez, Rosec, Richard, & Cheftel, 1993; Shigehisa, Ohmori, Saito, Taji, & Hayashi, 1991). In fact, for all the sample treatments in this study, no recovery of yeasts or moulds was observed in HPP samples during the entire storage period.

In this study samples processed with HPP at 450 and 600 MPa for 3.5 and 5 min showed an increase in APCs during storage. This could be attributed to the germination of spores activated by the pressures used. Germination of bacterial spores at relatively low pressures (30-50 MPa) is not uncommon (Gao et al., 2006). However, the germinated spores can be subsequently killed by relatively mild heat treatments or high pressure treatments. Process temperatures in the range of 80-100°C in conjunction with a pressure of about 600 MPa have been used to inactivate bacteria such as *Bacillus subtilis* propagated from inherent spores (Gao et al., 2006).

It has also been reported that pressure-assisted thermal processing (PATP) can inactivate spores, whereby the food product undergoes pre-heating and the adiabatic heat of compression is used to raise the temperature further (Balasubramaniam, 2010). When the pressure is released, the temperature rapidly drops, which minimizes the adverse effects of heating on the food product. Spores of *Clostridium* species have been associated with raw and cooked poultry meats which are not handled appropriately (Skariyachan, Mahajanakatti, Biradar, Sharma, & Abhilash, 2010). High pressure alone does not necessarily kill significant populations of spores, but it does appear to induce germination. Spores can be inactivated by combination of both pressure and heat. According to reports by Doona & Feeherry (2007) and Balasubramaniam (2010), spores of *Bacillus amyloliquefaciens* and selected surrogates of pathogenic *Clostridium* and *Bacillus* species were inactivated by application of high pressure and temperature combined. However, the mode of action of HPP on bacterial spores is still unclear.

6.1.2 Effect of HPP+LAE on the microbiological quality of RTE sliced chicken breast roast

Different studies have shown that HPP alone may not be sufficient for food preservation under all conditions; thus it may be necessary to use the hurdle technique by combining HPP with one or more other factors that can act synergistically (Hugas et al., 2002). Several factors have proved successful in this regard, such as low pH (Alpas et al., 2000), mild temperature processing with antimicrobial peptides (Garcia-Graells, Masschalck, & Michiels, 1999), lysozyme and the lactoperoxidase system in milk (Garcia-Graells, Valckx, & Michiels, 2000) the use of antimicrobial peptides *in vitro* (Kalchayanand et al., 1998) as well as in meat products (Garriga et al., *In Press*).

Masschalck, Van Houdt, and Michiels (2001) described two types of sensitization of bacteria to antimicrobial compounds by high pressure in buffer systems. One type is transient sensitization, whereby bacteria exhibit sensitivity to the antimicrobial only during the time they are being held under pressure, with resistance being restored to the level of unpressurized cells immediately upon relief. This is the case with lysozyme and nisin (Soontjes, Michiels, Wuytack, & Hauben, 1996). The other type is persistent sensitization, whereby bacteria remain sensitive for at least several hours after pressure treatment, like the sensitization at low pH and for the lactoperoxidase system (Garcia-Graells, Valckx et al., 2000). The latter form of sensitization involves small diffusible antimicrobial molecules that can penetrate the Gram-negative outer membrane. With larger antimicrobial molecules such as bacteriocins, no persistent sensitization was observed, probably because they failed to penetrate the outer membrane of Gram-negative bacteria after pressure treatment (Hugas et al., 2002).

In this study, HPP was applied to LAE-treated samples at 200 ppm concentration. The maximum inhibition of APCs, LAB and Y&M was observed in the samples treated with HPP+LAE (200 ppm) at 600 MPa for 1 min, 3.5 min and 5 min. No significant ($P>0.05$) difference was observed in all three samples. In an earlier study by Seemeen & Mutukumira (2010), RTE sliced chicken breast roast samples treated with LAE (200 ppm) alone were stable for 8 week (Seemeen & Mutukumira, 2010).

A combination treatment of HPP and LAE has not been reported by other researchers. However, HPP combined with nisin has been widely reported (Gao & Ju, 2008; Houdt et al., 2001; Knorr, Lee, & Heinz, 2003; Lee & Kaletunç, 2011; Yuste, Pla, Capellas, & Mor-Mur, 2011). A mechanism of pressure-promoted uptake of antimicrobials in bacteria has been used to explain the sensitization of test bacteria treated with nisin under pressure (Houdt et al., 2001). In a study conducted by Masschalck et al. (2001), lactoferrin (500 µg/ml), lactoferricin (20 µg/ml) and nisin (100 IU/ml) displayed bactericidal activity against strains of *E.coli, Salmonella* spp, *Shigella* spp., *Pseudomonas fluorescens* and *Staphylococcus aureus* when pressure-treated in the range of 155-400 MPa. The bactericidal efficacy and spectrum of nisin were enhanced under pressure (Masschalck, Van Houdt, & Michiels, 2001).

In general, the higher the pressure and the longer the time of treatment, the greater the microbial load reduction (Table 6). Buffered suspensions of vegetative bacterial cells or spores have been subjected to high pressure, nisin, and in some cases, other processes such as mild heat and low pH, and a synergy between combinations of HPP treatments was observed (Garcia-Graells, Masschalck, Michiels, & Haver, 2000; Hauben, Wuytack, Soontjens, & Michiels, 1996; Roberts & Hoover, 1996b; Stewart, Dunne, Sikes, & Hoover, 2000). Pressure has been shown to increase the effectiveness of nisin and other antimicrobial agents by sub-lethally damaging bacterial cells (Kalchayanand et al., 1994).

6.1.3 Effect of LAE on the microbiological quality of RTE sliced chicken breast roast

In this study, samples treated with LAE at 200 ppm and 315 ppm were able to extend the shelf-life of the RTE sliced chicken breast roast to 8 weeks during storage at 4°C. The application of the preservative within the food industry may be limited for a number of reasons: (1) its potency as an antimicrobial may be affected if it interacts with anionic components within the food matrix, (2) it may bind to anionic biopolymers in the mouth, leading to perceived bitterness, and (3) it tends to precipitate from solution at pH > 4.5 and at high ionic strength (in beverages) (Asker, Weiss, & McClements, 2009). The antimicrobial properties of LAE, are likely to depend on its interactions with other molecules in the systems in which it is used. In particular, this cationic surfactant may interact with anionic biopolymers through electrostatic interactions, which may either enhance or reduce its activity (Asker et al., 2009).

LAE is reported to be effective against microorganisms and pathogens, when included with lactates and diacetates in the formulation (Martin, Griffis, Vaughn, Bryan, Friedly, Marcy, Ricke, Crandall, Lary et al., 2009). Luchansky et al. (2005) observed that after the initial log reduction by LAE, the cell counts of *Listeria monocytogenes* increased by 2-5 log cfu/ham in samples without potassium lactate or sodium diacetate, when stored upto 60 days. LAE is a cationic surfactant, and it is suggested that it neutralizes molecules by competitively binding anionic cell surfaces, leading to alterations in the cell permeability of bacterial cells and hence inactivating or inhibiting them. The effect of LAE is also known to take place in the initial contact time and then remain stable (Rodriguez, Seguer, Rocabayera, & Manresa, 2004). Therefore, the time and temperature of exposure are perhaps important factors contributing to the cation reaching negatively charged cell surfaces.

LAE at 200 ppm inhibited the growth of microorganisms in cooked and vacuum-packaged RTE chicken breast roast stored for 49 days at 4°C (Seemeen & Mutukumira, 2010). LAE causes alterations in the cells that lead to disturbance in the membrane potential and cell structure, which can result in the loss of cell viability but not cell disruption or lysis (Kawamura & Whitehouse, 2008). This may further sensitize the bacterial cells to the applied high pressure.

The combined stresses of acid or alkaline pH, NaCl, and exposure to monolaurin or lauric acid were studied in meat and cheese isolates of *L. monocytogenes* (Vasseur, Rigaud, Herbaud, & Labadie, 2001). Lethality of cells that were exposed to 75 and 100 ppm of monolaurin or lauric acid was similar to that caused by exposure to 10% NaCl or pH 5.4 or 10.5. However, the combination of monolaurin, NaCl and alkaline pH was more than a hundred times more lethal to *L. monocytogenes* than NaCl or alkaline pH alone. The potential for NaCl or alkalinity to enhance efficacy of LAE was not investigated. Synergistic effects of fatty acid molecules and monolaurin with phenolic compounds and organic acids have also been reported (Blaszyk & Holley, 1998). Synergistic combinations of LAE with sodium lactate and sodium diacetate have also been reported (Martin, Griffis, Vaughn, Bryan, Friedly, Marcy, Ricke, Crandall, Lary et al., 2009; Porto-Fett et al., 2010; Sallen, Desvarenne, Quinn, Rajoharison, & Mabilat, 1996). However, combination studies of LAE with applied high pressure have been reported for the first time in this study.

6.2 Instrumental analysis of color

Color is the most important perceived attribute in meat products in terms of consumer acceptance (Jo et al., 2011). Lightness as indicated by the L* values was stable in all the HP-treated and HP+LAE- treated samples. However, this observation was not in agreement with the trends observed in restructured pork (Choi, Hong, Ko, & Min, 2008) and chicken breast meat (Betti et al., 2011), where increasing trends in the L* values was observed with an increase in applied pressure. Meat discoloration during high pressure processing can be due to (1) a “whitening” effect caused by myoglobin denaturation to heme displacement or release and (2) oxidation of the ferrous myoglobin to ferric myoglobin above 400 MPa (Carlez, Veciana-Nogues, & Cheftel, 1995). Previous studies have reported that the application of high pressure treatment affects the color of beef muscle (Serra et al., 2007; Shigehisa et al., 1991), fish minced muscle of albacore tuna

(Ramirez-Suarez & Morrissey, 2006), cod and mackerel (Ohshima et al., 1993), bluefish and sheephead (Ashie & Simpson, 1996a) as well as meat products such as sausages causing their "whitening" effect (Crehana, Troya, & Buckley, 2000). In a study by Carlez et al. (1995), the discoloration of meat was observed even at lower pressures between 200 MPa and 350 MPa.

The paleness of meat treated by HPP becomes brighter as measured by the L* value and this does not only account for a loss of the pigment, but also to protein coagulation that changes sample surface properties as well as the ratio of absorbed versus reflected light resulting in the "whitening" effect (Carlez et al., 1995; Mor-Mor & Yuste, 2003). It has been reported that pressure from 200 MPa inhibits calpastatin and further increase in pressure to over 400 MPa, can cause degradation of calpains (Hugas et al., 2002). In a study by Jo et al. (2011), the L* values of HPP-treated chicken breast fillet showed an increase by 32, 48 and 50% for pressures of 300 MPa, 450 MPa and 600 MPa, respectively. The L* values increased at pressure of 300 MPa and reached a maximum at pressure 450 MPa. There was no significant difference in L* value when pressure was increased to 600 MPa. A similar effect was observed in pork homogenates. The L* value started increasing with the pressure between 100 and 200 MPa and reached maximum between 300 and 400 MPa with no further increase at 600 MPa (Jo et al., 2011).

In most of the temperature-pressure combinations, L* values of NaCl-treated chicken breast samples were lower than that of the control, which could be attributed to the ability of water retention by NaCl samples (Betti et al., 2011). Increase in NaCl concentration results in an increase in water retention which can lead to darker color of meat (Baublits, Pohlman, Brown, Yancey, & Johnson, 2006). Higher L* values for meat are normally associated with poorer water holding capacity. However, in this study the L* values did not increase after pressure processing of samples (Figure 18 a & b). The discrepancy in the L* values could be accounted to the differences between the experiments, such as treatments and types of meat used. For semi-cooked or fully-cooked products, change in the L*values is not observed, since their proteins have already been denatured (Sorenson et al., 2011).

L* values of samples treated with LAE (200 and 315 ppm) showed a slight increase with storage time, which was not statistically significant (*P*>0.05) (Figure 18c). However, there have been no reported cases of the effect of LAE on the color of meat products.

The lower a* values of pressure-treated and pressure + LAE-treated RTE sliced chicken breast roast in this study (Figure 19a & b) were in agreement with the trends observed in chicken minced breast meat (Mariutti, Orlien, Bragagnolo, & Skibsted, 2008) as well as in beef muscle (Yamamoto, Lee, Lee, Kim, & Kim, 2007) where a* values decreased at higher pressure. The minced chicken breast in the experiment conducted by Mariutti et al. (2008) contained sage and garlic which are natural antioxidants. The presence of sage and garlic may have increased the antioxidant activity in the minced chicken breast. The antioxidant activity could have a protective effect, especially after the changes induced by the high pressure treatment which triggers ferrous oxidation of myoglobin which in turn, affects the color.

In another study, increase in pressure decreased the a* values of chicken breast meat (Betti et al., 2011). Cava, Gonzalez, Carrasco, and Ramirez (2009) showed that the discoloration through pressure processing appears as a result of oxidation of ferrous myoglobin to ferric myoglobin, with a decrease in the a* values. Also, the proportion of metmyoglobin increased at the expense of oxymyoglobin. The effects of high pressure treatment on myoglobin mainly depend on processing temperature (Cava et al., 2009). The decrease in a* values in pressure-treated mackerel and cod have been reported (Oshima, Nakagawa, & Koizumi, 1992).

A reduction in the a* values of LAE (200 and 315 ppm) treated chicken was observed in this study (Figure 19c). However, the results were not significant and this observation has not been reported before.

Contrasting reports about the effect of high pressure on the b* values in different experiments show that this color parameter may not always be affected by the high pressure treatment. In minced chicken breast meat, no effect of pressure on b* values was observed irrespective of the presence of spices, applied pressure or storage time (Mariutti et al., 2008). No changes in any of the color parameters studied by Ananth, Dickson, Olson, & Murano (1998) were affected by pressure of 414 MPa in raw loin pork. The b* (yellowness) values of Catalan thin, cured, dry the b* values of pork sausage (fuet) were reduced significantly by pressure, whereas no changes were observed in chorizo, a sausage made with ground pork and spicy seasonings (Aymerich, Marcos, & Garriga, 2005). Omana et al. (2011) reported decreasing b* values with increase in applied pressure in pressure-treated chicken breast fillet. In this study, the b* values of all the treatments of RTE sliced chicken breast roast did not change during storage, irrespective of the type of treatment, storage time or applied pressure (Figure 20 a,b &c). However, the disagreements between our results and other reports with respect to the a* and b* values of could be attributed to the "reversible" denaturation process of myoglobin which has been reported to occur through storage time (Cheah & Ledward, 1996).

6.3 Instrumental analysis of texture

In this study, shear force was used as an index to measure the firmness or hardness of the treated samples. It was observed that the texture of the samples was not affected by the applied pressure or concentrations of LAE used (Figure 21 a,b,c). There was no negatively perceived effect of pressure on the sensory texture (section 6.4). The firmness of the treated samples was not significantly (*P* >0.05) different from the untreated control samples on day 1.

In a study conducted by Jo et al. (2011), texture profile analysis parameters of chicken breast fillet such as cohesiveness, gumminess, hardness and chewiness increased with increasing pressure. Pressure applied at 450 and 600 MPa inflicted the most detrimental effects on the chicken breast fillet samples in the study conducted by Jo et al. (2011). In a study by Villacis, Rastogi and Balasubramaniam (2008), turkey breast muscles treated with pressures above 150 MPa became hard, gummy, and chewy with increasing pressure. Cohesiveness of the turkey breast muscles also increased with pressure holding time at pressures >150 MPa. A similar effect of increased pressure on hardness was observed in fish meat (Master, Stegeman, Kals, & Bartels, 2000) and beef muscles (Ma & Ledward, 2004). Firmness is the most important texture attribute to consumers and dictates the commercial value of a RTE meat product (Chamers & Bowers, 1993). The stability in texture of the samples treated with HP in this study, can be attributed to the cooking step prior to the pressure treatment.

In another study using high pressure on shredded stirred-curd cheddar cheese, it was observed that hardness increased at day 1; the hardness increased with higher pressure levels and pressurization times (Serrano, Velazquez, Lopetcharat, Ramirez, & Torres, 2004). The hardness values of cheese samples pressurized for 3 min at 345 MPa and 483 MPa were not significantly different but the hardness was markedly higher in the untreated samples.

From previous studies discussed in this section, high pressure does have an effect on the texture properties of food products. However, in the present study, the firmness of the RTE sliced chicken breast roast samples treated with high pressure was not significantly ($P > 0.05$) altered, which is in contradiction with other mentioned studies. The discrepancies between our studies and previous work may be attributed to different equipment used and processing parameters.

6.4 Consumer sensory analysis

Sensory attributes were evaluated for the selected pressure-treated, pressure + LAE-treated and LAE-treated samples. High pressure processing did not affect the flavour, texture and freshness of the RTE sliced chicken breast roast. Similar results were obtained for the HPP+LAE-treated samples. However, LAE (200 and 315 ppm)-treated samples were significantly ($P < 0.05$) different from the rest of the treatments in flavour, texture and freshness (Table 9). The instrumental color analysis revealed no changes in the L* and b* values. However, the decrease in the a* values was not detected by the consumer panelists.

In a similar study by Hayman et al. (2004), no evidence of deteriorating effects of high pressure treatment on sensory quality of various meat products were observed, even for products treated with 600 MPa pressure. Crehana, Troya and Buckley (2000) also concluded that high pressure processing does not markedly alter taste, flavour, or nutrient content of food. However, results obtained by Jo et al. (2011) demonstrated that pressure treatment can impact flavour, juiciness and aroma pleasantness of chicken breast fillet. Rivas-Canedo, Fernandez-Garcia, and Nunez (2008) showed that pressurization (400 MPa) of minced beef and chicken breast significantly changed the levels of some volatile compounds, where a few alcohols and aldehydes were decreased whereas other compounds were more abundant in the highly processed meats. High pressure may also accelerate other reactions that impact food flavour. Cheah and Ledward (1996) reported that the changes leading to catalysis of lipids oxidation in pressure processed meat were initiated at about 300 MPa at room temperature. In this study, the aroma pleasantness of the samples was not investigated as a sensory attribute. The effect of pressure on juiciness has been reported by Crehana et al. (2000), who demonstrated that the application of 300 MPa pressure significantly increased juiciness of frankfurters. In this study, the texture of the pressure-treated samples remained stable after pressure treatment.

The effect of high pressure treatment on sensory attributes of chicken meat has variable effects that are beneficial in some cases and detrimental in other (Jo et al., 2011). The mechanism of the variable effects is not fully understood and requires further research (Cheftel, 1995a).

The samples treated with LAE alone showed significant reduction in scores for flavour and freshness by the consumers. However, no similar sensory-related study has been reported by other researchers. The perceived change in flavour and freshness could be attributed to the increase in the microbial counts in the LAE (200 and 315 ppm)-treated samples by week 8 of storage at 4°C. Spoilage microorganisms in cooked chicken meats include *Staphylococcus epidermidis, Bacillus* species, *Micrococcus* species, *Streptococcus* species, *Pseudomonas putida, Pseudomonas fluorescens, Aerococcus* species, which are commonly isolated from chicken processing environments (Toule & Murphy, 1978). However, the microorganisms found in the RTE sliced chicken breast roast treated with LAE were not identified in this study.

The shelf-life of RTE chicken breast roast when sodium metabisulphite was used was between 6-8 weeks (R. Biggs, personal communication, May 26, 2010). LAE (200 and 315 ppm) may be a suitable replacement for the current preservative (sodium metabisulphite) used in the RTE sliced chicken breast roast; also, sodium metabisulphite is known to impart a metallic taste to the product (R. Biggs, personal communication, May 26, 2010).

6.5 Estimation of sensory shelf-life of RTE sliced chicken breast roast

Presently, the RTE sliced chicken breast roast has a shelf-life of 6-8 weeks at 4°C when sodium metabisulphite is used as a preservative (Seemeen & Mutukumira, 2010). Survival analysis was used as a methodology to determine the sensory shelf-life of the treated samples in this study. The results obtained in this study using the survival analysis were highly correlated to the results obtained from the consumer sensory analysis (Tables 9 and 11).

The application of survival analysis in shelf-life estimation of food products can be computed. Twenty to thirty sensory panelists are required to express their overall acceptability of samples at different storage times for different treatments. The number of sensory panelists who participated in this study was sufficient to estimate the shelf-life. Survival analysis is considered appropriate for this study as these measures are carried out directly on the data obtained in the consumer sensory tests (Hough, 2006). Consumer studies are an appropriate tool for determining the sensory shelf-life of foods. However, no studies have reported the use of consumer sensory data to determine the shelf-life of RTE sliced chicken breast roast when processed using HP and LAE.

In survival analysis statistics, the Weibull distribution model has been widely used to preview the shelf-life of dairy foods, such as cottage cheese (Cruz et al., 2010), concentrated yoghurt (Al-Kadamany et al., 2002), ricotta cheese (Hough, Pugleiso, Sanchez, & Silva, 1999), yayik butter (Arslan, Sert, Ayar, & Ozcan, 2009), shiitake mushrooms (Ares et al., 2006), and RTE lettuce (Araneda, Hough, & Penna, 2011).

In this study, twenty to thirty consumer panelists were recruited at different storage times to determine the shelf-life of RTE sliced chicken breast roast. The overall acceptability score was used to model the rejection time of the product/sample. A score of 5 (neither like nor dislike) or below 5 (dislike) was set as “not acceptable” on a 1 (dislike extremely) to 9 (like extremely) overall acceptability scale. Similarly, Kim et al. (2005) used three trained assessors to determine the shelf-life of modified-atmosphere packaged lettuce, arbitrarily setting a score of 3 (strong) as “not acceptable” on a 0 (none) to 4 (severe) off-odor scale. A similar scale and arbitrary cut off point was also used by McKellar et al. (2004) in their study on the influence of chlorinated water treatment and packaging on the shelf-life of RTE lettuce.

As discussed by Hough et al. (2003), food products do not have sensory shelf lives of their own; rather they depend on the interactions with the consumer. Jacxsens et al. (2002) reported a sensory shelf-life of 7 days for lettuce stored in an equillibrium modified-atmosphere package at 4°C. This value was obtained with 8-10 trained assessors who used a freshness scale from 1 (extremely fresh) to 10 (extremely deteriorated); the sample was considered unacceptable when a mean score above 5 was reached. In this study, a freshness scale of 1 (least fresh) to 9 (extremely fresh) was used and the sample was considered unacceptable when a mean score of 5 or below was obtained (Table 9). Hough et al. (2003) applied survival analysis statistics to determine the sensory shelf-life of foods based on the consumer rejecting the product, rather than on the product deteriorating.

As reported by Cruz et al. (2010), the shelf-life of probiotic yogurt was obtained by substituting the parameters found in the Weibull distribution model fit and subsequent consideration of the following values: 25% and 50% consumer rejection. The two rejection % values mentioned here were used in this study. Other researchers have also used this criterion in calculating the shelf-life of coffee (Cardelli & Labuza, 2001), yogurt (Curia, Aguerrido, Langohr, & Hough, 2005), and shiitake mushrooms (Ares et al., 2006).

In this study and in the various studies mentioned in this section, the focus of the shelf-life has been on the probability of a consumer rejecting a product after a certain storage time.

7.0 CONCLUSION:

Ready-to-eat chicken breast roast samples were treated with HP and HP+LAE (200 ppm), at 450 MPa for 1 min, 3.5 min and 5 min, HP and HP+LAE (200 ppm) at 600 MPa for 1 min, 3.5 min and 5 min, and LAE at 200 ppm. In the initial phase the treatments were screened based on their ability to inhibit the growth of APCs, LAB and Y&M when stored at 4°C for 16 weeks. The levels of LAB and Y&M were below levels of detection in all the treatment samples including the control (without preservative and/or pressure treatment). Samples treated with HP and HP+LAE at 450 MPa were capable of inhibiting the growth of APCs upto 12 weeks. Samples treated with HP and HP+LAE at 600 MPa were capable of inhibiting the growth of APCs upto 16 weeks. Samples treated with LAE at 200 ppm were successful in inhibiting the growth of APCs for 11 weeks. Therefore, a higher concentration of LAE was used in the subsequent investigations.

Results of the instrumental analysis of color for HPP treated samples, alone and in combination with LAE (200 ppm) showed that use of high pressure did not affect the L* and b* values. However, a decrease in the a* values was observed with increasing pressure. Texture analysis of the samples revealed that the texture of the samples was not affected by the treatment pressures, hold times, and levels of LAE used.

Samples treated with HP at 600 MPa for 1 min and 5 min, HP+LAE (200 ppm) at 600 MPa for 1 min, LAE at 200 ppm and 315 ppm were evaluated by consumer sensory panels. The results of the consumer sensory analysis showed no changes in color, texture, flavour and freshness of the HP-treated and HP+LAE (200 ppm)-treated samples. However, the LAE (200 and 315 ppm)-treated samples were described as “stale” and “distasteful” at week 12.

Survival analysis was employed as a tool for estimating the sensory shelf-life of the products. For a 25% consumer rejection, estimated sensory shelf-life of the samples treated with HP at 600 MPa for 1 min was 13.8 weeks, HP at 600 MPa for 5 min was 10.5 weeks, HP+LAE (200 ppm) at 600 MPa for 1 min was 8.3 weeks, LAE (200 ppm) was 7.2 weeks and LAE (315 ppm) was 6 weeks. Samples treated with HP alone at 600

MPa pressure for 1 min and 5 min had significantly higher sensory shelf-life as compared to the other treatments.

For 50% consumer rejection, the estimated shelf-life was >16 weeks for samples treated with 600 MPa pressure for 1 min and 5 min, 14.3 weeks for samples treated with HP+LAE (200 ppm), 10.1 for samples treated with LAE (200 ppm) and 9.2 for samples treated with LAE (315 ppm).

Therefore, results obtained in this study showed that HPP at 600 MPa for 1 min and 5 min, and HPP+LAE at 600 MPa for 1 min are efficient methods for extending the microbial shelf-life of the RTE sliced chicken breast roast. The study also showed that samples treated with 600 MPa had the best sensory properties during shelf-life.

8.0 RECOMMENDATIONS

Based on the experimental results and discussion, a few recommendations can be made for further research:

1. The pressure treatments used in this study were carried out at ambient temperatures. However, temperature-assisted high pressure processing can be explored as a possibility to further enhance the quality parameters (microbial, physicochemical and sensory) of products.

2. Physiochemical analysis of pressure treated and LAE-treated samples was not investigated in this study. Further research is needed in order to better understand the effect of these changes on the sensory characteristics of RTE chicken breast meat.

3. LAE alone was incapable of extending the shelf-life of the RTE sliced chicken breast roast to 16 weeks. However, LAE can be used as a replacement for currently used chemical preservatives in RTE meat products, such as sodium metabisulphite and nitrites. Further studies can be carried out to investigate the potential impact of the levels of LAE tested in this study on the sensory and functional attributes of the product.

4. Very little information is published on the degradation of the levels of LAE through the storage period. Analysis of the levels of LAE in the samples at various points of storage including the end of storage period is recommended.

5. Further work on microbial, sensory, physiochemical and morphological changes occurring in the LAE treated meat and its effect on the sensory properties of meat is suggested.

6. The bacterial populations (APCs) obtained in this study in all the treatments were not identified. Surviving populations of APCs after pressure treatment can be identified for better understanding of the pressure conditions and hold times that can be used for commercialization of the technology.

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APPENDIX 1

1.1 Specifications for product preparation (screening phase)

The following treatments are to be prepared in batches as explained below

Treatment 1- Control (No preservative/HPP)
Treatment 2- HPP alone (No preservative)
Treatment 3- LAE (200 ppm) + HPP
Treatment 4- LAE (200ppm) alone

1. The ingredients are listed in the order that they are to be added.

Batch 1: Treatment 1+2

Code	**Description**	**Quantity per batch (kg)**	**Product composition (%)**
DONOR			
026302	Fz Bflt Sl to FP Tempered	40	80
INGREDIENTS			
447848	Chilled water	8	16
441102	Brine Boneless Chicken #2 113796008	1.475	2.95
440154	Carrageenan Grindsted 62375	0.094	0.19
440272	Gelpro, Modified starch	0.20	0.40
447299	Salt Pure Dried Vacuum	0.235	0.47
TOTAL		**49.999 kg**	**100.01%**

Batch 2: Treatment 3+4

Code	**Description**	**Quantity per batch (kg)**	**Product composition (%)**
DONOR			
026302	Fz Bflt Sl to FP Tempered	40	79.86
INGREDIENTS			
447848	Chilled water	8	16
441102	Brine Boneless Chicken #2 113796008	1.475	2.94
440154	Carrageenan Grindsted 62375	0.094	0.19
-	LAE (Mirenat N)	0.08	0.16
440272	Gelpro, Modified starch	0.20	0.39
447299	Salt Pure Dried Vacuum	0.235	0.46
TOTAL		**49.99 kg**	**100%**

2. PACKAGING

PACKAGING REQUIREMENTS		
Code	**Description**	**Requirements (total for all the batches)**
420042	CASING 165 mm Flat Colour Case (Brown)	60m
420133	Clip E222	120 pcs
431740	WRAP H204 Hand 500mm × 440m 17	25m
420297	BULK BIN	1

3. Cooking will be carried out as per the set program. Batch 3 (highest concentration of LAE) will be placed at the top level of the oven, Batch 2 below that and Batch 1 in the lowest level (no LAE).

1.2 Specifications for product preparation (selection phase)

The following treatments are to be prepared in batches as explained below

Treatment 1- Control (No preservative/HPP)
Treatment 2- HPP alone (No preservative)
Treatment 3- LAE (200 ppm) + HPP
Treatment 4- LAE (200 ppm) alone
Treatment 5- LAE (315p pm) alone

1. The ingredients are listed in the order that they are to be added.

Batch 1: Treatment 1+2

Code	Description	Quantity per batch (kg)	Product composition (%)
DONOR			
026302	Fz Bflt Sl to FP Tempered	40	80
INGREDIENTS			
447848	Chilled water	8	16
441102	Brine Boneless Chicken #2 113796008	1.475	2.95
440154	Carrageenan Grindsted 62375	0.094	0.19
440272	Gelpro, Modified starch	0.2	0.40
447299	Salt Pure Dried Vacuum	0.235	0.47
TOTAL		**49.999 kg**	**100.01%**

Batch 2: Treatment 3+4

Code	Description	Quantity per batch (kg)	Product composition (%)
DONOR			
026302	Fz Bflt Sl to FP Tempered	40	79.86
INGREDIENTS			
447848	Chilled water	8	16
441102	Brine Boneless Chicken #2 113796008	1.475	2.94
440154	Carrageenan Grindsted 62375	0.094	0.19
-	LAE (Mirenat N)	0.08	0.16
440272	Gelpro, Modified starch	0.2	0.39
447299	Salt Pure Dried Vacuum	0.235	0.46
TOTAL		**49.999 kg**	**100%**

Batch 3: Treatment 5

Code	Description	Quantity per batch (kg)	Product composition (%)
DONOR			
026302	Fz Bflt Sl to FP Tempered	20	79.80
INGREDIENTS			
447848	Chilled water	4	16
441102	Brine Boneless Chicken #2 113796008	0.735	2.94
440154	Carrageenan Grindsted 62375	0.047	0.19
	LAE (Mirenat N)	0.063	0.25
440272	Gelpro, Modified starch	0.10	0.39
447299	Salt Pure Dried Vacuum	0.117	0.46
TOTAL		**25.063 kg**	**100.03%**

2. PACKAGING

PACKAGING REQUIREMENTS		
Code	**Description**	**Requirements (total for all the batches)**
420042	CASING 165 mm Flat Colour Case (Brown)	60m
420133	Clip E222	120 pcs
431740	WRAP H204 Hand 500mm × 440m 17	25m
420297	BULK BIN	1

3. Cooking will be carried out as per the set program. Batch 3 (highest concentration of LAE) will be placed at the top level of the oven, Batch 2 below that and Batch 1 in the lowest level (no LAE).

APPENDIX 2

2.1 Distribution temperature of the screened treatments during transport

Table 2.1: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
7/09/2010 11:54:50 A9P9	8.69
7/09/2010 12:54:50 A9P9	8.67
7/09/2010 13:54:50 A9P9	3.98
7/09/2010 14:54:50 A9P9	3.06
7/09/2010 15:54:50 A9P9	2.55
7/09/2010 16:54:50 A9P9	2.37
7/09/2010 17:54:50 A9P9	1.37
7/09/2010 18:54:50 A9P9	1.01
7/09/2010 19:54:50 A9P9	1.22
7/09/2010 20:54:50 A9P9	1.38
7/09/2010 21:54:50 A9P9	1.40
7/09/2010 22:54:50 A9P9	1.31
7/09/2010 23:54:50 A9P9	1.19
8/09/2010 0:54:50 A9P9	1.01
8/09/2010 1:54:50 A9P9	0.50
8/09/2010 2:54:50 A9P9	0.13
8/09/2010 3:54:50 A9P9	0.04
8/09/2010 4:54:50 A9P9	0.14
8/09/2010 5:54:50 A9P9	0.43
8/09/2010 6:54:50 A9P9	0.44
8/09/2010 7:54:50 A9P9	0.12
8/09/2010 8:54:50 A9P9	0.52
8/09/2010 9:54:50 A9P9	-0.38
8/09/2010 10:54:50 A9P9	-0.60
8/09/2010 11:54:50 A9P9	-0.68
8/09/2010 12:54:50 A9P9	-0.57
8/09/2010 13:54:50 A9P9	0.10
8/09/2010 14:54:50 A9P9	-0.03
8/09/2010 15:54:50 A9P9	0.11
8/09/2010 16:54:50 A9P9	0.29
8/09/2010 17:54:50 A9P9	-0.61
8/09/2010 18:54:50 A9P9	-0.98
8/09/2010 19:54:50 A9P9	-0.99
8/09/2010 20:54:50 A9P9	-0.64
8/09/2010 21:54:50 A9P9	-0.27
8/09/2010 22:54:50 A9P9	-0.15
8/09/2010 23:54:50 A9P9	-0.27
9/09/2010 0:54:50 A9P9	-0.19
9/09/2010 1:54:50 A9P9	-0.78
9/09/2010 2:54:50 A9P9	-1.02

Table 2.1$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
9/09/2010 3:54:50 A9P9	-1.08
9/09/2010 4:54:50 A9P9	-1.06
9/09/2010 5:54:50 A9P9	-1.10
9/09/2010 6:54:50 A9P9	-0.62
9/09/2010 7:54:50 A9P9	-0.39
9/09/2010 8:54:50 A9P9	-0.03
9/09/2010 9:54:50 A9P9	0.57
9/09/2010 10:54:50 A9P9	0.78
9/09/2010 11:54:50 A9P9	0.87
9/09/2010 12:54:50 A9P9	0.99
9/09/2010 13:54:50 A9P9	1.30
9/09/2010 14:54:50 A9P9	1.57
9/09/2010 15:54:50 A9P9	1.79
9/09/2010 16:54:50 A9P9	1.93
9/09/2010 17:54:50 A9P9	2.09
9/09/2010 18:54:50 A9P9	2.22
9/09/2010 19:54:50 A9P9	2.37
9/09/2010 20:54:50 A9P9	2.46
9/09/2010 21:54:50 A9P9	2.55
9/09/2010 22:54:50 A9P9	2.64
9/09/2010 23:54:50 A9P9	2.70
10/09/2010 0:54:50 A9P9	2.76
10/09/2010 1:54:50 A9P9	2.82
10/09/2010 2:54:50 A9P9	2.89
10/09/2010 3:54:50 A9P9	2.93
10/09/2010 4:54:50 A9P9	2.99
10/09/2010 5:54:50 A9P9	3.03
10/09/2010 6:54:50 A9P9	3.07
10/09/2010 7:54:50 A9P9	3.13
10/09/2010 8:54:50 A9P9	3.18
10/09/2010 9:54:50 A9P9	3.23
10/09/2010 10:54:50 A9P9	3.25
10/09/2010 11:54:50 A9P9	3.29
10/09/2010 12:54:50 A9P9	3.31
10/09/2010 13:54:50 A9P9	3.36
10/09/2010 14:54:50 A9P9	2.69
10/09/2010 15:54:50 A9P9	2.34
10/09/2010 16:54:50 A9P9	1.96
10/09/2010 17:54:50 A9P9	2.44
10/09/2010 18:54:50 A9P9	1.94
10/09/2010 19:54:50 A9P9	3.20
10/09/2010 20:54:50 A9P9	2.72
10/09/2010 21:54:50 A9P9	2.68

Table 2.1$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
10/09/2010 22:54:50 A9P9	2.68
10/09/2010 23:54:50 A9P9	2.69
11/09/2010 0:54:50 A9P9	2.70
11/09/2010 1:54:50 A9P9	2.77
11/09/2010 2:54:50 A9P9	2.69
11/09/2010 3:54:50 A9P9	2.58
11/09/2010 4:54:50 A9P9	2.46
11/09/2010 5:54:50 A9P9	2.44
11/09/2010 6:54:50 A9P9	2.40
11/09/2010 7:54:50 A9P9	2.38
11/09/2010 8:54:50 A9P9	3.63
11/09/2010 9:54:50 A9P9	4.12
11/09/2010 10:54:50 A9P9	4.30
11/09/2010 11:54:50 A9P9	4.35
11/09/2010 12:54:50 A9P9	4.51
11/09/2010 13:54:50 A9P9	4.51
11/09/2010 14:54:50 A9P9	3.15
11/09/2010 14:56:50 A9P9	3.17
11/09/2010 14:58:50 A9P9	3.16
11/09/2010 15:00:50 A9P9	3.17
11/09/2010 15:02:50 A9P9	3.17
11/09/2010 15:04:50 A9P9	3.17
11/09/2010 15:06:50 A9P9	3.17
11/09/2010 15:08:50 A9P9	3.17
11/09/2010 15:10:50 A9P9	3.17
11/09/2010 15:12:50 A9P9	3.17
11/09/2010 15:14:50 A9P9	3.19
11/09/2010 15:16:50 A9P9	3.19
11/09/2010 15:18:50 A9P9	3.19
11/09/2010 15:20:50 A9P9	3.19
11/09/2010 15:22:50 A9P9	3.21
11/09/2010 15:24:50 A9P9	3.19
11/09/2010 15:26:50 A9P9	3.21
11/09/2010 15:28:50 A9P9	3.21
11/09/2010 15:30:50 A9P9	3.21
11/09/2010 15:32:50 A9P9	3.21
11/09/2010 15:34:50 A9P9	3.21
11/09/2010 15:36:50 A9P9	3.21
11/09/2010 15:38:50 A9P9	3.23
11/09/2010 15:40:50 A9P9	3.23
11/09/2010 15:42:50 A9P9	3.23
11/09/2010 15:44:50 A9P9	3.25
11/09/2010 15:46:50 A9P9	3.25

Table 2.1$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
11/09/2010 15:48:50 A9P9	3.25
11/09/2010 15:50:50 A9P9	3.26
11/09/2010 15:52:50 A9P9	3.25
11/09/2010 15:54:50 A9P9	3.25
11/09/2010 16:54:50 A9P9	1.73
11/09/2010 17:54:50 A9P9	0.95
11/09/2010 18:54:50 A9P9	0.75
11/09/2010 19:54:50 A9P9	0.64
11/09/2010 20:54:50 A9P9	0.74
11/09/2010 21:54:50 A9P9	1.04
11/09/2010 22:54:50 A9P9	1.34
11/09/2010 23:54:50 A9P9	1.56
12/09/2010 0:54:50 A9P9	1.72
12/09/2010 1:54:50 A9P9	1.71
12/09/2010 2:54:50 A9P9	0.56
12/09/2010 3:54:50 A9P9	0.31
12/09/2010 4:54:50 A9P9	0.08
12/09/2010 4:56:50 A9P9	0.10
12/09/2010 5:54:50 A9P9	-0.01
12/09/2010 6:54:50 A9P9	0.08
12/09/2010 7:54:50 A9P9	0.54
12/09/2010 8:54:50 A9P9	0.58
12/09/2010 9:54:50 A9P9	0.61
12/09/2010 10:54:50 A9P9	0.61
12/09/2010 11:54:50 A9P9	0.61
12/09/2010 12:54:50 A9P9	0.58
12/09/2010 13:54:50 A9P9	0.58
12/09/2010 14:54:50 A9P9	0.60
12/09/2010 15:54:50 A9P9	0.57
12/09/2010 16:54:50 A9P9	0.54
12/09/2010 17:54:50 A9P9	0.52
12/09/2010 18:54:50 A9P9	0.50
12/09/2010 19:54:50 A9P9	0.45
12/09/2010 20:54:50 A9P9	0.43
12/09/2010 21:54:50 A9P9	0.66
12/09/2010 22:54:50 A9P9	0.62
12/09/2010 23:54:50 A9P9	0.64
13/09/2010 0:54:50 A9P9	0.54
13/09/2010 1:54:50 A9P9	0.50
13/09/2010 2:54:50 A9P9	0.45
13/09/2010 3:54:50 A9P9	0.45
13/09/2010 4:54:50 A9P9	1.02
13/09/2010 5:54:50 A9P9	0.72

Table 2.1$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
13/09/2010 6:54:50 A9P9	0.58
13/09/2010 7:54:50 A9P9	0.52
13/09/2010 8:54:50 A9P9	0.69
13/09/2010 9:54:50 A9P9	0.65
13/09/2010 10:54:50 A9P9	0.65
13/09/2010 11:54:50 A9P9	0.77
13/09/2010 12:54:50 A9P9	0.62
13/09/2010 13:54:50 A9P9	0.76
13/09/2010 14:54:50 A9P9	0.84
13/09/2010 15:54:50 A9P9	1.00
13/09/2010 16:54:50 A9P9	1.17
13/09/2010 17:54:50 A9P9	1.20
13/09/2010 18:54:50 A9P9	1.23
13/09/2010 19:54:50 A9P9	1.15
13/09/2010 20:54:50 A9P9	1.08
13/09/2010 21:54:50 A9P9	1.56
13/09/2010 22:54:50 A9P9	1.02
13/09/2010 23:54:50 A9P9	0.92
14/09/2010 0:54:50 A9P9	0.58
14/09/2010 1:54:50 A9P9	0.52
14/09/2010 2:54:50 A9P9	0.45
14/09/2010 3:54:50 A9P9	0.45
14/09/2010 4:56:50 A9P9	0.92
14/09/2010 5:54:50 A9P9	0.63
14/09/2010 6:54:50 A9P9	0.50
14/09/2010 7:54:50 A9P9	0.47
14/09/2010 8:54:50 A9P9	0.54
14/09/2010 9:54:50 A9P9	0.56
14/09/2010 10:54:50 A9P9	0.63
14/09/2010 11:54:50 A9P9	0.45
14/09/2010 12:54:50 A9P9	0.90
14/09/2010 13:54:50 A9P9	1.52
14/09/2010 14:54:50 A9P9	2.06
14/09/2010 15:54:50 A9P9	-0.86
14/09/2010 16:54:50 A9P9	3.02
14/09/2010 17:54:50 A9P9	2.78
14/09/2010 18:54:50 A9P9	2.54
14/09/2010 19:54:50 A9P9	2.90
14/09/2010 20:54:50 A9P9	2.70
14/09/2010 21:54:50 A9P9	3.06
14/09/2010 22:54:50 A9P9	2.83
14/09/2010 23:54:50 A9P9	3.19
15/09/2010 0:54:50 A9P9	3.00

Table 2.1$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
15/09/2010 1:54:50 A9P9	3.24
15/09/2010 2:54:50 A9P9	3.10
15/09/2010 3:54:50 A9P9	3.32
15/09/2010 4:54:50 A9P9	3.12
15/09/2010 5:54:50 A9P9	3.32
15/09/2010 6:54:50 A9P9	3.12
15/09/2010 7:54:50 A9P9	3.27
15/09/2010 8:54:50 A9P9	3.17
15/09/2010 9:54:50 A9P9	3.02
15/09/2010 10:54:50 A9P9	3.08
15/09/2010 11:54:50 A9P9	3.23
15/09/2010 12:54:50 A9P9	3.32
15/09/2010 13:54:50 A9P9	3.34
15/09/2010 14:54:50 A9P9	3.23
15/09/2010 15:54:50 A9P9	3.23
15/09/2010 16:54:50 A9P9	3.37
15/09/2010 17:54:50 A9P9	3.25
15/09/2010 18:54:50 A9P9	3.10
15/09/2010 19:54:50 A9P9	3.27
15/09/2010 20:54:50 A9P9	3.39
15/09/2010 21:54:50 A9P9	3.47
15/09/2010 22:54:50 A9P9	3.57
15/09/2010 23:54:50 A9P9	3.64
16/09/2010 0:54:50 A9P9	3.27
16/09/2010 1:54:50 A9P9	3.32
16/09/2010 2:54:50 A9P9	3.51
16/09/2010 3:54:50 A9P9	3.32
16/09/2010 4:54:50 A9P9	3.39
16/09/2010 5:54:50 A9P9	3.28
16/09/2010 6:54:50 A9P9	3.43
16/09/2010 7:54:50 A9P9	3.21
16/09/2010 8:54:50 A9P9	3.56
16/09/2010 9:54:50 A9P9	3.50
16/09/2010 10:54:50 A9P9	3.39
16/09/2010 11:54:50 A9P9	3.34
16/09/2010 12:54:50 A9P9	3.39
16/09/2010 13:54:50 A9P9	3.39
16/09/2010 14:54:50 A9P9	3.12
16/09/2010 15:54:50 A9P9	3.06
16/09/2010 16:54:50 A9P9	3.10
16/09/2010 17:54:50 A9P9	3.37
16/09/2010 18:54:50 A9P9	3.29
16/09/2010 19:54:50 A9P9	3.12

Table 2.1$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the first phase.

Date	Temperature
16/09/2010 20:54:50 A9P9	3.39
16/09/2010 21:54:50 A9P9	3.18
16/09/2010 22:54:50 A9P9	3.45
16/09/2010 23:54:50 A9P9	3.20
17/09/2010 0:54:50 A9P9	3.40
17/09/2010 1:54:50 A9P9	3.60
17/09/2010 2:54:50 A9P9	3.23
17/09/2010 3:54:50 A9P9	3.40
17/09/2010 4:54:50 A9P9	3.53
17/09/2010 5:54:50 A9P9	3.29
17/09/2010 6:54:50 A9P9	3.46
17/09/2010 7:54:50 A9P9	3.21
17/09/2010 8:54:50 A9P9	3.21
17/09/2010 9:12:50 A9P9	3.33
17/09/2010 9:14:50 A9P9	3.39
17/09/2010 9:16:50 A9P9	3.42
17/09/2010 9:18:50 A9P9	3.34
17/09/2010 9:20:50 A9P9	3.75
17/09/2010 9:22:50 A9P9	6.23

2.2 Distribution temperature of the selected treatments during transport

Table 2.2: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the second phase.

Date	Temperature
27/01/2011 15:59:06 A1P1	5.52
27/01/2011 16:59:06 A1P1	5.14
27/01/2011 17:59:06 A1P1	4.81
27/01/2011 18:59:06 A1P1	4.59
27/01/2011 19:59:06 A1P1	4.51
27/01/2011 20:59:06 A1P1	4.41
27/01/2011 21:59:06 A1P1	4.26
27/01/2011 22:59:06 A1P1	4.18
27/01/2011 23:59:06 A1P1	4.10
28/01/2011 0:59:06 A1P1	3.87
28/01/2011 1:59:06 A1P1	3.64
28/01/2011 2:59:06 A1P1	3.53
28/01/2011 3:59:06 A1P1	3.41
28/01/2011 4:59:06 A1P1	3.33
28/01/2011 5:59:06 A1P1	3.22
28/01/2011 6:59:06 A1P1	3.33
28/01/2011 7:59:06 A1P1	3.59
28/01/2011 8:59:06 A1P1	3.64
28/01/2011 9:59:06 A1P1	3.79
28/01/2011 10:59:06 A1P1	3.65
28/01/2011 11:59:06 A1P1	3.92
28/01/2011 12:59:06 A1P1	4.22
28/01/2011 13:59:06 A1P1	4.88
28/01/2011 14:59:06 A1P1	7.37
28/01/2011 15:59:06 A1P1	4.07
28/01/2011 16:59:06 A1P1	3.59
28/01/2011 17:59:06 A1P1	2.89
28/01/2011 18:59:06 A1P1	2.63
28/01/2011 19:59:06 A1P1	3.32
28/01/2011 20:59:06 A1P1	2.70
28/01/2011 21:59:06 A1P1	2.07
28/01/2011 22:59:06 A1P1	1.84
28/01/2011 23:59:06 A1P1	2.22
29/01/2011 0:59:06 A1P1	3.00
29/01/2011 1:59:06 A1P1	2.68
29/01/2011 2:59:06 A1P1	2.93

Table 2.2$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the second phase.

Date	Temperature
29/01/2011 3:59:06 A1P1	2.75
29/01/2011 4:59:06 A1P1	2.23
29/01/2011 5:59:06 A1P1	2.10
29/01/2011 6:59:06 A1P1	2.23
29/01/2011 7:59:06 A1P1	1.82
29/01/2011 8:59:06 A1P1	1.74
29/01/2011 9:59:06 A1P1	1.61
29/01/2011 10:59:06 A1P1	1.52
29/01/2011 11:59:06 A1P1	1.92
29/01/2011 12:59:06 A1P1	2.04
29/01/2011 13:59:06 A1P1	2.05
29/01/2011 14:59:06 A1P1	1.67
29/01/2011 15:59:06 A1P1	1.86
29/01/2011 16:59:06 A1P1	1.58
29/01/2011 17:59:06 A1P1	2.22
29/01/2011 18:59:06 A1P1	1.98
29/01/2011 19:59:06 A1P1	1.49
29/01/2011 20:59:06 A1P1	1.32
29/01/2011 21:59:06 A1P1	2.12
29/01/2011 22:59:06 A1P1	1.68
29/01/2011 23:59:06 A1P1	1.14
30/01/2011 0:59:06 A1P1	1.74
30/01/2011 1:59:06 A1P1	1.69
30/01/2011 2:59:06 A1P1	1.28
30/01/2011 3:59:06 A1P1	0.92
30/01/2011 4:59:06 A1P1	1.80
30/01/2011 5:59:06 A1P1	1.26
30/01/2011 6:59:06 A1P1	1.58
30/01/2011 7:59:06 A1P1	1.18
30/01/2011 8:59:06 A1P1	1.35
30/01/2011 9:59:06 A1P1	1.63
30/01/2011 10:59:06 A1P1	1.07
30/01/2011 11:59:06 A1P1	0.92
30/01/2011 12:59:06 A1P1	0.92
30/01/2011 13:59:06 A1P1	0.75
30/01/2011 14:59:06 A1P1	0.83
30/01/2011 15:59:06 A1P1	1.61
30/01/2011 16:59:06 A1P1	0.68
30/01/2011 17:59:06 A1P1	0.70
30/01/2011 18:59:06 A1P1	1.38
30/01/2011 19:59:06 A1P1	0.68
30/01/2011 20:59:06 A1P1	0.99
30/01/2011 21:59:06 A1P1	0.63

Table 2.2$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the second phase.

Date	Temperature
30/01/2011 22:59:06 A1P1	0.94
30/01/2011 23:59:06 A1P1	0.99
31/01/2011 0:59:06 A1P1	0.83
31/01/2011 1:59:06 A1P1	0.67
31/01/2011 2:59:06 A1P1	1.36
31/01/2011 3:59:06 A1P1	1.24
31/01/2011 4:59:06 A1P1	1.07
31/01/2011 5:59:06 A1P1	0.68
31/01/2011 6:59:06 A1P1	0.90
31/01/2011 7:59:06 A1P1	1.31
31/01/2011 8:59:06 A1P1	0.81
31/01/2011 9:59:06 A1P1	0.54
31/01/2011 10:59:06 A1P1	1.18
31/01/2011 11:59:06 A1P1	1.35
31/01/2011 12:59:06 A1P1	0.39
31/01/2011 13:59:06 A1P1	0.72
31/01/2011 14:59:06 A1P1	3.70
31/01/2011 15:59:06 A1P1	5.27
31/01/2011 16:59:06 A1P1	3.76
31/01/2011 17:59:06 A1P1	2.84
31/01/2011 18:59:06 A1P1	2.74
31/01/2011 19:59:06 A1P1	2.69
31/01/2011 20:59:06 A1P1	2.68
31/01/2011 21:59:06 A1P1	2.66
31/01/2011 22:59:06 A1P1	3.08
31/01/2011 23:59:06 A1P1	2.79
1/02/2011 0:59:06 A2P2	2.81
1/02/2011 1:59:06 A2P2	2.80
1/02/2011 2:59:06 A2P2	2.81
1/02/2011 3:59:06 A2P2	2.84
1/02/2011 4:59:06 A2P2	2.84
1/02/2011 5:59:06 A2P2	2.98
1/02/2011 6:59:06 A2P2	2.88
1/02/2011 7:59:06 A2P2	5.32
1/02/2011 8:59:06 A2P2	1.63
1/02/2011 9:59:06 A2P2	2.94
1/02/2011 10:59:06 A2P2	2.86
1/02/2011 11:59:06 A2P2	3.06
1/02/2011 12:59:06 A2P2	3.17
1/02/2011 13:59:06 A2P2	3.75
1/02/2011 14:59:06 A2P2	3.79
1/02/2011 15:59:06 A2P2	4.04
1/02/2011 16:59:06 A2P2	3.85

Table 2.2$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the second phase.

Date	Temperature
1/02/2011 17:59:06 A2P2	4.24
1/02/2011 18:59:06 A2P2	3.91
1/02/2011 19:59:06 A2P2	4.02
1/02/2011 20:59:06 A2P2	4.06
1/02/2011 21:59:06 A2P2	4.03
1/02/2011 22:59:06 A2P2	4.00
1/02/2011 23:59:06 A2P2	3.96
2/02/2011 0:59:06 A2P2	4.04
2/02/2011 1:59:06 A2P2	4.35
2/02/2011 2:59:06 A2P2	4.28
2/02/2011 3:59:06 A2P2	4.19
2/02/2011 4:59:06 A2P2	4.08
2/02/2011 5:59:06 A2P2	4.00
2/02/2011 6:59:06 A2P2	4.05
2/02/2011 7:59:06 A2P2	4.15
2/02/2011 8:59:06 A2P2	4.05
2/02/2011 9:59:06 A2P2	4.22
2/02/2011 10:59:06 A2P2	4.30
2/02/2011 11:59:06 A2P2	4.14
2/02/2011 12:59:06 A2P2	4.24
2/02/2011 13:59:06 A2P2	4.18
2/02/2011 14:59:06 A2P2	4.30
2/02/2011 15:59:06 A2P2	4.14
2/02/2011 16:59:06 A2P2	4.20
2/02/2011 17:59:06 A2P2	4.41
2/02/2011 18:59:06 A2P2	4.13
2/02/2011 19:59:06 A2P2	4.28
2/02/2011 20:59:06 A2P2	4.42
2/02/2011 21:59:06 A2P2	4.12
2/02/2011 22:59:06 A2P2	4.10
2/02/2011 23:59:06 A2P2	4.17
3/02/2011 0:59:06 A2P2	4.24
3/02/2011 1:59:06 A2P2	4.32
3/02/2011 2:59:06 A2P2	4.39
3/02/2011 3:59:06 A2P2	4.17
3/02/2011 4:59:06 A2P2	4.06
3/02/2011 5:59:06 A2P2	4.12
3/02/2011 6:59:06 A2P2	4.22
3/02/2011 7:01:06 A2P2	4.26
3/02/2011 7:03:06 A2P2	4.33
3/02/2011 7:05:06 A2P2	4.35
3/02/2011 7:07:06 A2P2	4.38
3/02/2011 7:09:06 A2P2	4.30

Table 2.2$_{\text{contd.}}$: Distribution temperature monitored during the transport of samples from Christchurch to Auckland during the second phase.

Date	Temperature
3/02/2011 7:11:06 A2P2	4.16
3/02/2011 7:13:06 A2P2	4.10
3/02/2011 7:15:06 A2P2	4.05
3/02/2011 7:17:06 A2P2	4.06
3/02/2011 7:19:06 A2P2	4.07
3/02/2011 7:21:06 A2P2	4.11
3/02/2011 7:23:06 A2P2	4.15
3/02/2011 7:25:06 A2P2	4.19
3/02/2011 7:27:06 A2P2	4.24
3/02/2011 7:29:06 A2P2	4.29
3/02/2011 7:31:06 A2P2	4.34

APPENDIX 3

3.1 Raw data for microbial counts (APCs) during screening of treatments

Table 3.1a: Growth of APCs in HP-treated RTE sliced chicken breast roast

Storage time		Control	HPP 450/1	HPP 450/3.5	HPP 450/5	HPP 600/1	HPP 600
Day 0	Sample 1	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 2	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 3	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 4	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	1.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Week 2	Sample 1	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 2	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 3	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 4	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	1.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Week 4	Sample 1	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 2	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 3	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 4	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	1.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**

Table 3.1a$_{\text{contd.}}$ Growth of APCs in HPP-treated RTE sliced chicken breast roast

Storage time		Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1	HPP/600
Week 6	Sample 1	2.79	1.00	1.00	1.00	1.00	1.00
	Sample 2	3.08	1.00	2.17	1.00	1.00	1.00
	Sample 3	2.86	1.00	2.04	1.00	1.00	1.00
	Sample 4	2.83	1.00	1.00	1.00	1.00	1.00
	Sample 5	2.93	1.00	1.00	1.00	1.48	1.00
	Mean	**2.90**	**1.00**	**1.44**	**1.00**	**1.10**	**1.00**
	SD	**0.11**	**0.00**	**0.61**	**0.00**	**0.21**	**0.00**
Week 8	Sample 1	3.85	1.00	1.00	1.00	1.10	1.00
	Sample 2	4.00	1.00	1.35	1.00	1.00	1.00
	Sample 3	3.07	1.00	1.04	1.00	1.00	1.00
	Sample 4	3.99	1.00	1.00	1.02	1.00	1.00
	Sample 5	3.76	1.00	1.00	1.00	1.00	1.00
	Mean	**3.73**	**1.00**	**1.08**	**1.00**	**1.02**	**1.00**
	SD	**0.38**	**0.00**	**0.61**	**0.38**	**0.90**	**0.00**
Week 9	Sample 1	3.71	1.00	1.56	1.20	1.00	1.00
	Sample 2	3.5	1.00	1.00	1.12	1.00	1.00
	Sample 3	3.78	1.00	1.47	1.20	1.00	2.23
	Sample 4	4.00	1.00	1.65	1.17	1.00	1.00
	Sample 5	3.89	1.00	1.32	1.16	1.00	1.00
	Mean	**3.78**	**1.00**	**1.40**	**1.17**	**1.00**	**1.25**
	SD	**0.39**	**0.00**	**0.25**	**0.01**	**0.00**	**0.55**
Week 10	Sample 1	4.01	1.00	1.47	1.10	1.00	1.81
	Sample 2	3.87	1.00	2.12	2.65	1.00	1.84
	Sample 3	4.43	1.00	2.20	2.40	1.00	2.01
	Sample 4	4.53	1.00	1.00	2.20	1.00	2.25
	Sample 5	3.22	1.00	1.56	1.00	1.00	2.17
	Mean	**4.01**	**1.00**	**1.67**	**1.87**	**1.00**	**2.02**
	SD	**0.52**	**0.00**	**0.50**	**0.77**	**0.00**	**0.19**

Table 3.1a$_{\text{contd.}}$ Growth of APCs in HP-treated RTE sliced chicken breast roast

Storage time		Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1	HPP/600
Week 11	Sample 1	3.15	1.00	2.45	2.07	1.00	2.20
	Sample 2	4.09	1.00	3.12	1.69	1.00	2.39
	Sample 3	4.79	1.00	2.76	1.90	1.00	2.65
	Sample 4	4.64	2.00	2.77	2.34	1.00	2.77
	Sample 5	4.05	2.10	3.00	2.18	1.00	2.90
	Mean	**4.14**	**1.42**	**2.82**	**2.04**	**1.00**	**2.58**
	SD	**0.57**	**0.58**	**0.26**	**0.25**	**0.00**	**0.28**
Week 12	Sample 1	4.25	2.20	3.34	1.45	1.00	3.08
	Sample 2	4.89	2.00	3.25	2.36	1.00	1.00
	Sample 3	4.70	1.97	3.78	2.69	1.00	3.34
	Sample 4	4.76	2.00	3.25	1.67	1.00	2.54
	Sample 5	4.50	2.10	3.70	2.41	1.00	2.40
	Mean	**4.62**	**2.05**	**3.46**	**2.12**	**1.00**	**2.47**
	SD	**1.49**	**0.10**	**0.28**	**0.73**	**0.00**	**1.28**
Week 13	Sample 1	4.98	1.97	4.10	1.77	1.00	2.09
	Sample 2	3.60	1.77	3.54	4.38	1.00	2.90
	Sample 3	4.56	2.00	4.01	4.44	2.39	2.13
	Sample 4	3.22	2.10	3.12	3.65	1.00	2.50
	Sample 5	4.20	2.30	3.55	3.50	1.00	2.94
	Mean	**4.11**	**2.03**	**3.66**	**3.53**	**1.46**	**2.51**
	SD	**1.80**	**0.13**	**0.52**	**1.52**	**0.80**	**0.64**
Week 14	Sample 1	4.74	4.43	4.47	3.30	1.00	5.40
	Sample 2	4.11	4.41	4.45	2.69	1.00	3.25
	Sample 3	4.67	4.14	4.46	3.23	2.34	2.41
	Sample 4	4.54	4.45	4.38	3.70	1.00	3.70
	Sample 5	4.15	4.30	4.60	2.70	1.00	3.69
	Mean	**4.44**	**4.35**	**4.46**	**3.12**	**1.45**	**3.69**
	SD	**0.29**	**0.13**	**0.01**	**0.61**	**0.77**	**1.54**

Table 3.1a$_{\text{contd.}}$ Growth of APCs in HP-treated RTE sliced chicken breast roast

Storage time		Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1	HPP/600
Week 15	Sample 1	4.50	4.86	4.74	3.40	1.00	4.45
	Sample 2	4.55	4.91	4.89	2.78	1.00	4.72
	Sample 3	4.67	4.75	4.97	3.25	1.00	4.46
	Sample 4	4.60	4.72	5.00	3.08	1.00	4.65
	Sample 5	4.23	4.90	4.9	2.85	1.00	4.40
	Mean	**4.51**	**4.83**	**4.87**	**3.07**	**1.00**	**4.54**
	SD	**0.20**	**0.09**	**0.12**	**0.33**	**0.00**	**0.15**
Week 16	Sample 1	5.11	4.55	4.78	3.21	1.00	5.10
	Sample 2	4.98	4.98	4.67	3.67	1.00	3.87
	Sample 3	4.56	5.32	4.91	3.98	1.00	4.75
	Sample 4	5.21	4.73	5.02	2.56	1.00	4.95
	Sample 5	5.20	5.15	5.12	2.76	1.00	4.70
	Mean	**5.01**	**4.95**	**4.90**	**3.24**	**1.00**	**4.67**
	SD	**0.89**	**0.16**	**0.45**	**0.78**	**0.00**	**0.32**

Note: Control= no HPP/no LAE; HPP/450/1= HPP at 450 MPa for 1 min; HPP/450/3.5= HP at 450 MPa for
HPP at 450 MPa for 5 min; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/3.5= HPP at 600 MPa for 3.5
600 MPa for 5 min.

Table 3.1b: Growth of APCs in HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	HPP+LAE 450/1	HPP+LAE 450/3.5	HPP+LAE 450/5	HPP+LAE 600/1
Day 0	Sample 1	1.00	1.00	1.00	1.00	1.00
	Sample 2	1.00	1.00	1.00	1.00	1.00
	Sample 3	1.00	1.00	1.00	1.00	1.00
	Sample 4	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Week 2	Sample 1	1.00	1.00	1.00	1.00	1.00
	Sample 2	1.00	1.00	1.00	1.00	1.00
	Sample 3	1.00	1.00	1.00	1.00	1.00
	Sample 4	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Week 4	Sample 1	1.00	1.00	1.00	1.00	1.00
	Sample 2	1.00	1.00	1.00	1.00	1.00
	Sample 3	1.00	1.00	1.00	1.00	1.00
	Sample 4	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.00**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Week 6	Sample 1	2.79	1.00	1.78	1.69	1.00
	Sample 2	3.08	1.00	1.06	1.00	1.00
	Sample 3	2.86	1.00	1.10	1.00	1.00
	Sample 4	2.83	1.00	1.00	1.00	1.00
	Sample 5	2.93	1.00	1.00	1.00	1.00
	Mean	**2.90**	**1.00**	**1.30**	**1.14**	**1.00**
	SD	**0.11**	**0.00**	**0.37**	**0.31**	**0.00**

Table 3.1b$_{\text{contd.}}$: Growth of APCs in HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	HPP+LAE 450/1	HPP+LAE 450/3.5	HPP+LAE 450/5	HPP+LAE 600/1
Week 8	Sample 1	3.85	2.04	1.00	3.51	1.00
	Sample 2	4.00	1.00	1.00	1.00	1.00
	Sample 3	3.07	1.00	2.76	1.00	1.00
	Sample 4	3.99	1.00	1.00	2.30	1.00
	Sample 5	3.76	1.00	1.00	1.00	1.00
	Mean	**3.73**	**1.21**	**1.35**	**1.76**	**1.00**
	SD	**0.38**	**0.47**	**1.88**	**1.13**	**0.00**
Week 9	Sample 1	3.71	1.00	1.00	1.00	1.00
	Sample 2	3.50	1.00	1.00	1.00	1.00
	Sample 3	3.78	1.50	2.34	1.00	1.00
	Sample 4	4.00	1.00	1.67	2.40	1.00
	Sample 5	3.89	1.00	1.00	2.00	1.00
	Mean	**3.78**	**1.10**	**1.40**	**1.48**	**1.00**
	SD	**0.39**	**0.22**	**0.60**	**0.67**	**0.00**
Week 10	Sample 1	4.01	1.00	1.00	1.00	1.00
	Sample 2	3.87	1.00	1.00	1.00	1.00
	Sample 3	4.43	1.20	1.67	1.00	1.00
	Sample 4	4.53	2.30	1.99	1.60	1.00
	Sample 5	3.22	1.00	1.00	1.00	1.00
	Mean	**4.01**	**1.30**	**1.33**	**1.12**	**1.00**
	SD	**0.52**	**0.40**	**0.33**	**0.27**	**0.00**
Week 11	Sample 1	3.15	1.00	1.00	1.00	1.00
	Sample 2	4.09	1.90	1.00	2.10	1.00
	Sample 3	4.79	1.67	2.24	1.80	1.00
	Sample 4	4.64	1.95	1.65	1.00	1.00
	Sample 5	4.05	1.00	1.00	1.00	1.00
	Mean	**4.14**	**1.50**	**1.38**	**1.38**	**1.00**
	SD	**0.57**	**0.30**	**0.42**	**0.53**	**0.00**

Table 3.1b$_{\text{contd.}}$: Growth of APCs in HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	HPP+LAE 450/1	HPP+LAE 450/3.5	HPP+LAE 450/5	HPP+LAE 600/1
Week 12	Sample 1	4.25	1.00	2.60	1.00	1.00
	Sample 2	4.89	2.20	3.40	1.75	1.00
	Sample 3	4.70	1.90	2.41	2.50	1.00
	Sample 4	4.76	2.10	3.00	1.00	1.00
	Sample 5	4.50	1.80	3.30	1.70	1.00
	Mean	**4.62**	**1.80**	**2.94**	**1.59**	**1.00**
	SD	**1.49**	**0.15**	**0.78**	**0.59**	**0.00**
Week 13	Sample 1	4.98	1.00	4.59	1.80	1.00
	Sample 2	3.60	2.20	2.61	1.70	1.00
	Sample 3	4.56	2.50	2.67	1.77	1.00
	Sample 4	3.22	1.30	2.78	1.79	1.00
	Sample 5	4.20	3.00	3.70	1.70	1.00
	Mean	**4.11**	**1.90**	**3.28**	**1.75**	**1.00**
	SD	**1.80**	**0.40**	**0.78**	**0.01**	**0.00**
Week 14	Sample 1	4.74	3.13	2.77	3.13	3.2
	Sample 2	4.11	2.17	3.25	4.19	1.00
	Sample 3	4.67	1.00	3.34	2.90	1.00
	Sample 4	4.54	2.90	2.72	3.00	1.00
	Sample 5	4.15	2.90	3.52	3.85	1.00
	Mean	**4.44**	**2.10**	**3.12**	**3.41**	**1.73**
	SD	**0.29**	**0.90**	**0.31**	**0.69**	**1.35**
Week 15	Sample 1	4.50	2.40	3.77	4.36	2.80
	Sample 2	4.55	2.39	4.20	4.54	4.00
	Sample 3	4.67	2.56	3.68	4.14	2.98
	Sample 4	4.60	3.00	3.50	4.60	2.00
	Sample 5	4.23	2.04	3.90	4.10	3.80
	Mean	**4.51**	**2.45**	**3.81**	**4.35**	**3.26**
	SD	**0.20**	**0.23**	**0.24**	**0.20**	**0.20**

Table 3.1b$_{\text{contd.}}$: Growth of APCs in HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	HPP+LAE 450/1	HPP+LAE 450/3.5	HPP+LAE 450/5	HPP+LAE 600/1
Week 16	Sample 1	5.11	2.50	3.98	4.90	3.40
	Sample 2	4.98	2.00	4.65	5.14	3.60
	Sample 3	4.56	2.60	4.00	4.50	2.50
	Sample 4	5.21	2.40	3.60	4.90	3.20
	Sample 5	5.20	2.15	4.35	4.55	4.30
	Mean	**5.01**	**2.33**	**4.12**	**4.80**	**3.40**
	SD	**0.89**	**0.34**	**0.54**	**0.45**	**0.85**

Note: Control= no HPP/no LAE; HPP+LAE/450/1= HPP + LAE (200 ppm) at 450 MPa for 1 min; HPP+LA
(200 ppm) at 450 MPa for 3.5 min; HPP+LAE/450/5= HPP + LAE (200 ppm) at 450 MPa for 5 min; HPP+L
(200 ppm) at 600 MPa for 1 min; HPP+LAE/600/3.5= HPP + LAE (200 ppm) at 600 MPa for 3.5 min; HPP+
(200 ppm) at 600 MPa for 5 min.

Table 3.1c: Growth of APCs in LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	LAE 200
Day 0	Sample 1	1.00	1.00
	Sample 2	1.00	1.00
	Sample 3	1.00	1.00
	Sample 4	1.00	1.00
	Sample 5	1.00	1.00
	Mean	**1.00**	**1.00**
	SD	**0.00**	**0.00**
Week 2	Sample 1	1.00	1.00
	Sample 2	1.00	1.00
	Sample 3	1.00	1.00
	Sample 4	1.00	1.00
	Sample 5	1.00	1.00
	Mean	**1.00**	**1.00**
	SD	**0.00**	**0.00**
Week 4	Sample 1	1.00	1.00
	Sample 2	1.00	1.00
	Sample 3	1.00	1.00
	Sample 4	1.00	1.00
	Sample 5	1.00	1.00
	Mean	**1.00**	**1.00**
	SD	**0.00**	**0.00**
Week 6	Sample 1	2.79	1.00
	Sample 2	3.08	1.00
	Sample 3	2.86	1.00
	Sample 4	2.83	1.00
	Sample 5	2.93	1.00
	Mean	**2.90**	**1.00**
	SD	**0.11**	**0.00**
Week 8	Sample 1	3.85	1.00
	Sample 2	4.00	1.00
	Sample 3	3.07	1.00
	Sample 4	3.99	1.00
	Sample 5	3.76	1.00
	Mean	**3.73**	**1.00**
	SD	**0.38**	**0.00**

Table 3.1c$_{\text{contd.}}$: Growth of APCs in LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	LAE 200
Week 9	Sample 1	3.71	1.00
	Sample 2	3.50	1.00
	Sample 3	3.78	1.30
	Sample 4	4.00	1.00
	Sample 5	3.89	1.00
	Mean	**3.78**	**1.06**
	SD	**0.39**	**0.22**
Week 10	Sample 1	4.01	1.00
	Sample 2	3.87	1.40
	Sample 3	4.43	1.00
	Sample 4	4.53	2.00
	Sample 5	3.22	1.00
	Mean	**4.01**	**1.28**
	SD	**0.52**	**0.40**
Week 11	Sample 1	3.15	1.00
	Sample 2	4.09	1.00
	Sample 3	4.79	1.00
	Sample 4	4.64	1.00
	Sample 5	4.05	1.90
	Mean	**4.14**	**1.18**
	SD	**0.57**	**0.30**
Week 12	Sample 1	4.25	1.45
	Sample 2	4.89	1.40
	Sample 3	4.70	1.50
	Sample 4	4.76	1.55
	Sample 5	4.50	1.10
	Mean	**4.62**	**1.40**
	SD	**1.49**	**0.15**
Week 13	Sample 1	4.98	2.00
	Sample 2	3.60	2.00
	Sample 3	4.56	1.50
	Sample 4	3.22	2.65
	Sample 5	4.20	2.01
	Mean	**4.11**	**2.03**
	SD	**1.80**	**0.40**

Table 3.1c$_{\text{contd.}}$: Growth of APCs in LAE (200 ppm)-treated RTE sliced chicken breast roast

Storage time		Control	LAE 200
Week 14	Sample 1	4.74	1.39
	Sample 2	4.11	4.10
	Sample 3	4.67	3.00
	Sample 4	4.54	2.30
	Sample 5	4.15	2.70
	Mean	**4.44**	**2.70**
	SD	**0.29**	**0.90**
Week 15	Sample 1	4.50	3.50
	Sample 2	4.55	3.50
	Sample 3	4.67	3.50
	Sample 4	4.60	3.10
	Sample 5	4.23	4.00
	Mean	**4.51**	**3.52**
	SD	**0.20**	**0.23**
Week 16	Sample 1	5.11	4.60
	Sample 2	4.98	3.90
	Sample 3	4.56	4.80
	Sample 4	5.21	5.00
	Sample 5	5.20	4.20
	Mean	**5.01**	**4.50**
	SD	**0.89**	**0.34**

Note: Control= no HPP/no LAE; LAE 200= LAE at 200 ppm.

3.2 Raw data for microbial counts (APCs) in the selected treatments

Table 3.2: Growth of APCs in the selected treatments of RTE sliced chicken breast roast.

Storage time		Control	HPP 600/1	HPP 600/5	HPP+LAE 600/1	LAE 200	LAE 315
Day 0	Sample 1	1.00	1.00	1.00	1.00	1.00	1.95
	Sample 2	1.00	1.00	1.00	1.00	1.00	2.98
	Sample 3	1.00	1.00	1.00	1.00	3.25	2.51
	Sample 4	1.00	1.00	1.00	1.00	1.00	1.00
	Sample 5	1.00	1.00	1.00	1.00	2.00	1.00
	Mean	**1.00**	**1.00**	**1.00**	**1.00**	**1.65**	**1.89**
	SD	**0.00**	**0.00**	**0.00**	**0.00**	**0.99**	**0.89**
Week 2	Sample 1	3.08	1.00	1.00	1.00	1.00	3.09
	Sample 2	3.01	1.00	1.00	1.00	1.00	2.87
	Sample 3	2.98	1.00	1.00	1.00	3.39	3.12
	Sample 4	2.73	1.00	1.00	1.00	3.14	2.07
	Sample 5	3.12	1.00	1.00	1.00	3.34	1.00
	Mean	**2.98**	**1.00**	**1.00**	**1.00**	**2.37**	**2.43**
	SD	**0.15**	**0.00**	**0.00**	**0.00**	**1.26**	**0.91**
Week 4	Sample 1	5.39	1.00	1.00	1.00	3.51	3.47
	Sample 2	4.48	1.00	1.00	1.00	3.47	3.49
	Sample 3	4.54	1.00	1.00	1.00	3.52	2.91
	Sample 4	4.47	1.00	1.00	1.00	3.51	3.31
	Sample 5	4.60	1.00	1.00	1.00	3.52	2.74
	Mean	**4.70**	**1.00**	**1.00**	**1.00**	**3.51**	**3.18**
	SD	**0.39**	**0.00**	**0.00**	**0.00**	**0.02**	**0.34**
Week 6	Sample 1	4.67	1.00	1.00	1.00	4.32	4.47
	Sample 2	4.66	1.00	1.00	1.00	4.51	4.21
	Sample 3	4.79	1.00	1.00	1.00	4.39	3.95
	Sample 4	4.86	1.00	1.00	1.00	4.57	4.31
	Sample 5	4.72	1.00	1.00	1.00	4.52	4.47
	Mean	**4.74**	**1.00**	**1.00**	**1.00**	**4.46**	**4.28**
	SD	**0.08**	**0.00**	**0.00**	**0.00**	**0.10**	**0.22**
Week 8	Sample 1	5.54	1.00	1.00	1.00	5.15	4.72
	Sample 2	5.01	1.00	1.00	1.00	5.47	4.6
	Sample 3	5.47	1.00	1.00	1.00	1.00	4.49
	Sample 4	4.53	1.00	1.00	1.00	4.67	4.62
	Sample 5	3.81	1.00	1.00	1.00	4.65	4.27
	Mean	**4.87**	**1.00**	**1.00**	**1.00**	**4.19**	**4.54**
	SD	**0.72**	**0.00**	**0.00**	**0.00**	**1.82**	**0.17**

Table 3.2$_{\text{contd.}}$: Growth of APCs in the selected treatments of RTE sliced chicken breast roast.

Storage time		Control	HPP 600/1	HPP 600/5	HPP+LAE 600/1	LAE 200	LAE 315
Week 10	Sample 1	5.01	1.00	1.00	1.00	4.89	3.66
	Sample 2	4.98	1.00	1.00	1.00	1.00	4.15
	Sample 3	5.06	1.00	1.00	1.00	5.05	5.17
	Sample 4	4.93	1.00	1.00	1.00	5.04	5.11
	Sample 5	5.02	1.00	1.00	1.00	4.99	5.20
	Mean	**5.00**	**1.00**	**1.00**	**1.00**	**4.19**	**4.66**
	SD	**0.05**	**0.00**	**0.00**	**0.00**	**1.79**	**0.71**
Week 12	Sample 1	5.14	1.00	1.00	1.00	7.80	5.16
	Sample 2	6.23	1.00	1.00	1.00	5.38	4.79
	Sample 3	5.09	1.00	2.42	1.00	4.78	4.67
	Sample 4	5.44	1.00	1.00	1.00	5.03	5.04
	Sample 5	5.04	1.00	2.32	1.00	3.90	4.98
	Mean	**5.39**	**1.00**	**1.55**	**1.00**	**5.38**	**4.93**
	SD	**0.50**	**0.00**	**0.75**	**0.00**	**1.46**	**0.20**
Week 14	Sample 1	4.89	1.00	1.10	1.00	6.45	5.16
	Sample 2	5.54	1.00	1.00	1.00	5.17	5.52
	Sample 3	5.90	1.00	1.00	1.00	4.56	4.68
	Sample 4	6.43	1.00	1.00	1.00	6.78	5.10
	Sample 5	4.75	1.00	1.00	1.00	4.25	5.10
	Mean	**5.50**	**1.00**	**1.02**	**1.00**	**5.44**	**5.11**
	SD	**0.60**	**0.00**	**0.10**	**0.00**	**1.12**	**0.33**
Week 16	Sample 1	6.34	1.00	1.00	1.00	5.87	5.23
	Sample 2	5.98	1.00	1.00	1.00	7.23	5.12
	Sample 3	5.31	1.00	1.00	1.00	3.54	4.51
	Sample 4	5.66	1.00	1.00	1.00	5.58	5.67
	Sample 5	5.65	1.00	1.00	1.00	5.80	5.20
	Mean	**5.79**	**1.00**	**1.00**	**1.00**	**5.60**	**5.15**
	SD	**0.43**	**0.00**	**0.00**	**0.00**	**1.20**	**0.45**

Note: Control= no HPP/no LAE; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/5= HPP at 600 MPa for 5 min; HPP+LAE/600/1= HPP + LAE (200 ppm) at 600 MPa for 1 min; LAE 200= LAE at 200 ppm; LAE 315= LAE at 315 ppm.

APPENDIX 4

4.1 Mean data for instrumental color during screening of treatments

Table 4.1a: L* values of the HP-treated RTE sliced chicken breast roast samples during storage at 4°C for 16

Storage time (in weeks)	Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1
0	66.40 ± 2.74	69.80 ± 2.47	72.94 ± 3.66	68.75 ± 1.63	70.27 ± 4.01
2	73.10 ± 2.94	73.23 ± 0.51	74.37 ± 2.20	72.42 ± 1.31	70.60 ± 2.31
4	73.76 ± 1.70	73.37 ± 1.34	72.97 ± 2.33	70.93 ± 0.59	71.34 ± 1.87
6	74.10 ± 1.60	72.55 ± 1.62	72.46 ± 1.80	72.22 ± 2.17	70.90 ± 0.41
8	72.66 ± 3.56	69.58 ± 1.86	72.52 ± 3.06	70.83 ± 0.87	69.73 ± 0.51
10	71.92 ± 0.67	71.39 ± 0.85	72.32 ± 0.71	70.90 ± 2.14	70.41 ± 1.14
12	71.68 ± 0.78	69.40 ± 2.71	71.98 ± 0.40	71.22 ± 0.89	70.57 ± 1.04
14	71.55 ± 0.28	71.08 ± 1.28	72.03 ± 0.17	71.45 ± 0.76	70.79 ± 0.33
16	72.56 ± 1.13	70.67 ± 1.34	72.17 ± 0.86	71.60 ± 1.10	70.84 ± 1.30

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP/450/1= HPP at 450 MPa for 1 min; HPP/450/3.5= HP at 450 MPa for
HPP at 450 MPa for 5 min; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/3.5= HPP at 600 MPa for 3.5
600 MPa for 5 min.

Table 4.1b: L* values of the HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast samples during st
weeks.

Storage time (in weeks)	Control	HPP+LAE/450/1	HPP+LAE/450/3.5	HPP+LAE/450/5	HPP+LAE/600/1	HP
0	66.40 ±2.74	71.33 ± 5.88	69.89 ± 3.65	71.15 ± 4.10	70.27 ± 4.01	
2	73.10 ± 2.94	69.90 ± 1.52	68.70 ± 1.18	71.39 ± 7.33	69.91 ± 1.59	
4	73.76 ± 1.7	71.32 ± 0.99	69.04 ± 1.57	72.18 ± 2.65	69.11 ± 2.04	
6	74.10 ± 1.60	70.87 ± 0.74	68.45 ± 2.88	70.96 ± 3.26	69.83 ± 3.00	
8	72.66 ± 3.56	70.36 ± 1.55	69.17 ± 1.56	70.75 ± 2.06	70.29 ± 2.75	
10	71.92 ± 0.67	70.92 ± 0.43	70.63 ± 0.88	72.29 ± 0.67	70.64 ± 0.94	
12	71.68 ± 0.78	71.28 ± 0.65	70.46 ± 0.99	71.39 ± 0.77	71.11 ± 1.31	
14	71.55 ± 0.28	71.00 ± 1.61	70.62 ± 0.59	71.74 ± 1.37	71.16 ± 1.32	
16	72.56 ± 1.13	71.22 ± 0.86	70.61 ± 0.72	71.70 ± 0.55	71.11 ± 0.67	

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP+LAE/450/1= HPP + LAE (200 ppm) at 450 MPa for 1 min; HPP+LA
(200 ppm) at 450 MPa for 3.5 min; HPP+LAE/450/5= HPP + LAE (200 ppm) at 450 MPa for 5 min; HPP+L
(200 ppm) at 600 MPa for 1 min; HPP+LAE/600/3.5= HPP + LAE (200 ppm) at 600 MPa for 3.5 min; HPP+
(200 ppm) at 600 MPa for 5 min.

Table 4.1c: L* values of LAE (200 ppm)-treated RTE sliced chicken breast roast samples during storage at 4

Storage time (in weeks)	Control	LAE 200
0	66.40 ± 2.74	70.11 ± 1.30
2	73.10 ± 2.94	72.12 ± 1.40
4	73.76 ± 1.70	73.40 ± 1.50
6	74.10 ± 1.60	72.30 ± 0.97
8	72.66 ± 3.56	71.98 ± 1.23
10	71.92 ± 0.67	71.66 ± 1.02
12	71.68 ± 0.78	72.32 ± 0.50
14	71.55 ± 0.28	71.45 ± 0.65
16	72.56 ± 1.13	71.77 ± 1.02

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; LAE 200= LAE at 200 ppm.

Table 4.1d: a* values of the HP-treated RTE sliced chicken breast roast samples during storage at 4°C for 16

Storage time (in weeks)	Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1
0	2.37 ± 1.65	1.34 ± 0.39	0.70 ± 0.47	1.08 ± 0.92	0.35 ± 0.94
2	1.50 ± 0.26	0.64 ± 0.40	0.69 ± 0.54	0.95 ± 0.42	0.28 ± 0.50
4	1.41 ± 0.40	0.85 ± 0.28	0.68 ± 0.35	1.06 ± 0.87	0.83 ± 0.19
6	1.37 ± 0.43	1.00 ± 0.40	0.73 ± 0.28	1.07 ± 0.76	0.86 ± 0.12
8	1.27 ± 0.90	0.37 ± 1.19	0.32 ± 0.31	1.05 ± 0.21	0.99 ± 0.12
10	1.39 ± 0.15	1.10 ± 0.12	0.70 ± 0.28	1.10 ± 0.03	0.95 ± 0.18
12	1.29 ± 0.16	0.24 ± 0.45	0.55 ± 0.21	0.97 ± 0.11	1.01 ± 0.10
14	1.40 ± 0.17	0.65 ± 1.07	0.75 ± 0.52	1.22 ± 0.20	0.95 ± 0.26
16	1.30 ± 0.20	0.85 ± 0.60	0.62 ± 0.43	0.96 ± 0.14	0.93 ± 0.33

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP/450/1= HPP at 450 MPa for 1 min; HPP/450/3.5= HP at 450 MPa for HPP at 450 MPa for 5 min; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/3.5= HPP at 600 MPa for 3.5 600 MPa for 5 min.

Table 4.1e: a* values of HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast samples during storag

Storage time (in weeks)	Control	HPP+LAE/450/1	HPP+LAE/450/3.5	HPP+LAE/450/5	HPP+LAE/600/1	HP
0	2.37 ± 1.65	1.29 ± 0.22	1.39 ± 0.91	0.65 ± 1.14	1.35 ± 0.94	
2	1.50 ± 0.26	0.96 ± 0.83	0.14 ± 0.35	0.82 ± 0.62	1.78 ± 0.99	
4	1.41 ± 0.40	1.43 ± 0.61	0.33 ± 0.59	1.06 ± 0.53	1.55 ± 0.90	
6	1.37 ± 0.43	1.41 ± 0.51	0.43 ± 0.63	1.31 ± 0.52	1.43 ± 1.03	
8	1.27 ± 0.90	0.09 ± 0.12	0.44 ± 0.25	0.99 ± 0.35	-0.01 ± 0.31	
10	1.39 ± 0.15	0.23 ± 0.18	0.66 ± 0.05	1.05 ± 0.24	0.44 ± 0.13	
12	1.29 ± 0.16	0.17 ± 0.07	0.53 ± 0.27	1.00 ± 0.37	0.05 ± 0.24	
14	1.40 ± 0.17	0.40 ± 0.24	0.48 ± 0.48	0.89 ± 0.23	0.18 ± 0.86	
16	1.30 ± 0.20	0.32 ± 0.67	0.45 ± 0.52	0.91 ± 0.34	0.12 ± 0.22	

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP+LAE/450/1= HPP + LAE (200 ppm) at 450 MPa for 1 min; HPP+LA (200 ppm) at 450 MPa for 3.5 min; HPP+LAE/450/5= HPP + LAE (200 ppm) at 450 MPa for 5 min; HPP+L (200 ppm) at 600 MPa for 1 min; HPP+LAE/600/3.5= HPP + LAE (200 ppm) at 600 MPa for 3.5 min; HPP+ (200 ppm) at 600 MPa for 5 min.

Table 4.1f: a* values of the LAE (200 ppm)-treated sliced chicken breast roast samples during storage at 4°C

Storage time (in weeks)	Control	LAE
0	2.37 ± 1.65	2.20 ± 0.75
2	1.50 ± 0.26	1.70 ± 0.43
4	1.41 ± 0.40	1.50 ± 0.54
6	1.37 ± 0.43	1.64 ± 0.65
8	1.27 ± 0.90	1.32 ± 1.02
10	1.39 ± 0.15	1.35 ± 0.69
12	1.29 ± 0.16	1.33 ± 0.74
14	1.40 ± 0.17	1.44 ± 0.77
16	1.30 ± 0.20	1.29 ± 0.89

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; LAE 200= LAE at 200 ppm.

Table 4.1g: b* values of the HP-treated RTE sliced chicken breast roast samples during storage at 4°C for 16

Storage time (in weeks)	Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1
0	5.99 ± 0.71	6.07 ± 0.68	6.14 ± 0.87	5.88 ± 0.36	5.83 ± 1.44
2	7.39 ± 0.43	7.06 ± 0.21	7.56 ± 0.72	7.11 ± 0.34	6.83 ± 0.40
4	6.76 ± 0.83	6.87 ± 0.55	6.33 ± 0.45	6.70 ± 0.45	7.06 ± 0.25
6	6.71 ± 0.68	7.04 ± 0.33	6.98 ± 0.85	6.60 ± 0.89	7.10 ± 0.17
8	6.91 ± 0.94	6.42 ± 0.71	6.63 ± 0.58	6.72 ± 0.52	6.50 ± 0.32
10	6.29 ± 0.50	6.00 ± 0.16	6.70 ± 0.56	7.04 ± 0.17	7.40 ± 0.15
12	6.01 ± 0.34	6.14 ± 0.64	6.80 ± 0.50	7.03 ± 0.56	6.90 ± 0.51
14	6.47 ± 0.35	7.28 ± 0.46	6.90 ± 0.39	7.21 ± 0.38	7.40 ± 0.24
16	6.33 ± 0.41	6.55 ± 0.76	6.75 ± 0.45	7.15 ± 0.42	7.10 ± 0.32

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP/450/1= HPP at 450 MPa for 1 min; HPP/450/3.5= HP at 450 MPa for HPP at 450 MPa for 5 min; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/3.5= HPP at 600 MPa for 3.5 600 MPa for 5 min.

Table 4.1h: b* values of HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast samples during storag

rage time (in weeks)	Control	HPP+LAE/450/1	HPP+LAE/450/3.5	HPP+LAE/450/5	HPP+LAE/600/1
0	5.99 ± 0.71	6.39 ± 1.15	5.44 ± 0.68	6.30 ± 1.56	5.83 ± 1.44
2	7.39 ± 0.43	5.17 ± 0.50	5.38 ± 0.78	5.89 ± 1.10	6.44 ± 0.57
4	6.76 ± 0.83	6.26 ± 0.42	5.17 ± 0.34	5.88 ± 0.56	6.59 ± 0.58
6	6.71 ± 0.68	6.34 ± 0.69	5.57 ± 0.94	6.07 ± 0.65	7.01 ± 0.86
8	6.91 ± 0.94	6.34 ± 0.23	6.01 ± 0.61	6.55 ± 0.48	6.75 ± 0.24
10	6.29 ± 0.50	6.67 ± 0.38	5.83 ± 0.34	7.04 ± 0.06	7.03 ± 0.14
12	6.01 ± 0.34	6.91 ± 0.53	7.16 ± 0.42	6.71 ± 0.45	7.03 ± 0.04
14	6.47 ± 0.35	7.06 ± 0.33	7.49 ± 0.27	7.08 ± 0.16	7.29 ± 0.57
16	6.33 ± 0.41	6.87 ± 0.54	7.12 ± 0.31	7.00 ± 0.41	7.13 ± 0.61

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP+LAE/450/1= HPP + LAE (200 ppm) at 450 MPa for 1 min; HPP+LA (200 ppm) at 450 MPa for 3.5 min; HPP+LAE/450/5= HPP + LAE (200 ppm) at 450 MPa for 5 min; HPP+L (200 ppm) at 600 MPa for 1 min; HPP+LAE/600/3.5= HPP + LAE (200 ppm) at 600 MPa for 3.5 min; HPP+ (200 ppm) at 600 MPa for 5 min.

Table 4.1i: b* values of LAE (200 ppm)-treated RTE sliced chicken breast roast samples during storage at 4°

Storage time (in weeks)	Control	LAE 200
0	5.99 ± 0.71	6.00 ± 1.10
2	7.39 ± 0.43	6.57 ± 0.54
4	6.76 ± 0.83	6.77 ± 0.98
6	6.71 ± 0.68	6.59 ±0.71
8	6.91 ± 0.94	6.89 ± 0.74
10	6.29 ± 0.50	6.40 ± 0.76
12	6.01 ± 0.34	6.12 ± 0.86
14	6.47 ± 0.35	6.55 ± 0.32
16	6.33 ± 0.41	6.43 ± 0.67

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; LAE 200= LAE at 200 ppm.

4.2 Mean data for instrumental color of the selected treatments

Table 4.2a: L* values of the selected treatments of RTE sliced chicken breast roast samples during storage at

Storage time (in weeks)	Control	HPP/600/1	HPP/600/5	HPP+LAE600/1	LAE 2
0	74.09 ± 1.55	71.17 ± 0.22	72.65 ± 1.22	71.11 ± 0.20	72.47 ±
2	74.10 ± 3.21	70.08 ± 0.62	72.17 ± 1.89	70.94 ± 0.44	72.10 ±
4	74.62 ± 4.41	70.73 ± 0.32	73.44 ± 2.08	72.15 ± 3.51	70.46 ±
6	72.43 ± 6.21	69.50 ± 0.79	72.81 ± 3.41	72.29 ± 1.81	72.42 ±
8	71.65 ± 0.97	73.41 ± 3.11	70.05 ± 0.47	70.77 ± 0.39	74.09 ±
10	73.38 ± 2.12	72.86 ± 2.34	71.54 ± 0.78	71.47 ± 0.56	75.58 ±
12	69.57 ± 1.53	70.62 ± 1.44	72.89 ± 2.39	68.82 ± 2.24	71.61 ±
14	65.85 ± 1.47	69.53 ± 1.27	70.72 ± 1.93	74.53 ± 1.27	70.64 ±
16	66.79 ± 1.87	71.44 ± 0.75	71.15 ± 0.07	71.91 ± 0.63	70.44 ±

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/5= HPP at 600 MPa for 5
HPP + LAE (200 ppm) at 600 MPa for 1 min; LAE 200= LAE at 200 ppm; LAE 315= LAE at 315 ppm.

Table 4.2b: a* values of the selected treatments of RTE sliced chicken breast roast samples during storage at

Storage time (in weeks)	Control	HPP/600/1	HPP 600/5	HPP+LAE600/1
0	2.06 ± 0.07	0.52 ± 0.17	0.96 ± 0.06	1.21 ± 0.11
2	2.22 ± 0.06	0.62 ± 0.11	0.85 ± 0.14	1.24 ± 0.17
4	2.16 ± 0.15	0.54 ± 0.29	0.82 ± 0.09	1.75 ± 0.02
6	1.85 ± 0.76	0.74 ± 0.86	1.04 ± 0.18	1.22 ± 0.11
8	2.44 ± 0.19	1.98 ± 0.48	2.33 ± 0.11	2.30 ± 0.39
10	2.37 ± 0.23	2.21 ± 0.29	2.48 ± 0.14	2.41 ±0.26
12	3.29 ± 0.44	3.11 ±0.25	1.79 ± 0.09	2.22 ± 0.26
14	2.45 ± 0.21	2.38 ± 0.09	1.40 ± 0.18	1.85 ±0.34
16	2.75 ± 0.40	2.30 ± 0.17	1.33 ± 0.13	1.73 ±0.35

*All values are means ± standard deviation

Note: Control= no HPP/no LAE; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/5= HPP at 600 MPa for
HPP + LAE (200 ppm) at 600 MPa for 1 min; LAE 200= LAE at 200 ppm; LAE 315= LAE at 315 ppm.

APPENDIX 5

5.1 Mean data for instrumental texture during screening of treatments

Table 5.1: Shear force* values of the HP-treated RTE sliced chicken breast roast samples during storage at 4

Storage time (in weeks)	Control	HPP/450/1	HPP/450/3.5	HPP/450/5	HPP/600/1	HPP
0	53.12 ± 0.64	52.34 ± 1.20	53.55 ± 1.03	53.98 ± 0.76	53.26 ± 0.34	54.3
2	52.91 ± 0.50	52.03 ± 1.47	53.12 ± 0.77	54.50 ± 1.52	53.63 ± 1.11	53.7
4	54.05 ± 1.41	53.52 ± 0.88	53.88 ± 0.58	54.01 ± 1.46	52.74 ± 1.33	54.6
6	52.16 ± 1.17	52.83 ± 0.99	54.05 ± 0.55	54.46 ± 1.01	52.77 ± 0.39	51.6
8	51.76 ± 0.65	53.12 ± 1.21	53.93 ± 1.56	51.30 ± 1.23	53.63 ± 1.33	53.0
10	50.35 ± 0.75	52.25 ± 1.39	54.63 ± 1.09	52.73 ± 1.38	53.77 ± 2.17	54.0
12	50.50 ± 0.68	51.95 ± 0.98	53.41 ± 1.49	50.84 ± 0.48	53.65 ± 0.87	54.0
14	50.70 ± 1.25	51.60 ± 1.28	53.84 ± 0.46	51.46 ± 1.44	52.93 ± 1.44	53.3
16	50.23 ± 1.04	51.30 ± 0.86	53.22 ± 0.87	51.50 ± 1.03	51.86 ± 0.92	52.4

*All values represent mean shear force (g) ± standard deviation of samples. Each value is the mean of six rea

Note: Control= no HPP/no LAE; HPP/450/1= HPP at 450 MPa for 1 min; HPP/450/3.5= HPP at 450 MPa fo HPP at 450 MPa for 5 min; HPP/600/1= HPP at 600 MPa for 1 min; HPP/600/3.5= HPP at 600 MPa for 3.5 600 MPa for 5 min.

Table 5.2: Shear force* values of the HPP+LAE (200 ppm)-treated RTE sliced chicken breast roast samples
16 weeks.

Storage time (in weeks)	Control	HPP + LAE/450/1	HPP +LAE/450/3.5	HPP+LAE/450/5	HPP+LAE/600/1	HPP
0	53.12 ± 0.64	53.40 ± 0.54	53.12 ± 2.10	52.78 ± 0.93	53.46 ± 0.87	5
2	52.91 ± 0.50	53.28 ± 0.82	51.05 ± 3.51	53.63 ± 1.22	53.39 ± 0.71	5
4	54.05 ± 1.41	53.88 ± 0.79	52.00 ± 1.46	53.06 ± 0.99	53.04 ± 0.92	5
6	52.16 ± 1.17	53.25 ± 0.20	50.92 ± 0.71	53.28 ± 0.65	52.85 ± 0.48	5
8	51.76 ± 0.65	53.80 ± 0.60	53.96 ± 1.11	52.46 ± 0.62	52.44 ± 0.46	5
10	50.35 ± 0.75	52.30 ± 1.97	53.17 ± 1.67	50.50 ± 0.75	51.40 ± 0.57	5
12	50.50 ± 0.68	52.80 ± 1.74	53.35 ± 1.49	53.98 ± 2.10	52.66 ± 1.36	50
14	50.70 ± 1.25	53.71 ±2.67	52.23 ± 1.11	51.85 ± 1.52	53.26 ± 1.65	5
16	50.23 ± 1.04	53.61 ± 1.13	52.10 ± 0.81	51.66 ± 0.78	52.45 ± 0.92	5

*All values represent mean shear force (g) ± standard deviation of samples. Each value is the mean of six rea

Note: Control= no HPP/no LAE; HPP+LAE/450/1= HPP + LAE (200 ppm) at 450 MPa for 1 min; HPP+LA
(200 ppm) at 450 MPa for 2.5 min; HPP+LAE /450/5= HPP + LAE (200 ppm) at 450 MPa for 5 min; HPP+
LAE (200 ppm) at 600 MPa for 1 min; HPP+LAE /600/3.5= HPP + LAE (200 ppm) at 600 MPa for 3.5 min
+ LAE (200 ppm) at 600 MPa for 5 min.

Table 5.3: Shear force* values of the LAE (200 ppm)-treated RTE sliced chicken breast roast samples during weeks.

Storage time (in weeks)	Control	LAE 200
0	53.12 ± 0.64	53.55 ± 0.75
2	52.91 ± 0.50	53.16 ± 0.67
4	54.05 ± 1.41	53.67 ± 0.14
6	52.16 ± 1.17	52.78 ± 0.71
8	51.76 ± 0.65	52.81 ± 0.56
10	50.35 ± 0.75	51.33 ± 0.77
12	50.50 ± 0.68	52.65 ± 0.84
14	50.70 ± 1.25	52.74 ± 0.52
16	50.23 ± 1.04	52.97 ± 0.94

*All values represent mean shear force (g) ± standard deviation of samples. Each value is the mean of six rea

Note: Control= no HPP/no LAE; LAE 200= LAE at 200 ppm.

APPENDIX 6

6.1 Consumer Consent Form

SENSORY PROPERTIES OF READY-TO-EAT CHICKEN BREAST ROAST

Consumer Sensory Evaluation

CONSENT FORM

THIS CONSENT FORM WILL BE HELD FOR 12 MONTHS FROM DATE OF SIGNING

(For minors aged 8-15 consent form to be signed by parent or guardian)

- I have read and understood the Information Sheet and have had the details of the study explained to me. My questions have been answered to my satisfaction, and I understand that I may ask further questions at any time.

- I agree to voluntarily participate in this study under the conditions set out in the Information Sheet.

- I understand I have the right to withdraw from the study at any time and to decline to answer any particular questions.

- I understand that the product does not comply with Halal or Kosher standards.

Participants Signature:……………………………………………………………………………..

Full Name- printed:…………………………..…………………Date:…………………………….

6.2 Sensory Evaluation Form

CONSUMER ACCEPTABILITY TEST

- Please rinse your mouth before starting.
- Evaluate the sample ____ in front of you for appearance and other attributes.

1. Indicate your overall opinion on the COLOR of the sample by checking ONLY one box [√]

☐	☐	☐	☐	☐	☐	☐	☐	☐
Dislike Extremely				Neither like nor dislike				Like extremely

2. Indicate your overall opinion on the TEXTURE of the sample by checking ONLY one box [√]

☐	☐	☐	☐	☐	☐	☐	☐	☐
Dislike Extremely				Neither like nor dislike				Like extremely

3. Indicate your overall opinion on the FLAVOUR of the sample by checking ONLY one box [√]

☐	☐	☐	☐	☐	☐	☐	☐	☐
Dislike Extremely				Neither like nor dislike				Like extremely

4. Indicate your overall opinion on the FRESHNESS of the sample by checking ONLY one box [√]

☐	☐	☐	☐	☐	☐	☐	☐	☐
Extremely stale				Neither like nor dislike				Extremely fresh

5. Indicate your overall degree of liking of the product by checking ONLY one box [√]

☐	☐	☐	☐	☐	☐	☐	☐	☐
Dislike Extremely				Neither like nor dislike				Like extremely

THANK YOU FOR YOUR PARTICIPATION

APPENDIX 7

7.1 Consumer sensory results for the selected treatments

Table 7.1: Consumer sensory evaluation of selected treatments at week 2

Panelist	Sample	Color	Texture	Flavor	Freshness	Overall
1	799	8	8	5	7	6
2	799	6	6	8	7	7
3	799	6	6	5	6	4
4	799	6	8	8	8	8
5	799	6	6	7	9	7
6	799	7	7	6	8	7
7	799	9	9	9	9	9
8	799	6	7	7	7	7
9	799	9	9	9	9	9
10	799	2	4	4	4	3
11	799	6	2	5	6	5
12	799	8	8	8	5	8
13	799	7	6	7	7	6
14	799	6	5	5	6	5
15	799	5	6	8	6	7
16	799	6	7	7	7	7
17	799	5	6	6	6	6
18	799	7	7	7	6	7
19	799	5	6	6	5	5
20	799	3	7	8	7	6
21	799	5	6	6	6	7
22	799	4	7	6	5	6
23	799	4	5	7	3	7
	Mean	5.91	6.43	6.70	6.48	6.48
	SD	1.73	1.56	1.36	1.53	1.44
1	126	6	8	7	9	8
2	126	6	7	7	8	7
3	126	5	4	3	4	4
4	126	4	8	8	8	7
5	126	7	7	8	9	8
6	126	8	7	5	7	5
7	126	9	9	9	9	9
8	126	6	7	5	7	6
9	126	9	9	9	9	9

Table 7.1$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 2

Panelist	Sample	Color	Texture	Flavor	Freshness	Overall
10	126	3	4	4	4	4
11	126	5	4	4	6	4
12	126	7	6	6	5	6
13	126	6	6	7	7	7
14	126	6	7	7	8	7
15	126	5	4	5	5	5
16	126	5	6	5	6	6
17	126	6	7	6	7	7
18	126	7	6	7	6	6
19	126	5	7	6	6	6
20	126	3	8	8	8	7
21	126	5	5	6	5	6
22	126	4	6	7	5	6
23	126	2	3	3	3	2
	Mean	5.61	6.30	6.173913	6.57	6.17
	SD	1.77	1.66	1.722899	1.78	1.67
1	853	7	8	6	8	7
2	853	7	8	5	5	6
3	853	6	6	4	5	6
4	853	2	4	7	5	4
5	853	7	7	7	9	8
6	853	6	7	7	7	7
7	853	7	9	8	8	8
8	853	6	7	8	7	8
9	853	9	9	9	9	9
10	853	3	4	4	5	4
11	853	3	2	1	6	2
12	853	8	8	4	5	4
13	853	6	7	6	7	7
14	853	7	8	7	7	7
15	853	6	6	7	3	7
16	853	7	6	4	5	5
17	853	6	7	6	7	7
18	853	7	7	7	7	7
19	853	5	6	7	6	6
20	853	3	7	8	7	6
21	853	5	5	6	5	6
22	853	4	4	6	4	4

Table 7.1$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 2

Panelist	Sample	Color	Texture	Flavor	Freshness	Overall
23	853	4	4	7	6	4
	Mean	5.70	6.35	6.13	6.22	6.04
	SD	1.79	1.80	1.79	1.54	1.72
1	342	7	7	8	8	7
2	342	7	8	7	6	7
3	342	6	6	6	6	6
4	342	7	6	6	7	5
5	342	7	7	7	9	8
6	342	4	4	4	6	4
7	342	7	8	8	8	9
8	342	6	7	7	7	7
9	342	9	9	8	9	9
10	342	4	5	3	3	3
11	342	7	3	4	7	4
12	342	8	8	8	5	8
13	342	7	7	6	6	7
14	342	8	8	9	8	8
15	342	3	5	7	5	5
16	342	7	6	6	6	6
17	342	6	6	5	6	6
18	342	7	7	7	7	7
19	342	7	6	6	6	6
20	342	4	7	7	7	7
21	342	4	5	6	5	7
22	342	5	7	6	5	6
23	342	4	4	3	4	3
	Mean	6.13	6.35	6.26	6.35	6.30
	SD	1.60	1.50	1.60	1.50	1.69
1	561	9	8	8	7	8
2	561	5	6	5	5	5
3	561	4	4	3	6	5
4	561	7	8	8	8	8
5	561	6	8	9	9	9
6	561	6	5	6	6	5
7	561	8	8	7	7	7
8	561	7	8	7	7	8
9	561	9	9	8	9	9
10	561	2	4	5	4	3

Table 7.1$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 2

Panelist	Sample	Color	Texture	Flavor	Freshness	Overall
11	561	4	3	7	7	5
12	561	8	8	3	5	4
13	561	3	7	6	6	6
14	561	7	7	7	6	6
15	561	5	6	7	5	6
16	561	6	7	6	6	6
17	561	6	4	5	4	5
18	561	7	7	6	7	7
19	561	4	5	5	4	4
20	561	4	6	8	7	6
21	561	1	4	3	3	3
22	561	4	4	4	5	4
23	561	4	4	7	6	5
	Mean	5.48	6.09	6.09	6.04	5.83
	SD	2.13	1.81	1.73	1.55	1.75

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.2: Consumer sensory evaluation of selected treatments at week 4

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	5	7	8	8	8
2	799	4	4	6	5	5
3	799	4	6	7	7	5
4	799	6	5	6	6	5
5	799	5	8	7	3	4
6	799	6	7	7	7	7
7	799	7	6	7	6	7
8	799	7	6	7	9	7
9	799	8	7	8	7	7
10	799	5	7	8	8	8
12	799	4	4	6	5	5
13	799	4	6	7	7	5
14	799	6	5	6	6	5
15	799	5	8	7	3	4
16	799	6	7	7	7	7
17	799	7	6	7	6	7
18	799	7	6	7	9	7
19	799	8	7	8	7	7
	Mean	5.78	6.22	7.00	6.44	6.11
	SD	1.39	1.20	0.71	1.74	1.36
1	126	6	6	6	6	6
2	126	7	7	7	6	7
3	126	6	7	6	6	6
4	126	3	2	3	3	3
5	126	3	6	4	6	6
6	126	7	6	6	6	6
7	126	7	7	6	7	7
8	126	8	7	6	6	6
9	126	7	8	7	6	8
11	126	6	6	6	6	6
12	126	7	7	7	6	7
13	126	6	7	6	6	6
14	126	3	2	3	3	3
15	126	3	6	4	6	6
16	126	7	6	6	6	6
17	126	7	7	6	7	7
18	126	8	7	6	6	6

Table 7.2$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 4

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
19	126	7	8	7	6	8
	Mean	6.00	6.22	5.67	5.78	6.11
	SD	1.80	1.72	1.32	1.09	1.36
1	853	5	6	5	6	5
2	853	4	4	4	5	4
3	853	7	7	7	7	7
4	853	5	7	8	9	7
5	853	3	6	3	3	6
6	853	7	5	6	6	6
7	853	7	7	7	7	7
8	853	4	7	8	7	8
9	853	8	7	8	8	8
11	853	5	6	5	6	5
12	853	4	4	4	5	4
13	853	7	7	7	7	7
14	853	5	7	8	9	7
15	853	3	6	3	3	6
16	853	7	5	6	6	6
17	853	7	7	7	7	7
18	853	4	7	8	7	8
19	853	8	7	8	8	8
	Mean	5.56	6.22	6.22	6.44	6.44
	SD	1.74	1.09	1.86	1.74	1.33
1	342	5	6	6	6	6
2	342	4	4	5	5	4
3	342	7	7	7	7	7
4	342	3	5	6	5	5
5	342	5	4	6	4	6
6	342	7	7	6	6	6
7	342	7	7	7	7	7
8	342	5	8	9	9	8
9	342	8	8	7	8	7
11	342	5	6	6	6	6
12	342	4	4	5	5	4
13	342	7	7	7	7	7
14	342	3	5	6	5	5
15	342	5	4	6	4	6

Table 7.2$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 4

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
16	342	7	7	6	6	6
17	342	7	7	7	7	7
18	342	5	8	9	9	8
19	342	8	8	7	8	7
	Mean	5.67	6.22	6.56	6.33	6.22
	SD	1.66	1.56	1.13	1.58	1.20
1	561	4	5	4	6	6
2	561	7	7	6	5	7
3	561	6	6	7	6	6
4	561	8	3	3	3	3
5	561	4	4	3	7	4
6	561	7	6	8	7	8
7	561	6	6	6	6	6
8	561	8	9	8	8	8
9	561	7	8	7	7	8
11	342	5	6	6	6	6
12	342	4	4	5	5	4
13	342	7	7	7	7	7
14	342	3	5	6	5	5
15	342	5	4	6	4	6
16	342	7	7	6	6	6
17	342	7	7	7	7	7
18	342	5	8	9	9	8
19	342	8	8	7	8	7
	Mean	6.33	6.00	5.78	6.11	6.22
	SD	1.50	1.87	1.99	1.45	1.79

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.3: Consumer sensory evaluation of selected treatments at week 6

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	7	7	6	8	7
2	799	4	4	4	4	4
3	799	5	4	5	5	6
4	799	5	7	8	7	7
5	799	6	5	6	4	4
6	799	3	6	8	7	7
7	799	7	7	8	8	7
8	799	6	7	6	6	7
9	799	6	3	4	3	3
10	799	6	6	7	7	6
11	799	6	6	5	5	6
12	799	7	8	8	8	9
13	799	6	6	6	7	6
14	799	5	7	8	7	7
15	799	1	1	3	6	1
16	799	8	8	8	7	8
17	799	7	3	5	8	7
18	799	7	4	7	5	6
19	799	6	7	7	7	7
20	799	7	7	5	7	6
21	799	7	7	6	5	6
22	799	3	7	5	4	5
23	799	7	6	6	6	7
	Mean	5.74	5.78	6.13	6.13	6.04
	SD	1.66	1.81	1.49	1.49	1.72
1	126	7	8	8	9	8
2	126	7	7	7	7	7
3	126	5	6	7	5	6
4	126	5	7	6	6	6
5	126	4	3	6	5	6
6	126	4	6	7	7	6
7	126	9	6	8	8	8
8	126	7	7	6	7	6
9	126	7	6	4	5	5
10	126	7	7	7	7	7
11	126	6	7	7	6	7
12	126	3	8	5	5	6

Table 7.3$_{contd.}$: Consumer sensory evaluation of selected treatments at week 6

Panellist	Attribute	Color	Texture	Flavour	Freshness	Overall
13	126	5	5	5	7	5
14	126	5	2	6	6	5
15	126	3	1	2	3	1
16	126	7	8	8	8	8
17	126	7	9	7	5	7
18	126	7	6	5	6	6
19	126	7	7	6	7	6
20	126	6	6	7	7	7
21	126	4	6	6	5	5
22	126	5	3	3	3	4
23	126	8	7	6	6	6
	Mean	5.87	6.00	6.04	6.09	6.00
	SD	1.60	2.00	1.52	1.47	1.51
1	853	5	6	7	9	7
2	853	7	5	6	5	6
3	853	5	5	5	5	6
4	853	5	4	5	6	6
5	853	6	6	4	5	4
6	853	7	7	7	8	7
7	853	9	6	4	6	6
8	853	7	3	6	6	4
9	853	7	4	3	3	4
10	853	7	7	6	7	7
11	853	5	5	4	5	5
12	853	5	8	8	6	7
13	853	5	5	5	5	6
14	853	6	5	7	6	6
15	853	1	1	4	1	1
16	853	7	7	8	7	8
17	853	4	5	3	7	3
18	853	7	6	7	6	7
19	853	7	7	6	7	6
20	853	8	7	7	7	7
21	853	4	5	4	5	4
22	853	2	3	3	3	3
23	853	6	8	6	7	7
	Mean	5.74	5.43	5.43	5.74	5.52
	SD	1.84	1.70	1.59	1.74	1.73

Table 7.3$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 6

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
1	342	6	7	4	9	5
2	342	4	5	6	6	5
3	342	5	9	6	5	6
4	342	5	7	5	7	5
5	342	6	5	7	5	7
6	342	7	6	6	7	7
7	342	7	8	8	8	8
8	342	5	4	6	6	5
9	342	6	6	7	7	7
10	342	6	7	6	7	6
11	342	3	4	4	5	4
12	342	8	5	3	7	4
13	342	3	5	4	3	4
14	342	3	5	1	5	1
15	342	8	8	9	9	9
16	342	8	7	6	7	7
17	342	7	7	2	5	1
18	342	6	5	4	4	4
19	342	7	5	6	6	6
20	342	7	6	4	3	3
21	342	7	7	7	5	7
22	342	3	3	4	4	4
23	342	6	6	6	5	5
	Mean	5.78	5.96	5.26	5.87	5.22
	SD	1.65	1.46	1.89	1.66	2.00
1	561	7	8	8	9	8
2	561	7	6	7	6	7
3	561	6	6	5	5	5
4	561	5	6	4	6	4
5	561	5	3	5	5	5
6	561	4	4	5	6	5
7	561	6	4	8	7	7
8	561	6	6	7	5	6
9	561	6	9	7	5	7
10	561	6	6	5	6	5
11	561	5	5	6	5	5
12	561	7	7	8	6	9
13	561	5	5	7	6	6

Table 7.3$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 6

Panellist	Attribute	Color	Texture	Flavour	Freshness	Overall
14	561	3	6	7	5	5
15	561	5	4	4	6	3
16	561	7	7	7	7	8
17	561	4	3	5	7	5
18	561	6	6	6	6	6
19	561	6	7	7	7	7
20	561	5	4	6	6	5
21	561	8	7	7	6	7
22	561	3	5	6	8	7
23	561	5	6	3	6	4
	Mean	5.52	5.65	6.09	6.13	5.91
	SD	1.27	1.53	1.38	1.01	1.47

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.4: Consumer sensory evaluation of selected treatments at week 8

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	2	3	3	3	2
2	799	3	4	6	8	3
3	799	6	8	7	6	6
4	799	5	7	7	7	7
5	799	6	6	7	7	6
6	799	7	6	3	5	5
7	799	7	7	6	7	7
8	799	5	5	7	4	6
9	799	8	8	8	8	8
10	799	5	6	7	7	6
11	799	7	7	7	7	8
12	799	7	7	7	7	7
13	799	6	6	5	6	6
14	799	2	3	8	4	6
15	799	2	3	3	3	2
16	799	3	4	6	8	3
17	799	6	8	7	6	6
18	799	5	7	7	7	7
19	799	6	6	7	7	6
20	799	7	6	3	5	5
21	799	7	7	6	7	7
22	799	5	5	7	4	6
23	799	8	8	8	8	8
24	799	5	6	7	7	6
25	799	7	7	7	7	8
26	799	7	7	7	7	7
27	799	6	6	5	6	6
28	799	2	3	8	4	6
	Mean	5.43	5.93	6.29	6.14	5.93
	SD	1.91	1.64	1.59	1.56	1.69
1	126	2	3	3	4	3
2	126	2	3	3	6	2
3	126	8	8	8	7	8
4	126	4	6	6	7	6
5	126	6	6	7	7	6
6	126	1	8	2	7	4
7	126	6	7	7	7	7

Table 7.4$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 8

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
8	126	2	3	7	5	4
9	126	4	5	6	5	4
10	126	4	6	7	7	7
11	126	7	7	6	7	7
12	126	4	6	5	6	5
13	126	5	6	6	5	5
14	126	2	6	4	5	5
15	126	2	3	3	4	3
16	126	2	3	3	6	2
17	126	8	8	8	7	8
18	126	4	6	6	7	6
19	126	6	6	7	7	6
20	126	1	8	2	7	4
21	126	6	7	7	7	7
22	126	2	3	7	5	4
23	126	4	5	6	5	4
24	126	4	6	7	7	7
25	126	7	7	6	7	7
26	126	4	6	5	6	5
27	126	5	6	6	5	5
28	126	2	6	4	5	5
	Mean	4.07	5.71	5.50	6.07	5.21
	SD	2.13	1.68	1.83	1.07	1.72
1	853	2	3	6	5	5
2	853	3	5	6	6	4
3	853	7	5	8	6	7
4	853	8	8	8	7	8
5	853	7	7	7	7	7
6	853	7	8	8	7	8
7	853	6	7	7	6	6
8	853	8	8	6	7	7
9	853	6	1	3	5	3
10	853	5	5	7	7	7
11	853	6	7	6	7	7
12	853	7	6	3	6	4
13	853	6	5	5	6	5
14	853	6	6	7	6	7

Table 7.4$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 8

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
15	853	2	3	6	5	5
16	853	3	5	6	6	4
17	853	7	5	8	6	7
18	853	8	8	8	7	8
19	853	7	7	7	7	7
20	853	7	8	8	7	8
21	853	6	7	7	6	6
22	853	8	8	6	7	7
23	853	6	1	3	5	3
24	853	5	5	7	7	7
25	853	6	7	6	7	7
26	853	7	6	3	6	4
27	853	6	5	5	6	5
28	853	6	6	7	6	7
	Mean	6.00	5.79	6.21	6.29	6.07
	SD	1.71	2.01	1.63	0.73	1.59
1	342	6	6	6	6	6
2	342	4	6	6	7	5
3	342	6	5	5	5	5
4	342	6	6	4	5	5
5	342	7	7	7	6	7
6	342	8	7	7	7	7
7	342	7	7	7	5	5
8	342	5	4	3	5	4
9	342	9	7	8	8	8
10	342	5	6	5	5	5
11	342	6	6	6	7	7
12	342	7	5	3	5	4
13	342	6	6	5	6	5
14	342	4	3	4	3	3
15	342	6	6	6	6	6
16	342	4	6	6	7	5
17	342	6	5	5	5	5
18	342	6	6	4	5	5
19	342	7	7	7	6	7
20	342	8	7	7	7	7
21	342	7	7	7	5	5
22	342	5	4	3	5	4
23	342	9	7	8	8	8

Table 7.4$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 8

Panellist	Sample	Color	Texture	Flavour	Freshness	Overall
24	342	5	6	5	5	5
25	342	6	6	6	7	7
26	342	7	5	3	5	4
27	342	6	6	5	6	5
28	342	4	3	4	3	3
	Mean	6.14	5.79	5.43	5.71	5.43
	SD	1.41	1.19	1.55	1.27	1.40
1	561	1	3	4	5	4
2	561	2	3	3	7	3
3	561	8	8	8	8	8
4	561	5	3	3	4	4
5	561	4	4	4	4	4
6	561	3	7	1	5	1
7	561	6	6	6	4	5
8	561	5	2	3	4	4
9	561	7	7	5	5	6
10	561	6	4	6	4	4
11	561	7	7	7	7	7
12	561	6	4	5	6	5
13	561	5	5	4	5	5
14	561	3	3	3	2	3
15	561	1	3	4	5	4
16	561	2	3	3	7	3
17	561	8	8	8	8	8
18	561	5	3	3	4	4
19	561	4	4	4	4	4
20	561	3	7	1	5	1
21	561	6	6	6	4	5
22	561	5	2	3	4	4
23	561	7	7	5	5	6
24	561	6	4	6	4	4
25	561	7	7	7	7	7
26	561	6	4	5	6	5
27	561	5	5	4	5	5
28	561	3	3	3	2	3
	Mean	4.86	4.71	4.43	5.00	4.50
	SD	2.03	1.94	1.87	1.57	1.74

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.5: Consumer sensory evaluation of selected treatments at week 10

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	6	5	7	5	5
2	799	7	4	6	5	5
3	799	7	8	6	8	7
4	799	4	7	7	8	7
5	799	4	6	6	4	4
6	799	6	5	7	5	5
7	799	7	4	6	5	5
8	799	7	8	6	8	7
9	799	4	7	7	8	7
10	799	4	6	6	4	4
11	799	6	5	7	5	5
12	799	7	4	6	5	5
13	799	7	8	6	8	7
14	799	4	7	7	8	7
15	799	4	6	6	4	4
16	799	6	5	7	5	5
17	799	7	4	6	5	5
18	799	7	8	6	8	7
19	799	4	7	7	8	7
20	799	4	6	6	4	4
	Mean	5.60	6.00	6.40	6.00	5.60
	SD	1.52	1.58	0.55	1.87	1.34
1	126	7	6	4	7	6
2	126	6	7	8	5	5
3	126	6	9	6	8	8
4	126	6	4	8	8	8
5	126	2	5	3	3	4
6	126	7	6	4	7	6
7	126	6	7	8	5	5
8	126	6	9	6	8	8
9	126	6	4	8	8	8
10	126	2	5	3	3	4
11	126	7	6	4	7	6
12	126	6	7	8	5	5
13	126	6	9	6	8	8
14	126	6	4	8	8	8
15	126	2	5	3	3	4

Table 7.5$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 10

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
16	126	7	6	4	7	6
17	126	6	7	8	5	5
18	126	6	9	6	8	8
19	126	6	4	8	8	8
20	126	2	5	3	3	4
	Mean	5.40	6.20	5.80	6.20	6.20
	SD	1.95	1.92	2.28	2.17	1.79
1	853	4	4	4	4	5
2	853	6	7	7	6	6
3	853	7	7	8	7	8
4	853	5	7	6	8	6
5	853	8	8	8	7	7
6	853	4	4	4	4	5
7	853	6	7	7	6	6
8	853	7	7	8	7	8
9	853	5	7	6	8	6
10	853	8	8	8	7	7
11	853	4	4	4	4	5
12	853	6	7	7	6	6
13	853	7	7	8	7	8
14	853	5	7	6	8	6
15	853	8	8	8	7	7
16	853	4	4	4	4	5
17	853	6	7	7	6	6
18	853	7	7	8	7	8
19	853	5	7	6	8	6
20	853	8	8	8	7	7
	Mean	6.00	6.60	6.60	6.40	6.40
	SD	1.58	1.52	1.67	1.52	1.14
1	342	6	2	2	1	3
2	342	4	4	5	4	4
3	342	7	4	5	6	5
4	342	4	7	7	8	8
5	342	3	3	3	1	3
6	342	6	2	2	1	3
7	342	4	4	5	4	4
8	342	7	4	5	6	5
9	342	4	7	7	8	8

Table 7.5$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 10

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
10	342	3	3	3	1	3
11	342	6	2	2	1	3
12	342	4	4	5	4	4
13	342	7	4	5	6	5
14	342	4	7	7	8	8
15	342	3	3	3	1	3
16	342	6	2	2	1	3
17	342	4	4	5	4	4
18	342	7	4	5	6	5
19	342	4	7	7	8	8
20	342	3	3	3	1	3
	Mean	4.80	4.00	4.40	4.00	4.60
	SD	1.64	1.87	1.95	3.08	2.07
1	561	7	4	3	3	3
2	561	7	7	6	5	6
3	561	9	4	4	6	5
4	561	8	7	7	8	8
5	561	2	3	1	1	1
6	561	7	4	3	3	3
7	561	7	7	6	5	6
8	561	9	4	4	6	5
9	561	8	7	7	8	8
10	561	2	3	1	1	1
11	561	7	4	3	3	3
12	561	7	7	6	5	6
13	561	9	4	4	6	5
14	561	8	7	7	8	8
15	561	2	3	1	1	1
16	561	7	4	3	3	3
17	561	7	7	6	5	6
18	561	9	4	4	6	5
19	561	8	7	7	8	8
20	561	2	3	1	1	1
	Mean	6.60	5.00	4.20	4.60	4.60
	SD	2.70	1.87	2.39	2.70	2.70

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.6: Consumer sensory evaluation of selected treatments at week 12

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	7	7	8	8	7
2	799	5	6	8	4	5
3	799	5	5	6	7	6
4	799	6	7	7	7	7
5	799	7	7	8	8	7
6	799	7	7	8	8	7
7	799	6	8	8	8	8
8	799	7	7	7	8	7
9	799	5	6	7	7	6
10	799	5	6	6	8	6
11	799	7	7	8	8	7
12	799	5	6	8	4	5
13	799	5	5	6	7	6
14	799	6	7	7	7	7
15	799	7	7	8	8	7
16	799	7	7	8	8	7
17	799	6	8	8	8	8
18	799	7	7	7	8	7
19	799	5	6	7	7	6
20	799	5	6	6	8	6
	Mean	6.00	6.60	7.30	7.30	6.60
	SD	0.94	0.84	0.82	1.25	0.84
1	126	8	6	5	5	5
2	126	5	8	8	8	8
3	126	5	5	6	5	5
4	126	7	7	7	7	7
5	126	7	7	6	8	7
6	126	6	6	6	7	6
7	126	5	6	4	6	5
8	126	7	6	6	7	7
9	126	6	6	5	6	6
10	126	5	7	6	8	7
11	126	8	6	5	5	5
12	126	5	8	8	8	8
13	126	5	5	6	5	5
14	126	7	7	7	7	7
15	126	7	7	6	8	7

Table 7.6$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 12

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
16	126	6	6	6	7	6
17	126	5	6	4	6	5
18	126	7	6	6	7	7
19	126	6	6	5	6	6
20	126	5	7	6	8	7
	Mean	6.10	6.40	5.90	6.70	6.30
	SD	1.10	0.84	1.10	1.16	1.06
1	853	6	6	4	5	5
2	853	5	7	8	7	7
3	853	5	5	6	5	5
4	853	7	6	6	7	6
5	853	7	6	7	7	6
6	853	5	6	5	7	6
7	853	6	5	6	6	6
8	853	4	6	6	8	5
9	853	5	7	5	7	6
10	853	7	6	7	6	7
11	853	6	6	4	5	5
12	853	5	7	8	7	7
13	853	5	5	6	5	5
14	853	7	6	6	7	6
15	853	7	6	7	7	6
16	853	5	6	5	7	6
17	853	6	5	6	6	6
18	853	4	6	6	8	5
19	853	5	7	5	7	6
20	853	7	6	7	6	7
	Mean	5.70	6.00	6.00	6.50	5.90
	SD	1.06	0.67	1.15	0.97	0.74
1	342	7	8	6	6	6
2	342	4	8	8	8	8
3	342	6	6	6	6	6
4	342	7	7	7	7	7
5	342	6	5	4	5	5
6	342	5	6	5	5	5
7	342	6	7	4	4	5
8	342	7	4	5	6	5

Table 7.6$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 12

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
9	342	6	5	3	2	4
10	342	4	5	1	1	3
11	342	7	8	6	6	6
12	342	4	8	8	8	8
13	342	6	6	6	6	6
14	342	7	7	7	7	7
15	342	6	5	4	5	5
16	342	5	6	5	5	5
17	342	6	7	4	4	5
18	342	7	4	5	6	5
19	342	6	5	3	2	4
20	342	4	5	1	1	3
	Mean	5.80	6.10	4.90	5.00	5.40
	SD	1.14	1.37	2.02	2.16	1.43
1	561	8	8	4	6	4
2	561	5	7	3	4	3
3	561	4	4	3	3	3
4	561	6	7	5	7	6
5	561	5	7	3	5	4
6	561	6	6	5	4	5
7	561	5	7	4	5	5
8	561	6	5	4	3	4
9	561	7	7	3	3	4
10	561	4	6	5	1	5
11	561	8	8	4	6	4
12	561	5	7	3	4	3
13	561	4	4	3	3	3
14	561	6	7	5	7	6
15	561	5	7	3	5	4
16	561	6	6	5	4	5
17	561	5	7	4	5	5
18	561	6	5	4	3	4
19	561	7	7	3	3	4
20	561	4	6	5	1	5
	Mean	5.60	6.40	3.90	4.10	4.30
	SD	1.26	1.17	0.88	1.73	0.95

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.7: Consumer sensory evaluation of selected treatments at week 14

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	6	7	8	8	7
2	799	6	7	7	6	6
3	799	4	5	6	7	6
4	799	6	7	7	7	7
5	799	5	7	8	8	6
6	799	7	7	8	5	6
7	799	5	8	6	7	7
8	799	7	8	7	8	7
9	799	6	6	7	7	6
10	799	7	6	6	8	7
11	799	6	7	8	8	7
12	799	6	7	7	6	6
13	799	4	5	6	7	6
14	799	6	7	7	7	7
15	799	5	7	8	8	6
16	799	7	7	8	5	6
17	799	5	8	6	7	7
18	799	7	8	7	8	7
19	799	6	6	7	7	6
20	799	7	6	6	8	7
	Mean	5.90	6.80	7.00	7.10	6.50
	SD	0.99	0.92	0.82	0.99	0.53
1	126	8	6	5	5	6
2	126	5	8	7	8	7
3	126	5	5	7	6	6
4	126	6	7	6	7	7
5	126	5	6	7	8	7
6	126	6	7	5	7	6
7	126	5	6	4	7	5
8	126	7	5	5	7	6
9	126	6	6	5	6	6
10	126	5	7	6	8	7
11	126	8	6	5	5	6
12	126	5	8	7	8	7
13	126	5	5	7	6	6
14	126	6	7	6	7	7
15	126	5	6	7	8	7

Table 7.7$_{contd.}$: Consumer sensory evaluation of selected treatments at week 14

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
16	126	6	7	5	7	6
17	126	5	6	4	7	5
18	126	7	5	5	7	6
19	126	6	6	5	6	6
20	126	5	7	6	8	7
	Mean	5.80	6.30	5.70	6.90	6.30
	SD	1.03	0.95	1.06	0.99	0.67
1	853	6	6	4	5	5
2	853	5	6	7	6	6
3	853	5	5	5	5	5
4	853	6	5	6	7	6
5	853	6	6	7	6	6
6	853	4	5	5	7	5
7	853	5	5	6	6	6
8	853	4	6	5	6	5
9	853	6	5	5	6	6
10	853	6	6	5	6	6
11	853	6	6	4	5	5
12	853	5	6	7	6	6
13	853	5	5	5	5	5
14	853	6	5	6	7	6
15	853	6	6	7	6	6
16	853	4	5	5	7	5
17	853	5	5	6	6	6
18	853	4	6	5	6	5
19	853	6	5	5	6	6
20	853	6	6	5	6	6
	Mean	5.30	5.50	5.50	6.00	5.60
	SD	0.82	0.53	0.97	0.67	0.52
1	342	7	8	4	5	5
2	342	4	7	6	5	6
3	342	5	6	4	4	5
4	342	7	6	5	5	6
5	342	6	6	4	4	4
6	342	6	5	4	4	5
7	342	4	6	4	3	5
8	342	6	5	4	1	5
9	342	5	4	3	1	4
10	342	4	4	1	1	3

Table 7.7contd.: Consumer sensory evaluation of selected treatments at week 14

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
11	342	7	8	4	5	5
12	342	4	7	6	5	6
13	342	5	6	4	4	5
14	342	7	6	5	5	6
15	342	6	6	4	4	4
16	342	6	5	4	4	5
17	342	4	6	4	3	5
18	342	6	5	4	1	5
19	342	5	4	3	1	4
20	342	4	4	1	1	3
	Mean	5.40	5.70	3.90	3.30	4.80
	SD	1.17	1.25	1.29	1.70	0.92
1	561	7	7	3	5	3
2	561	6	6	2	3	2
3	561	4	5	1	2	4
4	561	5	6	2	5	5
5	561	5	6	3	4	3
6	561	6	5	4	3	5
7	561	5	7	3	4	5
8	561	7	6	4	2	4
9	561	8	7	3	2	4
10	561	7	6	4	1	5
11	561	7	7	3	5	3
12	561	6	6	2	3	2
13	561	4	5	1	2	4
14	561	5	6	2	5	5
15	561	5	6	3	4	3
16	561	6	5	4	3	5
17	561	5	7	3	4	5
18	561	7	6	4	2	4
19	561	8	7	3	2	4
20	561	7	6	4	1	5
	Mean	6.00	6.10	2.90	3.10	4.00
	SD	1.25	0.74	0.99	1.37	1.05

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

Table 7.8: Consumer sensory evaluation of selected treatments at week 16

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
1	799	6	7	8	8	7
2	799	6	7	7	6	6
3	799	4	5	6	7	6
4	799	6	7	7	7	7
5	799	5	7	8	8	6
6	799	7	7	8	5	6
7	799	5	8	6	7	7
8	799	7	7	7	6	7
9	799	6	6	7	7	6
10	799	7	6	6	8	7
11	799	6	7	8	8	7
12	799	6	7	7	7	6
13	799	4	5	6	7	6
14	799	6	7	7	7	7
15	799	5	7	8	8	6
16	799	7	7	8	6	6
17	799	5	8	6	7	7
18	799	7	8	7	8	7
19	799	6	6	7	7	6
20	799	7	6	6	8	7
	Mean	5.90	6.75	7.00	7.10	6.50
	SD	0.97	0.85	0.79	0.85	0.51
1	126	7	6	5	5	6
2	126	5	7	7	8	7
3	126	5	5	7	6	6
4	126	6	7	6	7	7
5	126	5	6	6	7	7
6	126	6	7	5	7	6
7	126	5	6	4	7	5
8	126	7	5	5	7	6
9	126	6	6	5	6	6
10	126	5	7	6	7	7
11	126	6	6	5	5	7
12	126	5	7	7	8	7
13	126	5	5	7	6	6
14	126	6	7	6	7	7
15	126	5	6	7	8	7

Table 7.8$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 16

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
16	126	6	7	6	7	6
17	126	5	6	5	7	6
18	126	7	5	5	7	7
19	126	6	6	5	6	6
20	126	5	7	6	8	7
	Mean	5.65	6.20	5.75	6.80	6.45
	SD	0.75	0.77	0.91	0.89	0.60
1	853	6	6	5	6	6
2	853	5	6	7	6	6
3	853	5	5	5	5	5
4	853	6	5	6	7	6
5	853	6	6	7	6	6
6	853	5	5	5	7	5
7	853	5	5	6	6	6
8	853	6	6	5	6	5
9	853	6	5	5	6	6
10	853	6	6	5	6	6
11	853	6	6	5	5	5
12	853	5	6	7	6	6
13	853	5	6	5	5	5
14	853	6	5	6	7	6
15	853	6	6	7	6	6
16	853	4	5	6	7	5
17	853	5	6	6	7	6
18	853	4	6	5	6	6
19	853	6	5	5	6	6
20	853	6	6	5	6	6
	Mean	5.45	5.6	5.65	6.1	5.7
	SD	0.69	0.50	0.81	0.64	0.47
1	342	6	8	4	4	4
2	342	4	7	5	5	5
3	342	5	6	4	4	5
4	342	7	6	4	5	5
5	342	6	6	3	4	4
6	342	6	5	3	4	4
7	342	4	6	3	2	4
8	342	6	6	3	1	4
9	342	5	5	3	1	4
10	342	5	5	1	1	2

Table 7.8$_{\text{contd.}}$: Consumer sensory evaluation of selected treatments at week 16

Panelist	Sample	Color	Texture	Flavour	Freshness	Overall
11	342	7	8	3	4	3
12	342	4	7	4	4	4
13	342	5	6	4	4	4
14	342	6	6	4	5	5
15	342	6	5	3	2	4
16	342	6	5	3	2	3
17	342	4	6	3	1	4
18	342	6	5	4	1	4
19	342	4	4	2	1	3
20	342	4	4	1	1	3
	Mean	5.30	5.80	3.20	2.80	3.90
	SD	1.03	1.11	1.01	1.61	0.79
1	561	6	6	3	4	3
2	561	6	6	2	3	2
3	561	4	5	1	2	4
4	561	5	6	2	4	4
5	561	5	6	3	4	3
6	561	6	5	3	3	4
7	561	5	6	3	4	4
8	561	5	6	4	2	4
9	561	6	5	3	2	4
10	561	6	6	4	1	4
11	561	7	5	3	4	3
12	561	6	6	2	3	2
13	561	4	5	2	2	4
14	561	5	6	2	4	4
15	561	5	6	3	4	3
16	561	6	5	5	3	4
17	561	5	5	3	3	4
18	561	7	6	5	2	4
19	561	6	5	3	2	4
20	561	7	6	4	1	5
	Mean	5.60	5.60	3.00	2.85	3.65
	SD	0.88	0.50	1.03	1.04	0.75

Note: 799= HPP/600/1; 126= HPP/600/5; 853= HP+LAE/600/1; 342= LAE 200; 561= LAE 315.

APPENDIX 8

Statistical analysis

8.1 Minitab output for the growth of APCs in the screening phase

General Linear Model: Control, HPP 450/1, ... versus Storage time

```
Factor        Type   Levels  Values
Storage time  fixed      13  0, 2, 4, 6, 8, 9, 10, 11, 12, 13, 14, 15, 16


Analysis of Variance for Control, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  128.122  128.122  10.677  84.64  0.000
Error         52    6.560    6.560   0.126
Total         64  134.682


S = 0.355170   R-Sq = 95.13%   R-Sq(adj) = 94.01%


Unusual Observations for Control

Obs  Control      Fit   SE Fit  Residual  St Resid
 23  3.07000  3.73400  0.15884  -0.66400     -2.09 R
 35  3.22000  4.01200  0.15884  -0.79200     -2.49 R
 36  3.15000  4.14400  0.15884  -0.99400     -3.13 R
 38  4.79000  4.14400  0.15884   0.64600      2.03 R
 46  4.98000  4.11200  0.15884   0.86800      2.73 R
 49  3.22000  4.11200  0.15884  -0.89200     -2.81 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP 450/1, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS       F      P
Storage time  12  147.453  147.453  12.288  319.88  0.000
Error         52    1.998    1.998   0.038
Total         64  149.450


S = 0.195995   R-Sq = 98.66%   R-Sq(adj) = 98.35%


Unusual Observations for HPP 450/1

Obs  HPP 450/1      Fit   SE Fit  Residual  St Resid
 36    1.00000  1.42000  0.08765  -0.42000     -2.40 R
 37    1.00000  1.42000  0.08765  -0.42000     -2.40 R
 38    1.00000  1.42000  0.08765  -0.42000     -2.40 R
 39    2.00000  1.42000  0.08765   0.58000      3.31 R
 40    2.10000  1.42000  0.08765   0.68000      3.88 R
 61    4.55000  4.94600  0.08765  -0.39600     -2.26 R
 63    5.32000  4.94600  0.08765   0.37400      2.13 R

R denotes an observation with a large standardized residual.
```

```
Analysis of Variance for HPP 450/3.5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS       F      P
Storage time  12  147.892  147.892  12.324  153.70  0.000
Error         52    4.169    4.169   0.080
Total         64  152.061


S = 0.283165   R-Sq = 97.26%   R-Sq(adj) = 96.63%


Unusual Observations for HPP 450/3.5

Obs  HPP 450/3.5      Fit   SE Fit  Residual  St Resid
 17      2.17000  1.44200  0.12664   0.72800      2.87 R
 18      2.04000  1.44200  0.12664   0.59800      2.36 R
 33      2.20000  1.67000  0.12664   0.53000      2.09 R
 34      1.00000  1.67000  0.12664  -0.67000     -2.65 R
 49      3.12000  3.66400  0.12664  -0.54400     -2.15 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP 450/5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  59.9845  59.9845  4.9987  24.01  0.000
Error         52  10.8272  10.8272  0.2082
Total         64  70.8117


S = 0.456305   R-Sq = 84.71%   R-Sq(adj) = 81.18%


Unusual Observations for HPP 450/5

Obs  HPP 450/5      Fit   SE Fit  Residual  St Resid
 35    1.00000  1.87000  0.20407  -0.87000     -2.13 R
 46    1.77000  3.54800  0.20407  -1.77800     -4.36 R
 47    4.38000  3.54800  0.20407   0.83200      2.04 R
 48    4.44000  3.54800  0.20407   0.89200      2.19 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP 600/1, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS   Adj MS     F      P
Storage time  12  0.62506  0.62506  0.05209  0.85  0.597
Error         52  3.17448  3.17448  0.06105
Total         64  3.79954


S = 0.247078   R-Sq = 16.45%   R-Sq(adj) = 0.00%


Unusual Observations for HPP 600/1

Obs  HPP 600/1      Fit   SE Fit  Residual  St Resid
 48    2.39000  1.27800  0.11050   1.11200      5.03 R
 53    2.34000  1.26800  0.11050   1.07200      4.85 R
```

```
R denotes an observation with a large standardized residual.


Analysis of Variance for HPP 600/3.5, using Adjusted SS for Tests

Source        DF    Seq SS    Adj SS  Adj MS      F      P
Storage time  12  111.2882  111.2882  9.2740  42.34  0.000
Error         52   11.3889   11.3889  0.2190
Total         64  122.6771


S = 0.467993   R-Sq = 90.72%   R-Sq(adj) = 88.57%


Unusual Observations for HPP 600/3.5

Obs  HPP 600/3.5      Fit   SE Fit  Residual  St Resid
 28      2.23000  1.24600  0.20929   0.98400      2.35 R
 42      1.00000  2.47200  0.20929  -1.47200     -3.52 R
 43      3.34000  2.47200  0.20929   0.86800      2.07 R
 51      5.40000  3.69000  0.20929   1.71000      4.09 R
 53      2.41000  3.69000  0.20929  -1.28000     -3.06 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP/600/5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  31.4986  31.4986  2.6249  14.07  0.000
Error         52   9.7018   9.7018  0.1866
Total         64  41.2004


S = 0.431940   R-Sq = 76.45%   R-Sq(adj) = 71.02%


Unusual Observations for HPP/600/5

Obs  HPP/600/5      Fit   SE Fit  Residual  St Resid
 51    2.85000  1.92800  0.19317   0.92200      2.39 R
 62    2.12000  2.89800  0.19317  -0.77800     -2.01 R
 63    1.34000  2.89800  0.19317  -1.55800     -4.03 R
 64    4.60000  2.89800  0.19317   1.70200      4.41 R

R denotes an observation with a large standardized residual.
```

General Linear Model: Control, HPP+LAE 450/1, ... versus Storage time

```
Factor        Type   Levels  Values
Storage time  fixed      13  0, 2, 4, 6, 8, 9, 10, 11, 12, 13, 14, 15, 16


Analysis of Variance for Control, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  128.122  128.122  10.677  84.64  0.000
Error         52    6.560    6.560   0.126
Total         64  134.682
```

```
S = 0.355170   R-Sq = 95.13%   R-Sq(adj) = 94.01%


Unusual Observations for Control

Obs  Control      Fit  SE Fit  Residual  St Resid
 23  3.07000  3.73400  0.15884  -0.66400     -2.09 R
 35  3.22000  4.01200  0.15884  -0.79200     -2.49 R
 36  3.15000  4.14400  0.15884  -0.99400     -3.13 R
 38  4.79000  4.14400  0.15884   0.64600      2.03 R
 46  4.98000  4.11200  0.15884   0.86800      2.73 R
 49  3.22000  4.11200  0.15884  -0.89200     -2.81 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP+LAE 450/1, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS     F      P
Storage time  12  20.4278  20.4278  1.7023  8.28  0.000
Error         52  10.6933  10.6933  0.2056
Total         64  31.1211


S = 0.453475   R-Sq = 65.64%   R-Sq(adj) = 57.71%


Unusual Observations for HPP+LAE 450/1

     HPP+LAE
Obs    450/1      Fit  SE Fit  Residual  St Resid
 21  2.04000  1.20800  0.20280   0.83200      2.05 R
 34  2.30000  1.30000  0.20280   1.00000      2.47 R
 46  1.00000  2.00000  0.20280  -1.00000     -2.47 R
 50  3.00000  2.00000  0.20280   1.00000      2.47 R
 53  1.00000  2.42000  0.20280  -1.42000     -3.50 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP+LAE 450/3.5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  83.5724  83.5724  6.9644  31.20  0.000
Error         52  11.6084  11.6084  0.2232
Total         64  95.1808


S = 0.472481   R-Sq = 87.80%   R-Sq(adj) = 84.99%


Unusual Observations for HPP+LAE 450/3.5

     HPP+LAE
Obs  450/3.5      Fit  SE Fit  Residual  St Resid
 23  2.76000  1.35200  0.21130   1.40800      3.33 R
 28  2.34000  1.40200  0.21130   0.93800      2.22 R
 38  2.24000  1.37800  0.21130   0.86200      2.04 R
 46  4.59000  3.27000  0.21130   1.32000      3.12 R

R denotes an observation with a large standardized residual.
```

```
Analysis of Variance for HPP+LAE 450/5, using Adjusted SS for Tests

Source        DF    Seq SS    Adj SS  Adj MS      F      P
Storage time  12  103.9807  103.9807  8.6651  37.35  0.000
Error         52   12.0651   12.0651  0.2320
Total         64  116.0458


S = 0.481686   R-Sq = 89.60%   R-Sq(adj) = 87.20%


Unusual Observations for HPP+LAE 450/5

     HPP+LAE
Obs    450/5      Fit   SE Fit  Residual  St Resid
 21  3.51000  1.76200  0.21542   1.74800      4.06 R
 29  2.40000  1.48000  0.21542   0.92000      2.14 R
 43  2.50000  1.59000  0.21542   0.91000      2.11 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP+LAE 600/1, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  42.7084  42.7084  3.5590  22.61  0.000
Error         52   8.1851   8.1851  0.1574
Total         64  50.8935


S = 0.396744   R-Sq = 83.92%   R-Sq(adj) = 80.21%


Unusual Observations for HPP+LAE 600/1

     HPP+LAE
Obs    600/1      Fit   SE Fit  Residual  St Resid
 51  3.20000  1.44000  0.17743   1.76000      4.96 R
 57  4.00000  3.11600  0.17743   0.88400      2.49 R
 59  2.00000  3.11600  0.17743  -1.11600     -3.14 R
 63  2.50000  3.40000  0.17743  -0.90000     -2.54 R
 65  4.30000  3.40000  0.17743   0.90000      2.54 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP+LAE 600/3.5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS     F      P
Storage time  12  33.0723  33.0723  2.7560  4.58  0.000
Error         52  31.3023  31.3023  0.6020
Total         64  64.3746


S = 0.775865   R-Sq = 51.37%   R-Sq(adj) = 40.15%


Unusual Observations for HPP+LAE 600/3.5

     HPP+LAE
Obs  600/3.5      Fit   SE Fit  Residual  St Resid
 42  1.30000  2.94400  0.34698  -1.64400     -2.37 R
```

```
 43  5.38000  2.94400  0.34698   2.43600      3.51 R
 44  4.90000  2.94400  0.34698   1.95600      2.82 R
 45  1.00000  2.94400  0.34698  -1.94400     -2.80 R
 53  4.50000  1.78000  0.34698   2.72000      3.92 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP+LAE 600/5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS     F      P
Storage time  12  32.7114  32.7114  2.7260  9.16  0.000
Error         52  15.4758  15.4758  0.2976
Total         64  48.1872


S = 0.545537   R-Sq = 67.88%   R-Sq(adj) = 60.47%


Unusual Observations for HPP+LAE 600/5

     HPP+LAE
Obs    600/5      Fit   SE Fit  Residual  St Resid
 43  2.61000  1.59000  0.24397   1.02000      2.09 R
 46  4.70000  2.40800  0.24397   2.29200      4.70 R
 48  1.00000  2.40800  0.24397  -1.40800     -2.89 R

R denotes an observation with a large standardized residual.
```

General Linear Model: Control, LAE 200 versus Storage time

```
Factor        Type   Levels  Values
Storage time  fixed      13  0, 2, 4, 6, 8, 9, 10, 11, 12, 13, 14, 15, 16


Analysis of Variance for Control, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
Storage time  12  128.122  128.122  10.677  84.64  0.000
Error         52    6.560    6.560   0.126
Total         64  134.682


S = 0.355170   R-Sq = 95.13%   R-Sq(adj) = 94.01%


Unusual Observations for Control

Obs  Control      Fit   SE Fit  Residual  St Resid
 23  3.07000  3.73400  0.15884  -0.66400     -2.09 R
 35  3.22000  4.01200  0.15884  -0.79200     -2.49 R
 36  3.15000  4.14400  0.15884  -0.99400     -3.13 R
 38  4.79000  4.14400  0.15884   0.64600      2.03 R
 46  4.98000  4.11200  0.15884   0.86800      2.73 R
 49  3.22000  4.11200  0.15884  -0.89200     -2.81 R

R denotes an observation with a large standardized residual.


Analysis of Variance for LAE 200, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS      F      P
```

```
Storage time  12  78.1502  78.1502  6.5125  45.67  0.000
Error         52   7.4146   7.4146  0.1426
Total         64  85.5647


S = 0.377608   R-Sq = 91.33%   R-Sq(adj) = 89.33%


Unusual Observations for LAE 200

Obs  LAE 200      Fit   SE Fit  Residual  St Resid
 34  2.00000  1.28000  0.16887   0.72000      2.13 R
 40  1.90000  1.18000  0.16887   0.72000      2.13 R
 51  1.39000  2.69800  0.16887  -1.30800     -3.87 R
 52  4.10000  2.69800  0.16887   1.40200      4.15 R

R denotes an observation with a large standardized residual.
```

8.2 Minitab output for the growth of APCs in the selected treatments

General Linear Model: Control, HPP 600/1, ... versus Storage time

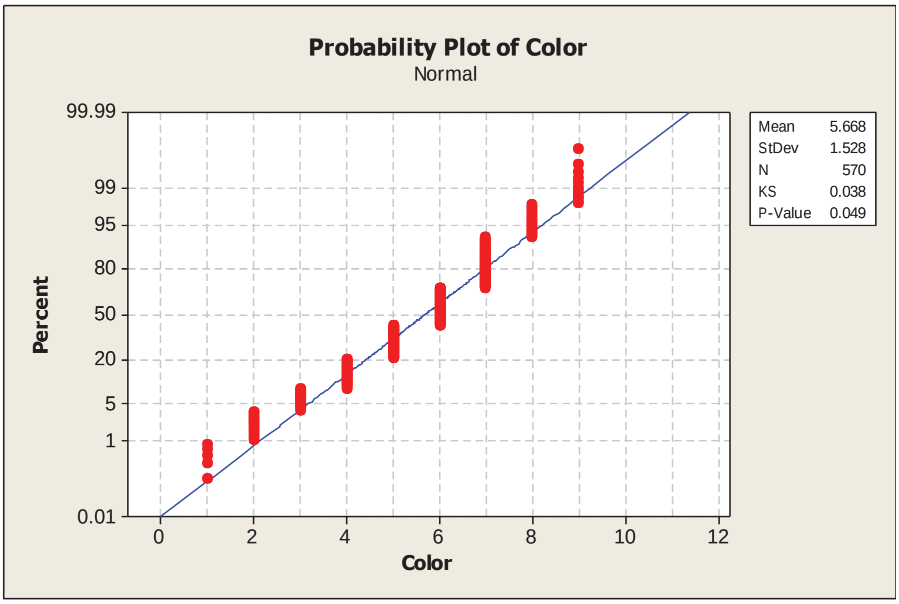
```
Factor        Type   Levels  Values
Storage time  fixed       9  0, 2, 4, 6, 8, 10, 12, 14, 16


Analysis of Variance for Control, using Adjusted SS for Tests

Source        DF  Seq SS  Adj SS  Adj MS      F      P
Storage time   8  92.265  92.265  11.533  65.27  0.000
Error         36   6.361   6.361   0.177
Total         44  98.626


S = 0.420356   R-Sq = 93.55%   R-Sq(adj) = 92.12%


Unusual Observations for Control

Obs  Control      Fit   SE Fit  Residual  St Resid
 25  3.81000  4.87200  0.18799  -1.06200     -2.82 R
 32  6.23000  5.38800  0.18799   0.84200      2.24 R
 39  6.43000  5.50200  0.18799   0.92800      2.47 R
 40  4.75000  5.50200  0.18799  -0.75200     -2.00 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP 600/1, using Adjusted SS for Tests

Source        DF     Seq SS     Adj SS     Adj MS   F  P
Storage time   8  0.0000000  0.0000000  0.0000000  **
Error         36  0.0000000  0.0000000  0.0000000
Total         44  0.0000000

** Denominator of F-test is zero.


S = 0   R-Sq = *%   R-Sq(adj) = *%


Analysis of Variance for HPP 600/5, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS   Adj MS     F      P
Storage time   8  1.32428  1.32428  0.16554  2.63  0.022
Error         36  2.26528  2.26528  0.06292
Total         44  3.58956


S = 0.250847   R-Sq = 36.89%   R-Sq(adj) = 22.87%


Unusual Observations for HPP 600/5

Obs  HPP 600/5      Fit   SE Fit  Residual  St Resid
 31    1.00000  1.54800  0.11218  -0.54800     -2.44 R
 32    1.00000  1.54800  0.11218  -0.54800     -2.44 R
 33    2.42000  1.54800  0.11218   0.87200      3.89 R
 34    1.00000  1.54800  0.11218  -0.54800     -2.44 R
```

```
 35   2.32000  1.54800  0.11218   0.77200      3.44 R

R denotes an observation with a large standardized residual.


Analysis of Variance for HPP+LAE 600/1, using Adjusted SS for Tests

Source        DF     Seq SS      Adj SS      Adj MS   F  P
Storage time   8  0.0000000  0.0000000  0.0000000  **
Error         36  0.0000000  0.0000000  0.0000000
Total         44  0.0000000

** Denominator of F-test is zero.


S = 0   R-Sq = *%   R-Sq(adj) = *%


Analysis of Variance for LAE 200, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS  Adj MS     F      P
Storage time   8   75.886  75.886   9.486  6.00  0.000
Error         36   56.891  56.891   1.580
Total         44  132.776


S = 1.25710   R-Sq = 57.15%   R-Sq(adj) = 47.63%


Unusual Observations for LAE 200

Obs  LAE 200      Fit   SE Fit  Residual  St Resid
 23  1.00000  4.18800  0.56219  -3.18800     -2.84 R
 27  1.00000  4.19400  0.56219  -3.19400     -2.84 R
 31  7.80000  5.37800  0.56219   2.42200      2.15 R

R denotes an observation with a large standardized residual.


Analysis of Variance for LAE 315, using Adjusted SS for Tests

Source        DF   Seq SS   Adj SS   Adj MS      F      P
Storage time   8  59.0165  59.0165  7.3771  25.48  0.000
Error         36  10.4226  10.4226  0.2895
Total         44  69.4391


S = 0.538069   R-Sq = 84.99%   R-Sq(adj) = 81.65%


Unusual Observations for LAE 315

Obs  LAE 315      Fit   SE Fit  Residual  St Resid
  2  2.98000  1.88800  0.24063   1.09200      2.27 R
 10  1.00000  2.43000  0.24063  -1.43000     -2.97 R
 26  3.66000  4.65800  0.24063  -0.99800     -2.07 R

R denotes an observation with a large standardized residual.
```

8.3 Minitab output for the sensory evaluation of selected treatments

Week 2

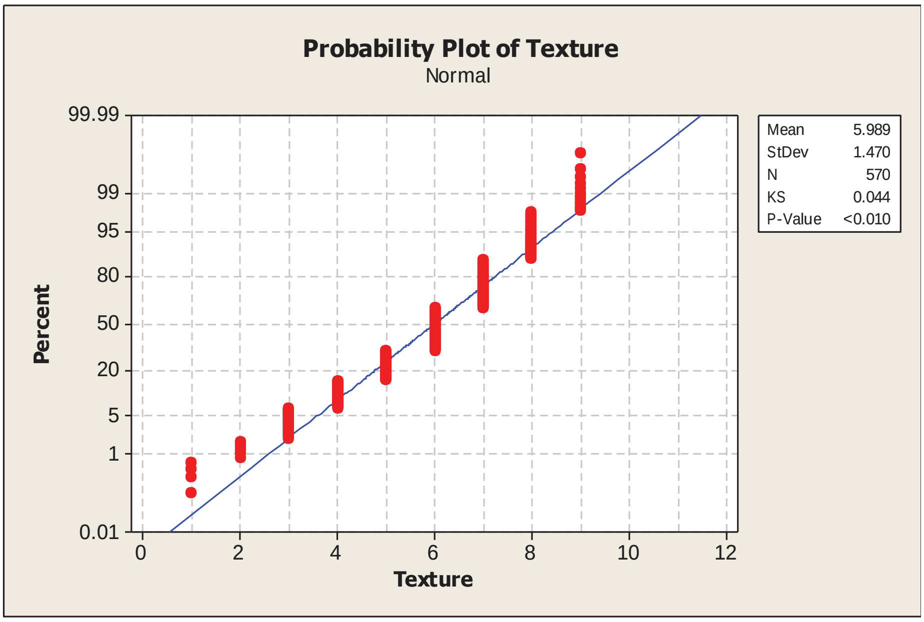
Normality plot for color



One-way ANOVA: Color versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    6.14  1.53  0.47  0.761
Error   110  362.52  3.30
Total   114  368.66

S = 1.815   R-Sq = 1.67%   R-Sq(adj) = 0.00%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev   -+---------+---------+---------+--------
126    23  5.609  1.777    (-----------*------------)
342    23  6.130  1.604           (-----------*------------)
561    23  5.478  2.129  (-----------*------------)
799    23  5.913  1.730        (------------*-----------)
853    23  5.696  1.795     (------------*-----------)
                          -+---------+---------+---------+--------
                          4.80      5.40      6.00      6.60

Pooled StDev = 1.815
```

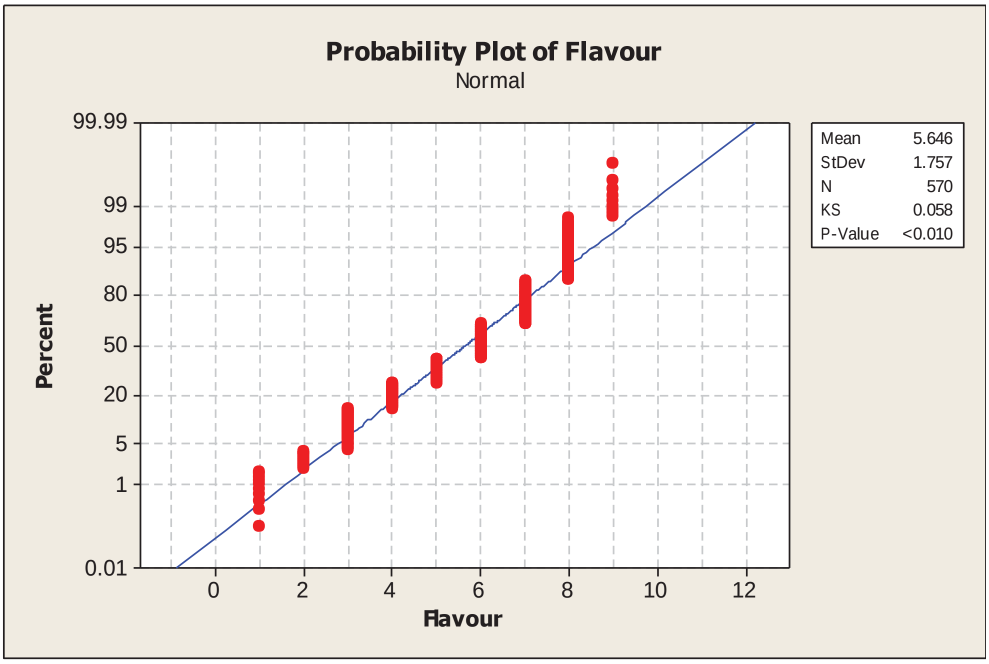
Normality plot for texture



One-way ANOVA: Texture versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    1.57  0.39  0.14  0.967
Error   110  306.78  2.79
Total   114  308.35

S = 1.670   R-Sq = 0.51%   R-Sq(adj) = 0.00%


                           Individual 95% CIs For Mean Based on
                           Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    23  6.304  1.663     (-------------*-------------)
342    23  6.348  1.496      (-------------*-------------)
561    23  6.087  1.807  (-------------*-------------)
799    23  6.435  1.562        (-------------*------------)
853    23  6.348  1.799      (-------------*-------------)
                         --+---------+---------+---------+-------
                         5.50      6.00      6.50      7.00

Pooled StDev = 1.670
```

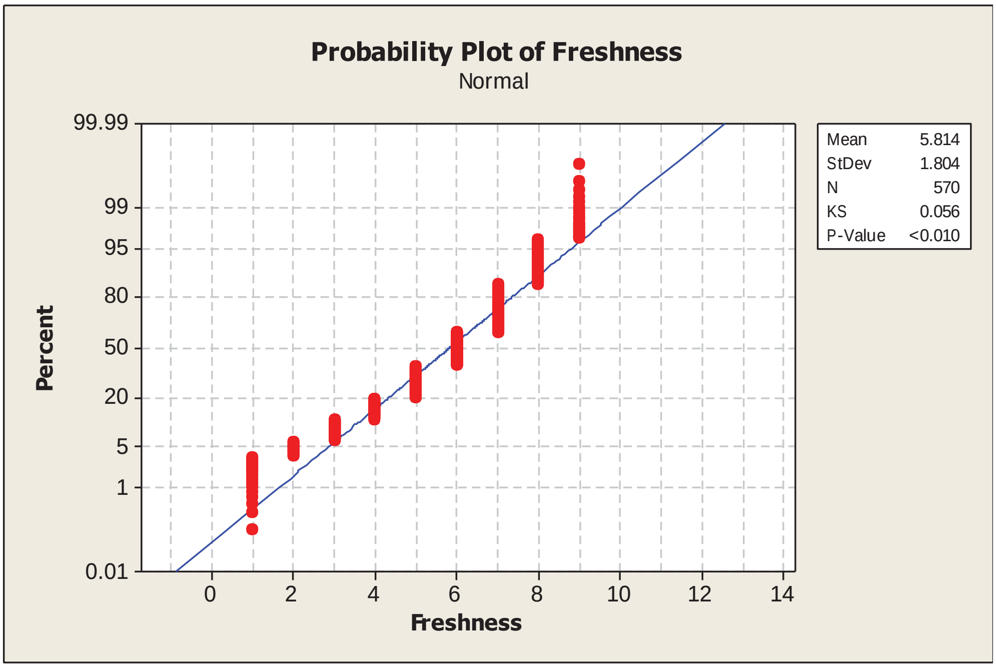
Normality plot for Flavour



One-way ANOVA: Flavor versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    5.60  1.40  0.51  0.725
Error   110  299.04  2.72
Total   114  304.64

S = 1.649   R-Sq = 1.84%   R-Sq(adj) = 0.00%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    23  6.174  1.723     (------------*-------------)
342    23  6.261  1.602       (------------*-------------)
561    23  6.087  1.730  (-------------*------------)
799    23  6.696  1.363             (-------------*-------------)
853    23  6.130  1.792   (-------------*------------)
                          --+---------+---------+---------+-------
                          5.50      6.00      6.50      7.00

Pooled StDev = 1.649
```

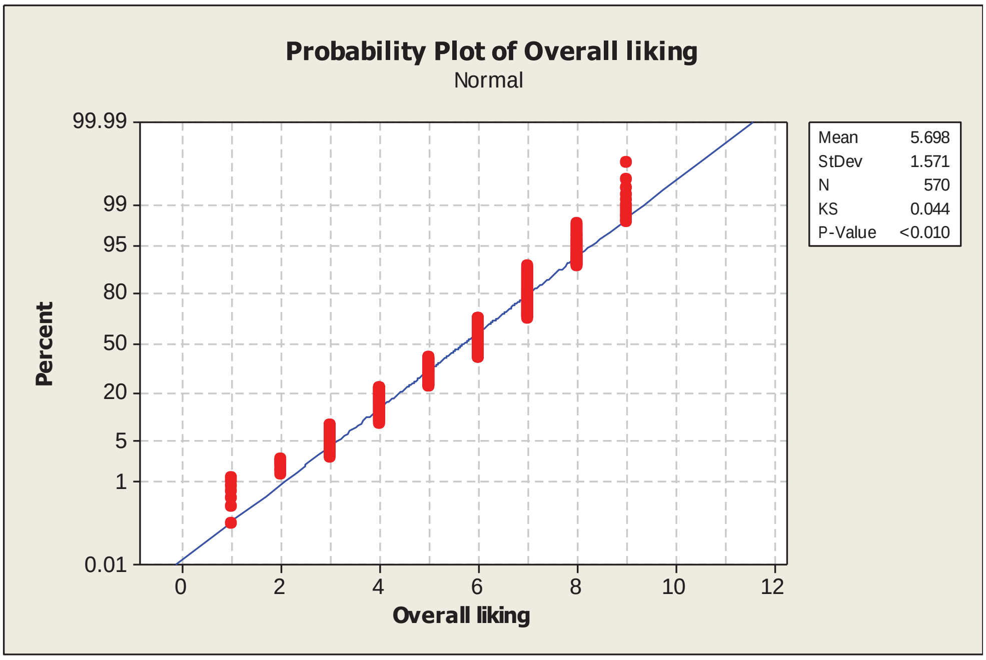
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    3.97  0.99  0.40  0.811
Error   110  275.48  2.50
Total   114  279.44

S = 1.583   R-Sq = 1.42%   R-Sq(adj) = 0.00%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    23  6.565  1.779          (------------*------------)
342    23  6.348  1.496       (------------*------------)
561    23  6.043  1.551  (------------*------------)
799    23  6.478  1.534        (-------------*------------)
853    23  6.217  1.536     (------------*------------)
                          --+---------+---------+---------+-------
                          5.50      6.00      6.50      7.00

Pooled StDev = 1.583
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    5.69  1.42  0.52  0.723
Error   110  302.17  2.75
Total   114  307.86

S = 1.657   R-Sq = 1.85%   R-Sq(adj) = 0.00%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev  ----+---------+---------+---------+-----
126    23  6.174  1.669    (-----------*----------)
342    23  6.304  1.690       (----------*----------)
561    23  5.826  1.749  (----------*-----------)
799    23  6.478  1.442          (----------*----------)
853    23  6.043  1.718   (-----------*----------)
                          ----+---------+---------+---------+-----
                            5.40      6.00      6.60      7.20

Pooled StDev = 1.657
```

General Linear Model: Color, Texture, ... versus Sample

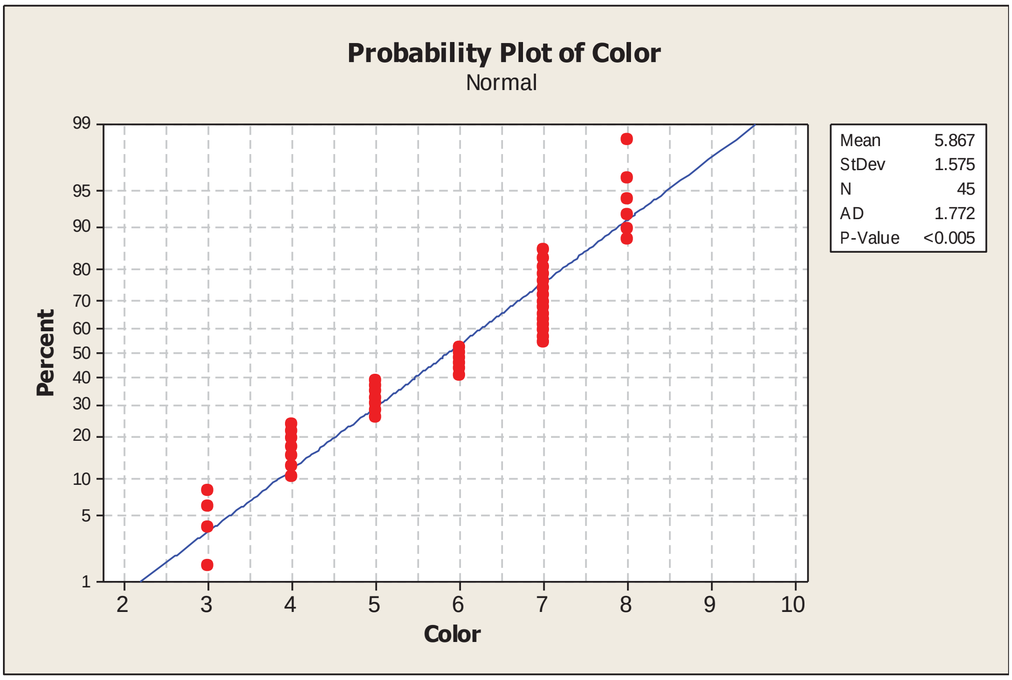
```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4    6.139    6.139   1.535  0.47  0.761
Error   110  362.522  362.522   3.296
```

```
Total   114  368.661


S = 1.81539   R-Sq = 1.67%   R-Sq(adj) = 0.00%


Unusual Observations for Color

Obs    Color      Fit   SE Fit  Residual  St Resid
 10  2.00000  5.91304  0.37854  -3.91304     -2.20 R
 46  2.00000  5.60870  0.37854  -3.60870     -2.03 R
 50  2.00000  5.69565  0.37854  -3.69565     -2.08 R
113  1.00000  5.47826  0.37854  -4.47826     -2.52 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4    1.565    1.565   0.391  0.14  0.967
Error   110  306.783  306.783   2.789
Total   114  308.348


S = 1.67001   R-Sq = 0.51%   R-Sq(adj) = 0.00%


Unusual Observations for Texture

Obs  Texture      Fit   SE Fit  Residual  St Resid
 11  2.00000  6.43478  0.34822  -4.43478     -2.72 R
 46  3.00000  6.30435  0.34822  -3.30435     -2.02 R
 57  2.00000  6.34783  0.34822  -4.34783     -2.66 R
 80  3.00000  6.34783  0.34822  -3.34783     -2.05 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavor, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4    5.600    5.600   1.400  0.51  0.725
Error   110  299.043  299.043   2.719
Total   114  304.643


S = 1.64881   R-Sq = 1.84%   R-Sq(adj) = 0.00%


Unusual Observations for Flavor

Obs   Flavor      Fit   SE Fit  Residual  St Resid
 57  1.00000  6.13043  0.34380  -5.13043     -3.18 R
 79  3.00000  6.26087  0.34380  -3.26087     -2.02 R
 92  3.00000  6.26087  0.34380  -3.26087     -2.02 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Freshness, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
```

```
Sample    4    3.965    3.965   0.991  0.40  0.811
Error   110  275.478  275.478   2.504
Total   114  279.443


S = 1.58251   R-Sq = 1.42%   R-Sq(adj) = 0.00%


Unusual Observations for Freshness

Obs  Freshness      Fit   SE Fit  Residual  St Resid
 23    3.00000  6.47826  0.32998  -3.47826     -2.25 R
 46    3.00000  6.56522  0.32998  -3.56522     -2.30 R
 61    3.00000  6.21739  0.32998  -3.21739     -2.08 R
 79    3.00000  6.34783  0.32998  -3.34783     -2.16 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Overall, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4    5.687    5.687   1.422  0.52  0.723
Error   110  302.174  302.174   2.747
Total   114  307.861


S = 1.65742   R-Sq = 1.85%   R-Sq(adj) = 0.00%


Unusual Observations for Overall

Obs  Overall      Fit   SE Fit  Residual  St Resid
 10  3.00000  6.47826  0.34560  -3.47826     -2.15 R
 46  2.00000  6.17391  0.34560  -4.17391     -2.57 R
 57  2.00000  6.04348  0.34560  -4.04348     -2.49 R
 79  3.00000  6.30435  0.34560  -3.30435     -2.04 R
 92  3.00000  6.30435  0.34560  -3.30435     -2.04 R

R denotes an observation with a large standardized residual.
```

Week 4

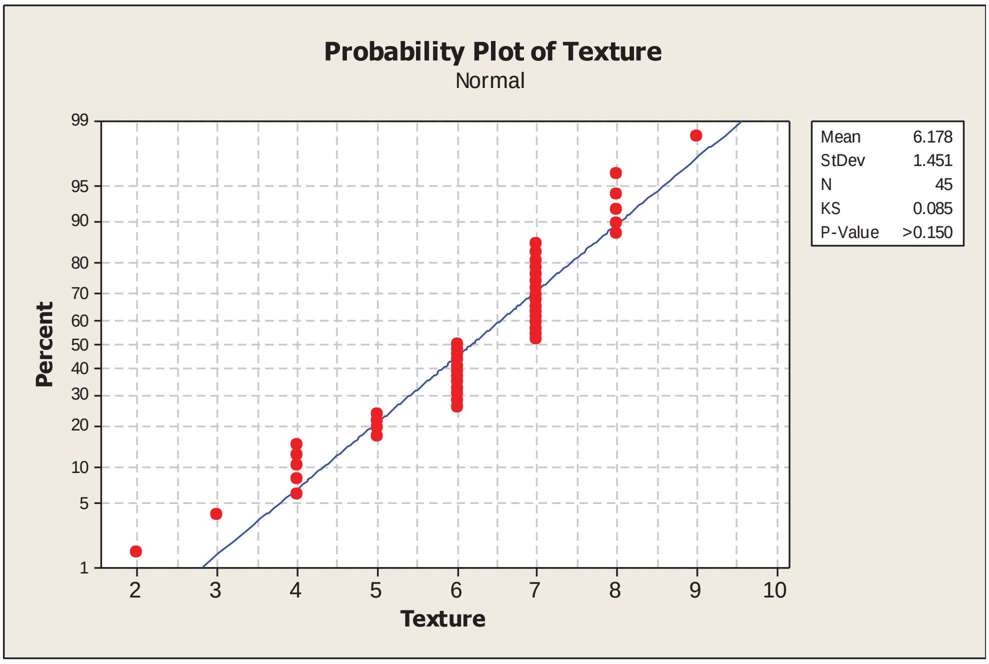
Normality plot for Color



One-way ANOVA: Color versus Sample

```
Source  DF      SS    MS     F      P
Sample   4    4.84  1.21  0.48  0.752
Error   85  215.56  2.54
Total   89  220.40

S = 1.592   R-Sq = 2.20%   R-Sq(adj) = 0.00%


                      Individual 95% CIs For Mean Based on
                      Pooled StDev
Level   N   Mean  StDev  -+---------+---------+---------+--------
126    18  6.000  1.749       (----------*---------)
342    27  5.667  1.593   (--------*--------)
561     9  6.333  1.500     (--------------*---------------)
799    18  5.778  1.353   (----------*---------)
853    18  5.556  1.688  (---------*----------)
                         -+---------+---------+---------+--------
                       4.90      5.60      6.30      7.00

Pooled StDev = 1.592
```

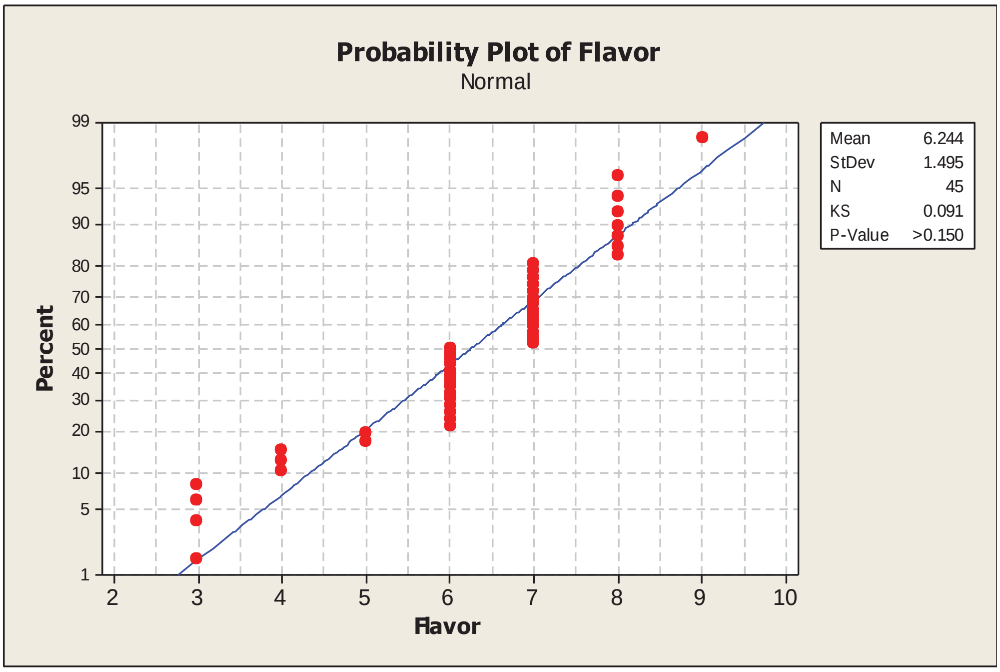
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source  DF      SS    MS     F      P
Sample   4    0.40  0.10  0.05  0.996
Error   85  176.00  2.07
Total   89  176.40

S = 1.439   R-Sq = 0.23%   R-Sq(adj) = 0.00%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev  ---------+---------+---------+---------+
126    18  6.222  1.665         (------------*-------------)
342    27  6.222  1.502           (----------*----------)
561     9  6.000  1.871  (------------------*------------------)
799    18  6.222  1.166         (------------*-------------)
853    18  6.222  1.060         (------------*-------------)
                          ---------+---------+---------+---------+
                                5.50      6.00      6.50      7.00

Pooled StDev = 1.439
```

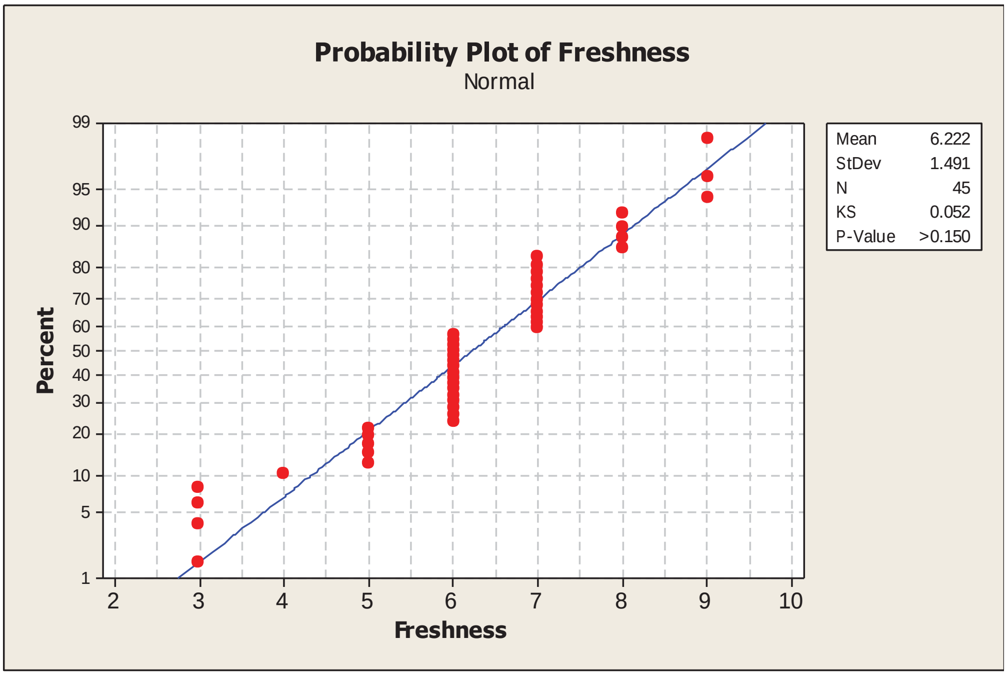
Normality plot for Flavour



One-way ANOVA: Flavor versus Sample

```
Source  DF      SS    MS     F      P
Sample   4   20.32  5.08  2.82  0.030
Error   85  153.33  1.80
Total   89  173.66

S = 1.343   R-Sq = 11.70%   R-Sq(adj) = 7.55%


                          Individual 95% CIs For Mean Based on Pooled StDev
Level   N   Mean  StDev   +---------+---------+---------+---------
126    18  5.667  1.283    (--------*--------)
342    27  6.556  1.086             (-------*------)
561     9  5.778  1.986   (------------*-----------)
799    18  7.000  0.686                 (--------*--------)
853    18  6.222  1.801          (--------*--------)
                         +---------+---------+---------+---------
                        4.90      5.60      6.30      7.00

Pooled StDev = 1.343
```

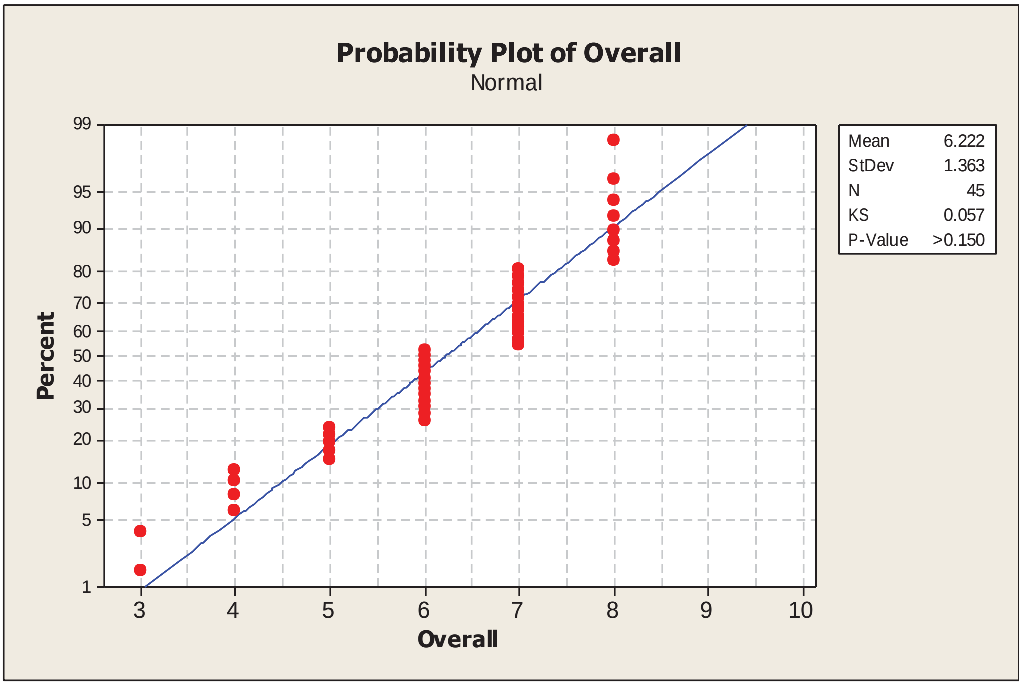
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source  DF      SS    MS     F      P
Sample   4    5.73  1.43  0.63  0.641
Error   85  192.89  2.27
Total   89  198.62

S = 1.506   R-Sq = 2.89%   R-Sq(adj) = 0.00%


                       Individual 95% CIs For Mean Based on
                       Pooled StDev
Level   N   Mean  StDev  -----+---------+---------+---------+----
126    18  5.778  1.060  (----------*-----------)
342    27  6.333  1.519           (---------*--------)
561     9  6.111  1.453  (----------------*---------------)
799    18  6.444  1.688           (----------*-----------)
853    18  6.444  1.688           (----------*-----------)
                         -----+---------+---------+---------+----
                            5.40      6.00      6.60      7.20

Pooled StDev = 1.506
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source  DF      SS    MS     F      P
Sample   4    1.33  0.33  0.19  0.942
Error   85  148.22  1.74
Total   89  149.56

S = 1.321   R-Sq = 0.89%   R-Sq(adj) = 0.00%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ---+---------+---------+---------+------
126    18  6.111  1.323     (-----------*------------)
342    27  6.222  1.155        (---------*----------)
561     9  6.222  1.787  (----------------*-----------------)
799    18  6.111  1.323     (-----------*------------)
853    18  6.444  1.294          (-----------*-----------)
                         ---+---------+---------+---------+------
                          5.50      6.00      6.50      7.00

Pooled StDev = 1.321
```

General Linear Model: Color, Texture, ... versus Sample

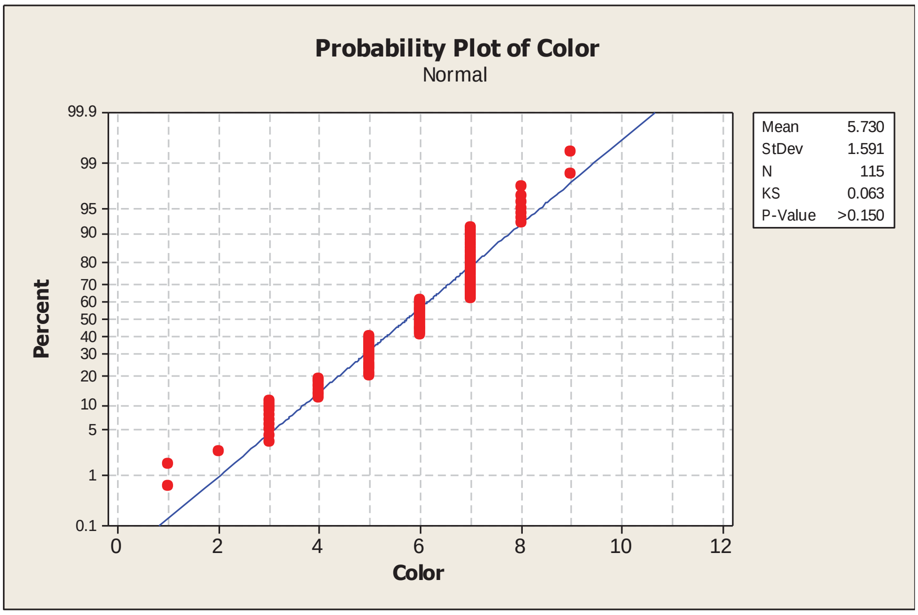
```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4    4.844    4.844   1.211  0.48  0.752
Error   85  215.556  215.556   2.536
```

```
Total   89  220.400


S = 1.59247   R-Sq = 2.20%   R-Sq(adj) = 0.00%


Analysis of Variance for Texture, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4    0.400    0.400   0.100  0.05  0.996
Error   85  176.000  176.000   2.071
Total   89  176.400


S = 1.43895   R-Sq = 0.23%   R-Sq(adj) = 0.00%


Unusual Observations for Texture

Obs  Texture      Fit  SE Fit  Residual  St Resid
 22  2.00000  6.22222  0.33916  -4.22222     -3.02 R
 31  2.00000  6.22222  0.33916  -4.22222     -3.02 R
 76  3.00000  6.00000  0.47965  -3.00000     -2.21 R
 80  9.00000  6.00000  0.47965   3.00000      2.21 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavor, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   20.322   20.322   5.081  2.82  0.030
Error   85  153.333  153.333   1.804
Total   89  173.656


S = 1.34310   R-Sq = 11.70%   R-Sq(adj) = 7.55%


Unusual Observations for Flavor

Obs   Flavor      Fit  SE Fit  Residual  St Resid
 22  3.00000  5.66667  0.31657  -2.66667     -2.04 R
 31  3.00000  5.66667  0.31657  -2.66667     -2.04 R
 41  3.00000  6.22222  0.31657  -3.22222     -2.47 R
 50  3.00000  6.22222  0.31657  -3.22222     -2.47 R
 76  3.00000  5.77778  0.44770  -2.77778     -2.19 R
 77  3.00000  5.77778  0.44770  -2.77778     -2.19 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Freshness, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4    5.733    5.733   1.433  0.63  0.641
Error   85  192.889  192.889   2.269
Total   89  198.622


S = 1.50641   R-Sq = 2.89%   R-Sq(adj) = 0.00%
```

```
Unusual Observations for Freshness

Obs  Freshness      Fit  SE Fit  Residual  St Resid
  5    3.00000  6.44444  0.35507  -3.44444     -2.35 R
 14    3.00000  6.44444  0.35507  -3.44444     -2.35 R
 41    3.00000  6.44444  0.35507  -3.44444     -2.35 R
 50    3.00000  6.44444  0.35507  -3.44444     -2.35 R
 76    3.00000  6.11111  0.50214  -3.11111     -2.19 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Overall, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4    1.333    1.333   0.333  0.19  0.942
Error   85  148.222  148.222   1.744
Total   89  149.556


S = 1.32053   R-Sq = 0.89%   R-Sq(adj) = 0.00%


Unusual Observations for Overall

Obs  Overall      Fit  SE Fit  Residual  St Resid
 22  3.00000  6.11111  0.31125  -3.11111     -2.42 R
 31  3.00000  6.11111  0.31125  -3.11111     -2.42 R
 76  3.00000  6.22222  0.44018  -3.22222     -2.59 R

R denotes an observation with a large standardized residual.
```

Week 6

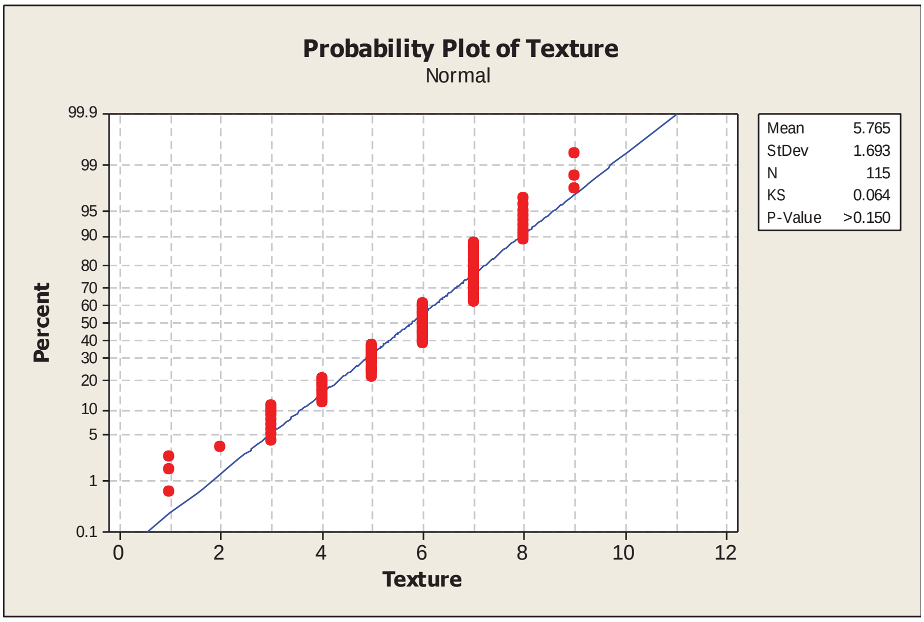
Normality plot for Color



One-way ANOVA: Color versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    1.51  0.38  0.14  0.965
Error   110  287.13  2.61
Total   114  288.64

S = 1.616   R-Sq = 0.52%   R-Sq(adj) = 0.00%


                        Individual 95% CIs For Mean Based on
                        Pooled StDev
Level   N   Mean  StDev  ---+---------+---------+---------+------
126    23  5.870  1.604       (------------*-------------)
342    23  5.783  1.650     (-------------*------------)
561    23  5.522  1.275  (------------*-------------)
799    23  5.739  1.657     (-------------*------------)
853    23  5.739  1.839     (-------------*------------)
                        ---+---------+---------+---------+------
                         5.00      5.50      6.00      6.50

Pooled StDev = 1.616
```

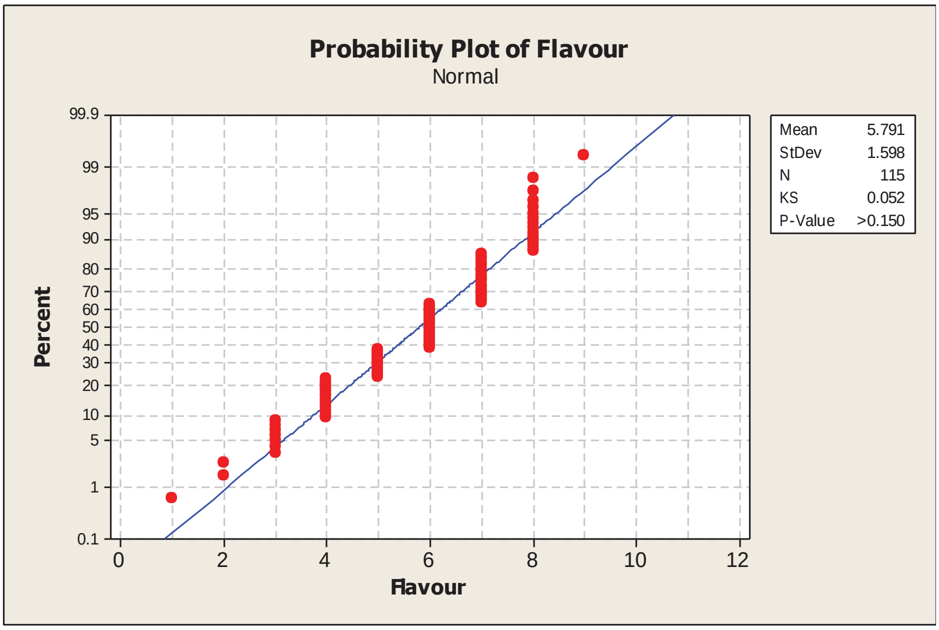
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source   DF      SS    MS     F      P
Sample    4    4.92  1.23  0.42  0.793
Error   110  321.74  2.92
Total   114  326.66

S = 1.710   R-Sq = 1.51%   R-Sq(adj) = 0.00%


                           Individual 95% CIs For Mean Based on
                           Pooled StDev
Level   N   Mean  StDev  -----+---------+---------+---------+----
126    23  6.000  2.000          (-------------*-------------)
342    23  5.957  1.461         (-------------*-------------)
561    23  5.652  1.526     (-------------*-------------)
799    23  5.783  1.808       (-------------*-------------)
853    23  5.435  1.701  (-------------*-------------)
                           -----+---------+---------+---------+----
                              5.00      5.50      6.00      6.50

Pooled StDev = 1.710
```

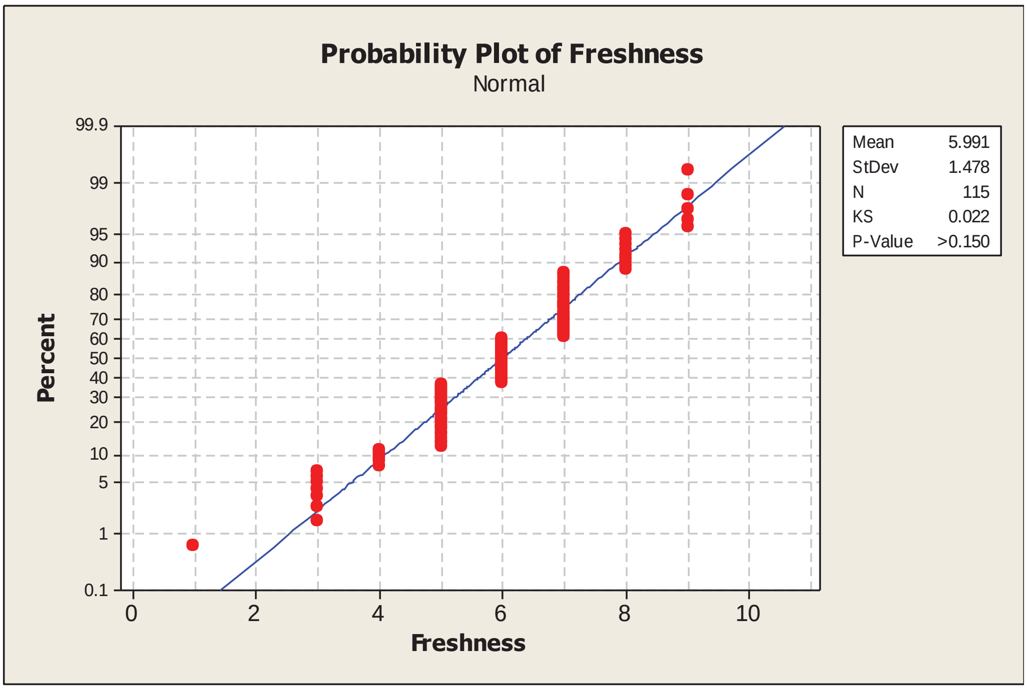
Normality test for Flavour



One-way ANOVA: Flavor versus Sample

```
Source   DF      SS    MS     F      P
Sample    4   15.51  3.88  1.55  0.193
Error   110  275.48  2.50
Total   114  290.99

S = 1.583   R-Sq = 5.33%   R-Sq(adj) = 1.89%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ---+---------+---------+---------+------
126    23  6.043  1.522            (----------*----------)
342    23  5.261  1.888  (----------*----------)
561    23  6.087  1.379             (---------*----------)
799    23  6.130  1.486             (----------*----------)
853    23  5.435  1.590     (----------*---------)
                         ---+---------+---------+---------+------
                          4.80      5.40      6.00      6.60

Pooled StDev = 1.583
```

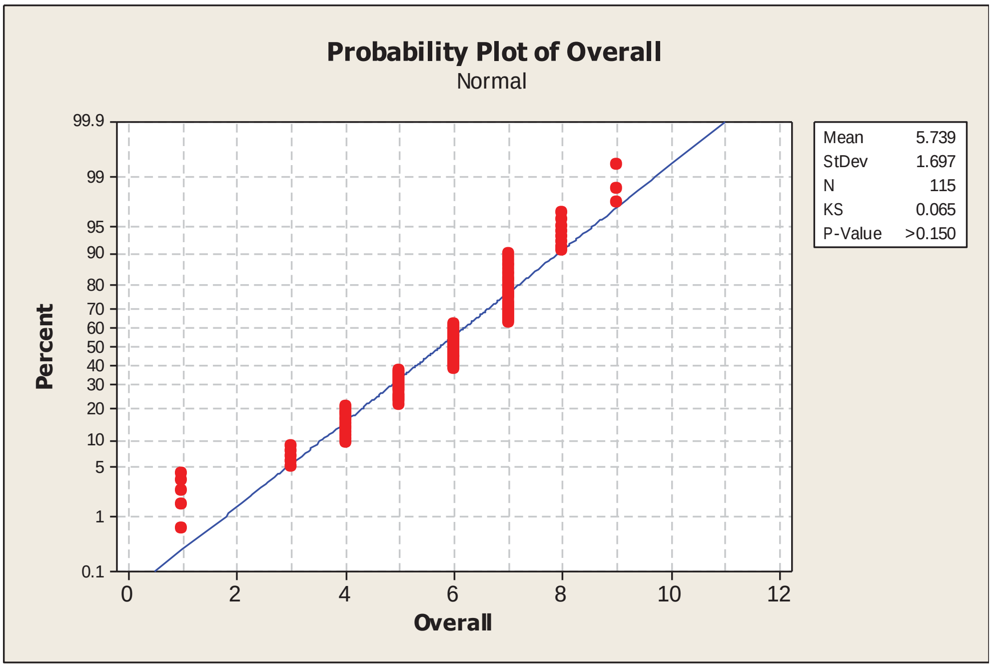
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source   DF     SS    MS     F      P
Sample    4   2.90  0.73  0.32  0.861
Error   110  246.09  2.24
Total   114  248.99

S = 1.496   R-Sq = 1.17%   R-Sq(adj) = 0.00%


                       Individual 95% CIs For Mean Based on
                       Pooled StDev
Level   N   Mean  StDev  --------+---------+---------+---------+-
126    23  6.087  1.474      (------------*-----------)
342    23  5.870  1.660   (-----------*------------)
561    23  6.130  1.014       (------------*-----------)
799    23  6.130  1.486       (------------*-----------)
853    23  5.739  1.738  (------------*-----------)
                       --------+---------+---------+---------+-
                            5.50      6.00      6.50      7.00

Pooled StDev = 1.496
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source   DF      SS    MS     F      P
Sample    4   11.74  2.93  1.02  0.400
Error   110  316.43  2.88
Total   114  328.17

S = 1.696   R-Sq = 3.58%   R-Sq(adj) = 0.07%


                       Individual 95% CIs For Mean Based on
                       Pooled StDev
Level   N   Mean  StDev  -----+---------+---------+---------+----
126    23  6.000  1.508             (-----------*-----------)
342    23  5.217  1.999  (-----------*-----------)
561    23  5.913  1.474            (-----------*----------)
799    23  6.043  1.718              (-----------*----------)
853    23  5.522  1.729       (-----------*-----------)
                       -----+---------+---------+---------+----
                          4.80      5.40      6.00      6.60

Pooled StDev = 1.696
```

General Linear Model: Color, Texture, ... versus Sample

```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF  Seq SS  Adj SS  Adj MS     F      P
Sample   4   1.513   1.513   0.378  0.14  0.965
```

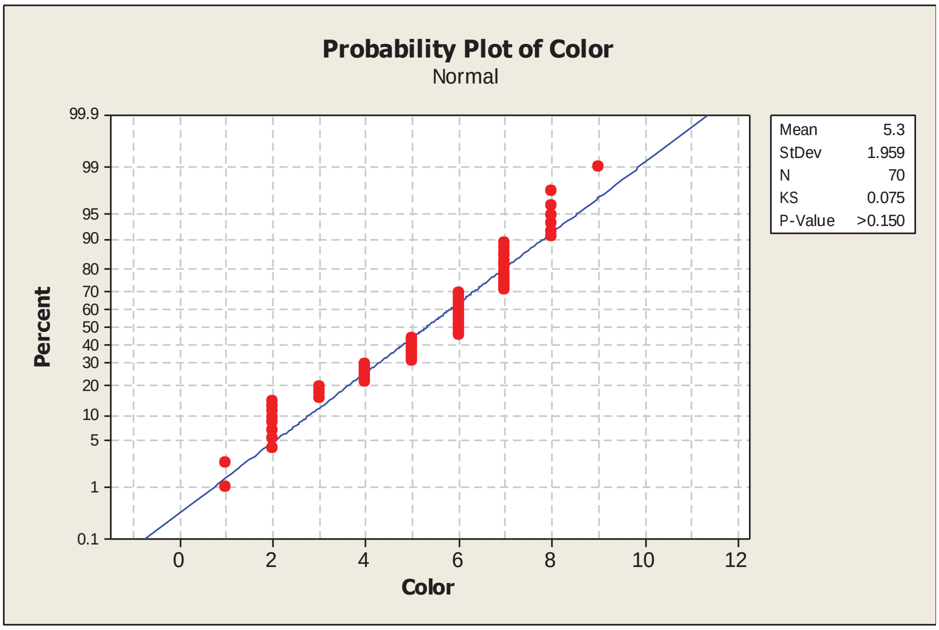
```
Error   110  287.130  287.130   2.610
Total   114  288.643


S = 1.61564   R-Sq = 0.52%   R-Sq(adj) = 0.00%


Unusual Observations for Color

Obs    Color      Fit   SE Fit  Residual  St Resid
 15  1.00000  5.73913  0.33688  -4.73913     -3.00 R
 53  9.00000  5.73913  0.33688   3.26087      2.06 R
 61  1.00000  5.73913  0.33688  -4.73913     -3.00 R
 68  2.00000  5.73913  0.33688  -3.73913     -2.37 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4    4.922    4.922   1.230  0.42  0.793
Error   110  321.739  321.739   2.925
Total   114  326.661


S = 1.71023   R-Sq = 1.51%   R-Sq(adj) = 0.00%


Unusual Observations for Texture

Obs  Texture      Fit   SE Fit  Residual  St Resid
 15  1.00000  5.78261  0.35661  -4.78261     -2.86 R
 37  2.00000  6.00000  0.35661  -4.00000     -2.39 R
 38  1.00000  6.00000  0.35661  -5.00000     -2.99 R
 61  1.00000  5.43478  0.35661  -4.43478     -2.65 R
101  9.00000  5.65217  0.35661   3.34783      2.00 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavor, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4   15.513   15.513   3.878  1.55  0.193
Error   110  275.478  275.478   2.504
Total   114  290.991


S = 1.58251   R-Sq = 5.33%   R-Sq(adj) = 1.89%


Unusual Observations for Flavor

Obs   Flavor      Fit   SE Fit  Residual  St Resid
 15  3.00000  6.13043  0.32998  -3.13043     -2.02 R
 38  2.00000  6.04348  0.32998  -4.04348     -2.61 R
 83  1.00000  5.26087  0.32998  -4.26087     -2.75 R
 84  9.00000  5.26087  0.32998   3.73913      2.42 R
 86  2.00000  5.26087  0.32998  -3.26087     -2.11 R

R denotes an observation with a large standardized residual.
```

```
Analysis of Variance for Freshness, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4    2.904    2.904   0.726  0.32  0.861
Error   110  246.087  246.087   2.237
Total   114  248.991


S = 1.49571   R-Sq = 1.17%   R-Sq(adj) = 0.00%


Unusual Observations for Freshness

Obs  Freshness      Fit  SE Fit  Residual  St Resid
  9    3.00000  6.13043  0.31188  -3.13043     -2.14 R
 38    3.00000  6.08696  0.31188  -3.08696     -2.11 R
 45    3.00000  6.08696  0.31188  -3.08696     -2.11 R
 47    9.00000  5.73913  0.31188   3.26087      2.23 R
 61    1.00000  5.73913  0.31188  -4.73913     -3.24 R
 70    9.00000  5.86957  0.31188   3.13043      2.14 R
 84    9.00000  5.86957  0.31188   3.13043      2.14 R

R denotes an observation with a large standardized residual.
```

```
Analysis of Variance for Overall, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4   11.739   11.739   2.935  1.02  0.400
Error   110  316.435  316.435   2.877
Total   114  328.174


S = 1.69608   R-Sq = 3.58%   R-Sq(adj) = 0.07%


Unusual Observations for Overall

Obs  Overall      Fit   SE Fit  Residual  St Resid
 15  1.00000  6.04348  0.35366  -5.04348     -3.04 R
 38  1.00000  6.00000  0.35366  -5.00000     -3.01 R
 61  1.00000  5.52174  0.35366  -4.52174     -2.73 R
 83  1.00000  5.21739  0.35366  -4.21739     -2.54 R
 84  9.00000  5.21739  0.35366   3.78261      2.28 R
 86  1.00000  5.21739  0.35366  -4.21739     -2.54 R

R denotes an observation with a large standardized residual.
```

Week 8

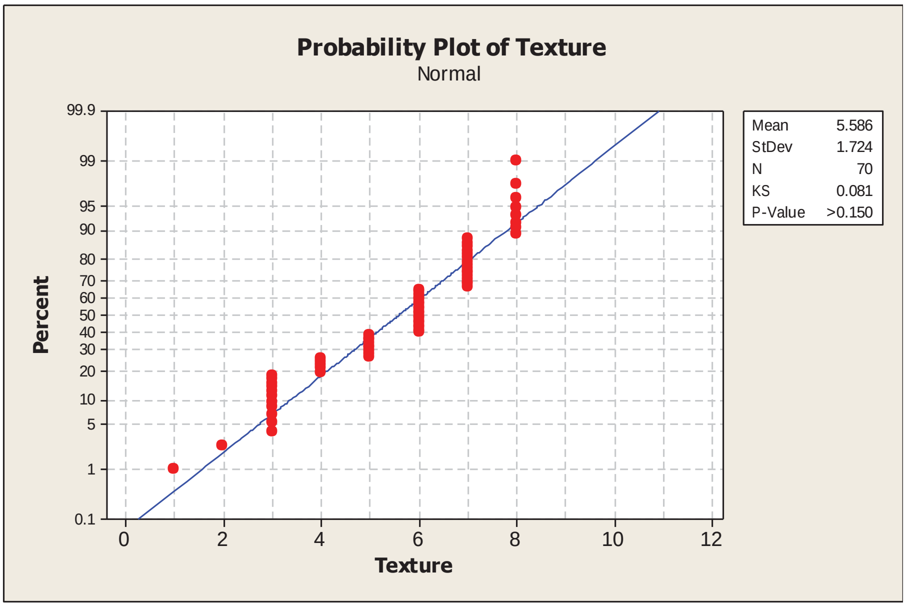
Normality plot for Color



One-way ANOVA: Color versus Sample

```
Source   DF      SS     MS     F      P
Sample    4   81.83  20.46  6.17  0.000
Error   135  447.57   3.32
Total   139  529.40

S = 1.821   R-Sq = 15.46%   R-Sq(adj) = 12.95%


                           Individual 95% CIs For Mean Based on
                           Pooled StDev
Level   N   Mean  StDev  ------+---------+---------+---------+---
126    28  4.071  2.089  (------*------)
342    28  6.143  1.380                 (-----*------)
561    28  4.857  1.995         (------*-----)
799    28  5.429  1.874            (------*------)
853    28  6.000  1.678                (------*------)
                         ------+---------+---------+---------+---
                             4.0       5.0       6.0       7.0

Pooled StDev = 1.821
```

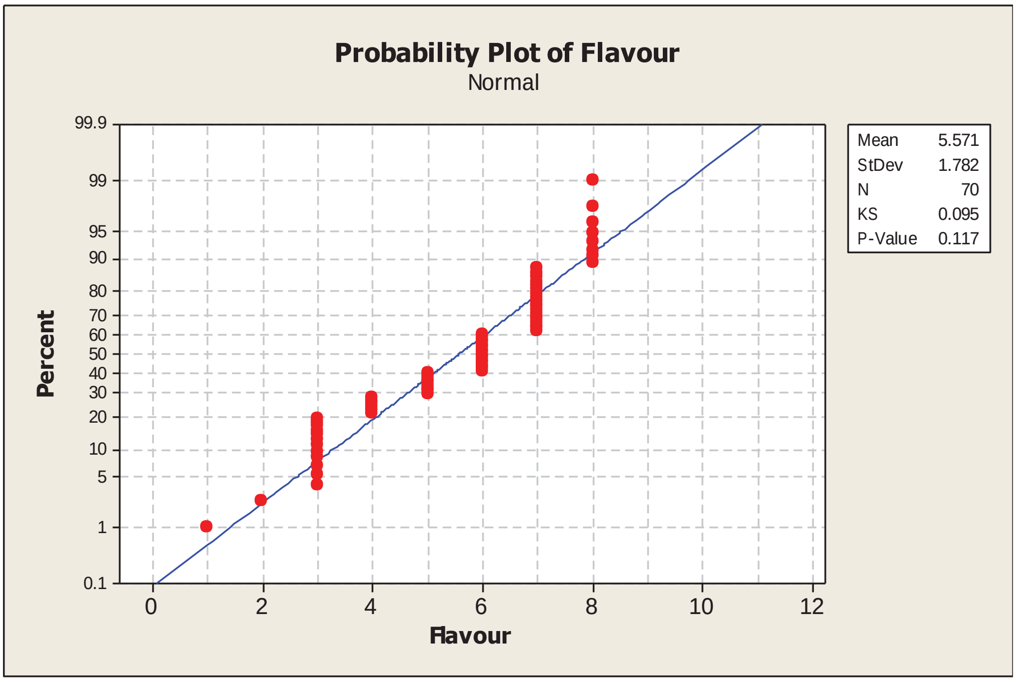
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source   DF      SS    MS     F      P
Sample    4   27.26  6.81  2.40  0.053
Error   135  382.71  2.83
Total   139  409.97

S = 1.684   R-Sq = 6.65%   R-Sq(adj) = 3.88%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    28  5.714  1.652             (--------*--------)
342    28  5.786  1.166              (--------*--------)
561    28  4.714  1.902  (--------*--------)
799    28  5.929  1.609                (--------*--------)
853    28  5.786  1.969              (--------*--------)
                         --+---------+---------+---------+-------
                         4.20      4.90      5.60      6.30

Pooled StDev = 1.684
```

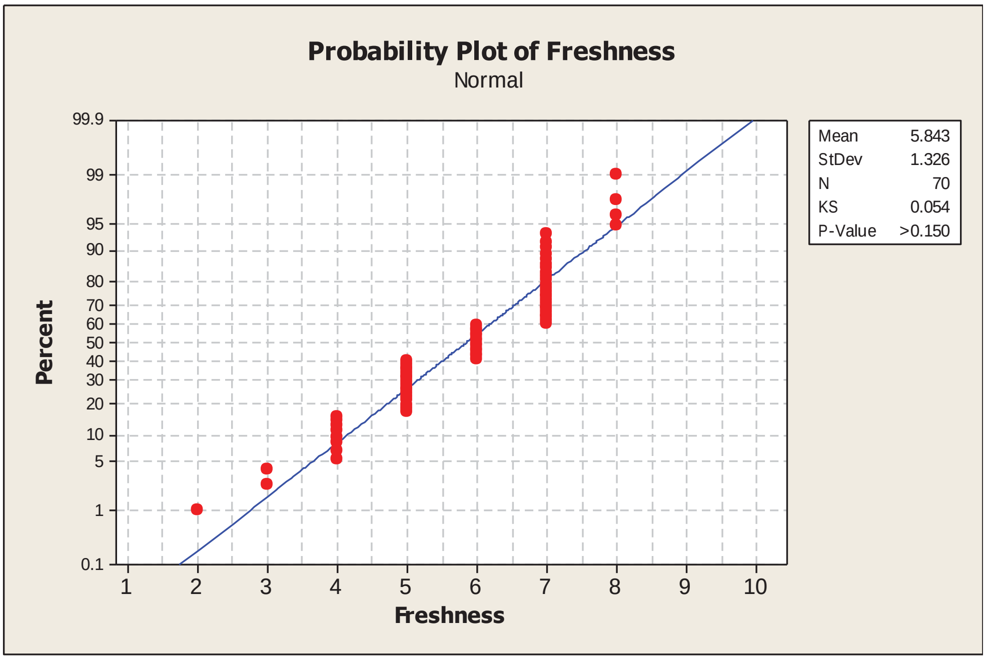
Normality plot for Flavour



One-way ANOVA: Flavour versus Sample

```
Source   DF      SS     MS     F      P
Sample    4   63.14  15.79  5.68  0.000
Error   135  375.14   2.78
Total   139  438.29

S = 1.667   R-Sq = 14.41%   R-Sq(adj) = 11.87%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    28  5.500  1.795             (-------*-------)
342    28  5.429  1.526            (-------*-------)
561    28  4.429  1.834  (------*-------)
799    28  6.286  1.560                        (-------*------)
853    28  6.214  1.595                       (-------*------)
                         --+---------+---------+---------+-------
                          4.00      4.80      5.60      6.40

Pooled StDev = 1.667
```

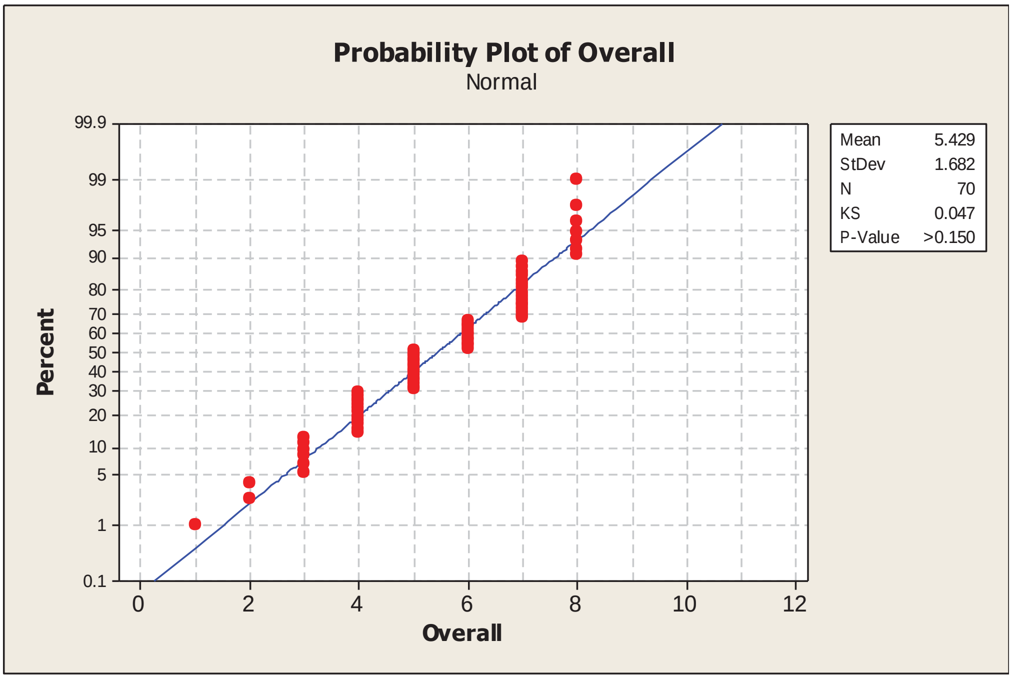
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source   DF      SS    MS     F      P
Sample    4   29.83  7.46  4.73  0.001
Error   135  212.71  1.58
Total   139  242.54

S = 1.255   R-Sq = 12.30%   R-Sq(adj) = 9.70%


                       Individual 95% CIs For Mean Based on
                       Pooled StDev
Level   N   Mean  StDev  ----+---------+---------+---------+-----
126    28  6.071  1.052              (-------*-------)
342    28  5.714  1.243           (-------*-------)
561    28  5.000  1.540  (------*-------)
799    28  6.143  1.533               (------*-------)
853    28  6.286  0.713                (-------*-------)
                         ----+---------+---------+---------+-----
                           4.80      5.40      6.00      6.60

Pooled StDev = 1.255
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source   DF      SS     MS     F      P
Sample    4   44.00  11.00  4.29  0.003
Error   135  346.29   2.57
Total   139  390.29

S = 1.602   R-Sq = 11.27%   R-Sq(adj) = 8.64%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ----+---------+---------+---------+-----
126    28  5.214  1.686        (-------*--------)
342    28  5.429  1.372           (--------*-------)
561    28  4.500  1.711  (-------*--------)
799    28  5.929  1.654                 (--------*-------)
853    28  6.071  1.562                   (--------*-------)
                         ----+---------+---------+---------+-----
                           4.20      4.90      5.60      6.30

Pooled StDev = 1.602
```

General Linear Model: Color, Texture, ... versus Sample

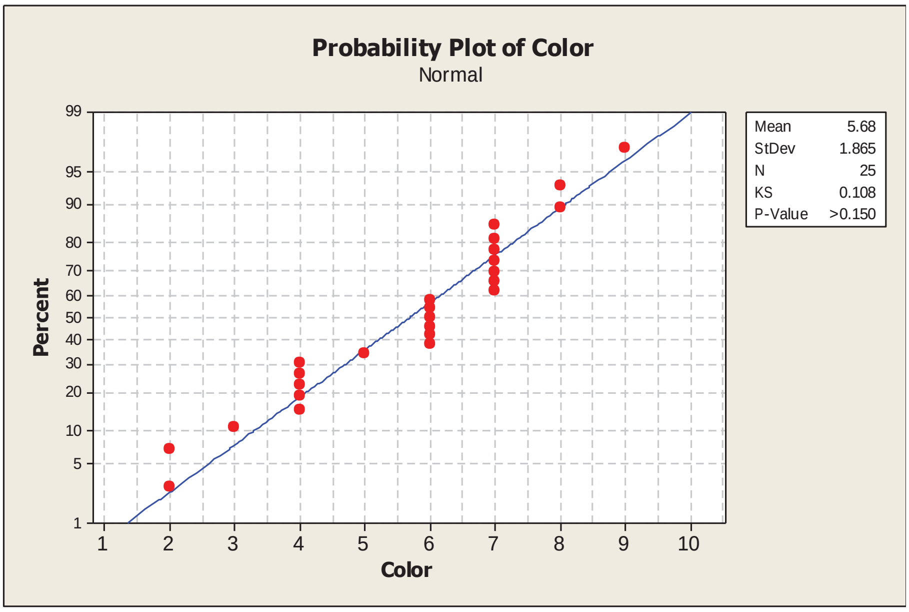
```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF  Seq SS   Adj SS  Adj MS     F      P
Sample   4  81.829   81.829  20.457  6.17  0.000
```

```
Error   135  447.571  447.571   3.315
Total   139  529.400


S = 1.82081   R-Sq = 15.46%   R-Sq(adj) = 12.95%


Unusual Observations for Color

Obs    Color      Fit   SE Fit  Residual  St Resid
 31  8.00000  4.07143  0.34410   3.92857      2.20 R
 45  8.00000  4.07143  0.34410   3.92857      2.20 R
 57  2.00000  6.00000  0.34410  -4.00000     -2.24 R
 71  2.00000  6.00000  0.34410  -4.00000     -2.24 R
113  1.00000  4.85714  0.34410  -3.85714     -2.16 R
127  1.00000  4.85714  0.34410  -3.85714     -2.16 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4   27.257   27.257   6.814  2.40  0.053
Error   135  382.714  382.714   2.835
Total   139  409.971


S = 1.68372   R-Sq = 6.65%   R-Sq(adj) = 3.88%


Unusual Observations for Texture

Obs  Texture      Fit   SE Fit  Residual  St Resid
 65  1.00000  5.78571  0.31819  -4.78571     -2.89 R
 79  1.00000  5.78571  0.31819  -4.78571     -2.89 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavour, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4   63.143   63.143  15.786  5.68  0.000
Error   135  375.143  375.143   2.779
Total   139  438.286


S = 1.66698   R-Sq = 14.41%   R-Sq(adj) = 11.87%


Unusual Observations for Flavour

Obs  Flavour      Fit   SE Fit  Residual  St Resid
  1  3.00000  6.28571  0.31503  -3.28571     -2.01 R
  6  3.00000  6.28571  0.31503  -3.28571     -2.01 R
 15  3.00000  6.28571  0.31503  -3.28571     -2.01 R
 20  3.00000  6.28571  0.31503  -3.28571     -2.01 R
 34  2.00000  5.50000  0.31503  -3.50000     -2.14 R
 48  2.00000  5.50000  0.31503  -3.50000     -2.14 R
115  8.00000  4.42857  0.31503   3.57143      2.18 R
118  1.00000  4.42857  0.31503  -3.42857     -2.09 R
129  8.00000  4.42857  0.31503   3.57143      2.18 R
```

```
132  1.00000  4.42857  0.31503  -3.42857     -2.09 R

R denotes an observation with a large standardized residual.
\
Analysis of Variance for Freshness, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4   29.829   29.829   7.457  4.73  0.001
Error   135  212.714  212.714   1.576
Total   139  242.543


S = 1.25525   R-Sq = 12.30%   R-Sq(adj) = 9.70%


Unusual Observations for Freshness

Obs  Freshness      Fit   SE Fit  Residual  St Resid
  1    3.00000  6.14286  0.23722  -3.14286     -2.55 R
 15    3.00000  6.14286  0.23722  -3.14286     -2.55 R
 98    3.00000  5.71429  0.23722  -2.71429     -2.20 R
112    3.00000  5.71429  0.23722  -2.71429     -2.20 R
115    8.00000  5.00000  0.23722   3.00000      2.43 R
126    2.00000  5.00000  0.23722  -3.00000     -2.43 R
129    8.00000  5.00000  0.23722   3.00000      2.43 R
140    2.00000  5.00000  0.23722  -3.00000     -2.43 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Overall, using Adjusted SS for Tests

Source   DF   Seq SS   Adj SS  Adj MS     F      P
Sample    4   44.000   44.000  11.000  4.29  0.003
Error   135  346.286  346.286   2.565
Total   139  390.286


S = 1.60159   R-Sq = 11.27%   R-Sq(adj) = 8.64%

Unusual Observations for Overall

Obs  Overall      Fit   SE Fit  Residual  St Resid
  1  2.00000  5.92857  0.30267  -3.92857     -2.50 R
 15  2.00000  5.92857  0.30267  -3.92857     -2.50 R
 30  2.00000  5.21429  0.30267  -3.21429     -2.04 R
 44  2.00000  5.21429  0.30267  -3.21429     -2.04 R
115  8.00000  4.50000  0.30267   3.50000      2.23 R
118  1.00000  4.50000  0.30267  -3.50000     -2.23 R
129  8.00000  4.50000  0.30267   3.50000      2.23 R
132  1.00000  4.50000  0.30267  -3.50000     -2.23 R

R denotes an observation with a large standardized residual.
```

Week 10

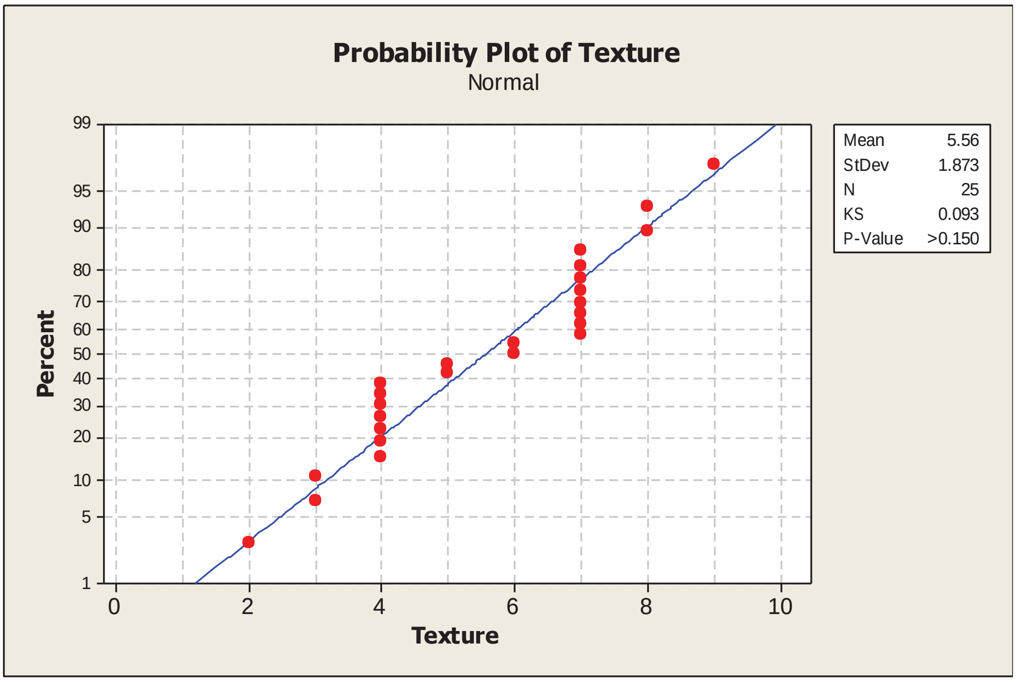
Normality plot for Color



One-way ANOVA: Color versus Sample

```
Source  DF      SS    MS     F      P
Sample   4   36.16  9.04  2.89  0.026
Error   95  297.60  3.13
Total   99  333.76

S = 1.770   R-Sq = 10.83%   R-Sq(adj) = 7.08%


                         Individual 95% CIs For Mean Based on Pooled StDev
Level   N   Mean  StDev   +---------+---------+---------+---------
126    20  5.400  1.789        (-------*-------)
342    20  4.800  1.508   (-------*-------)
561    20  6.600  2.479                  (-------*-------)
799    20  5.600  1.392          (-------*-------)
853    20  6.000  1.451             (-------*-------)
                          +---------+---------+---------+---------
                        4.0       5.0       6.0       7.0

Pooled StDev = 1.770
```

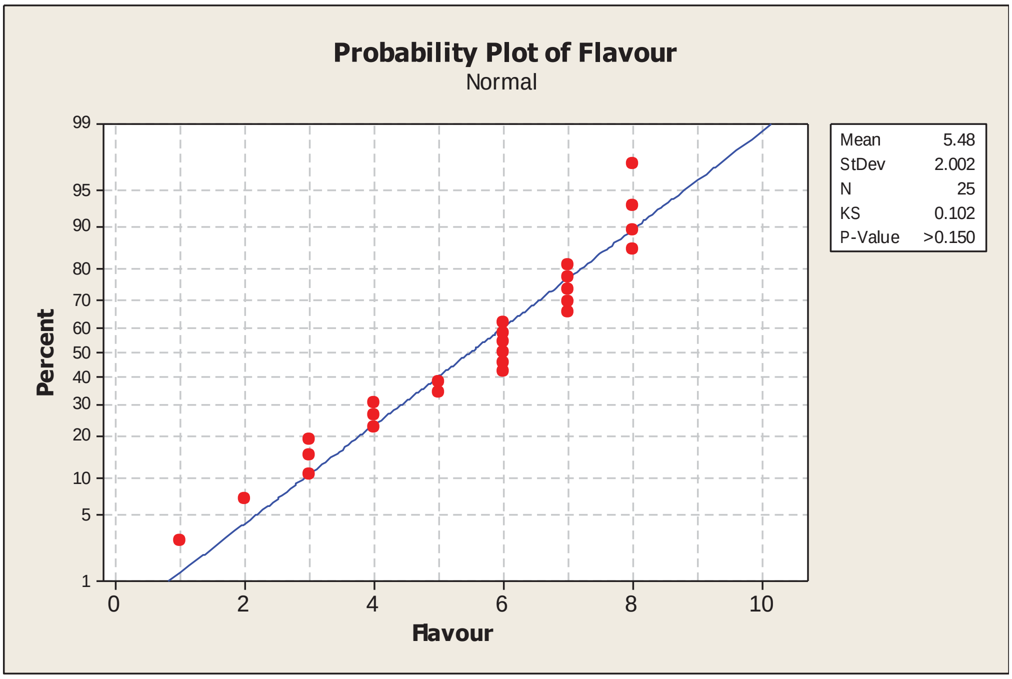
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source  DF      SS     MS     F      P
Sample   4   88.64  22.16  8.49  0.000
Error   95  248.00   2.61
Total   99  336.64

S = 1.616   R-Sq = 26.33%   R-Sq(adj) = 23.23%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ---+---------+---------+---------+------
126    20  6.200  1.765                 (-----*-----)
342    20  4.000  1.717  (-----*-----)
561    20  5.000  1.717           (-----*-----)
799    20  6.000  1.451                (-----*-----)
853    20  6.600  1.392                     (-----*-----)
                         ---+---------+---------+---------+------
                          3.6       4.8       6.0       7.2

Pooled StDev = 1.616
```

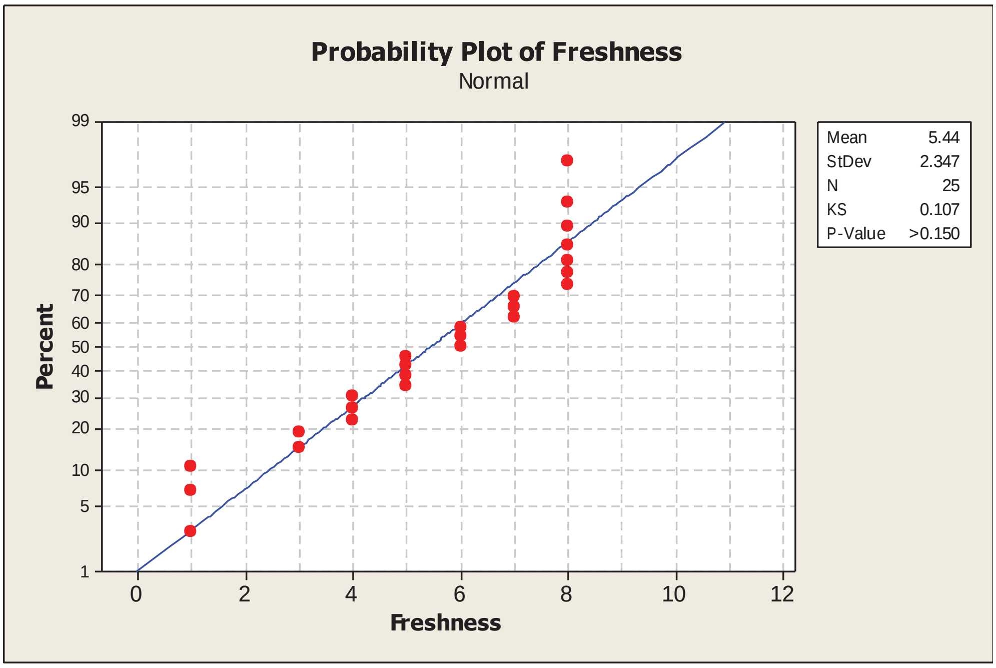
Normality plot for Flavour



One-way ANOVA: Flavour versus Sample

```
Source  DF      SS     MS     F      P
Sample   4  100.16  25.04  8.35  0.000
Error   95  284.80   3.00
Total   99  384.96

S = 1.731   R-Sq = 26.02%   R-Sq(adj) = 22.90%


                       Individual 95% CIs For Mean Based on
                       Pooled StDev
Level   N   Mean  StDev  ------+---------+---------+---------+---
126    20  5.800  2.093             (-------*-------)
342    20  4.400  1.789  (-------*-------)
561    20  4.200  2.191 (-------*-------)
799    20  6.400  0.503                  (-------*-------)
853    20  6.600  1.536                    (-------*-------)
                       ------+---------+---------+---------+---
                           4.0       5.0       6.0       7.0

Pooled StDev = 1.731
```

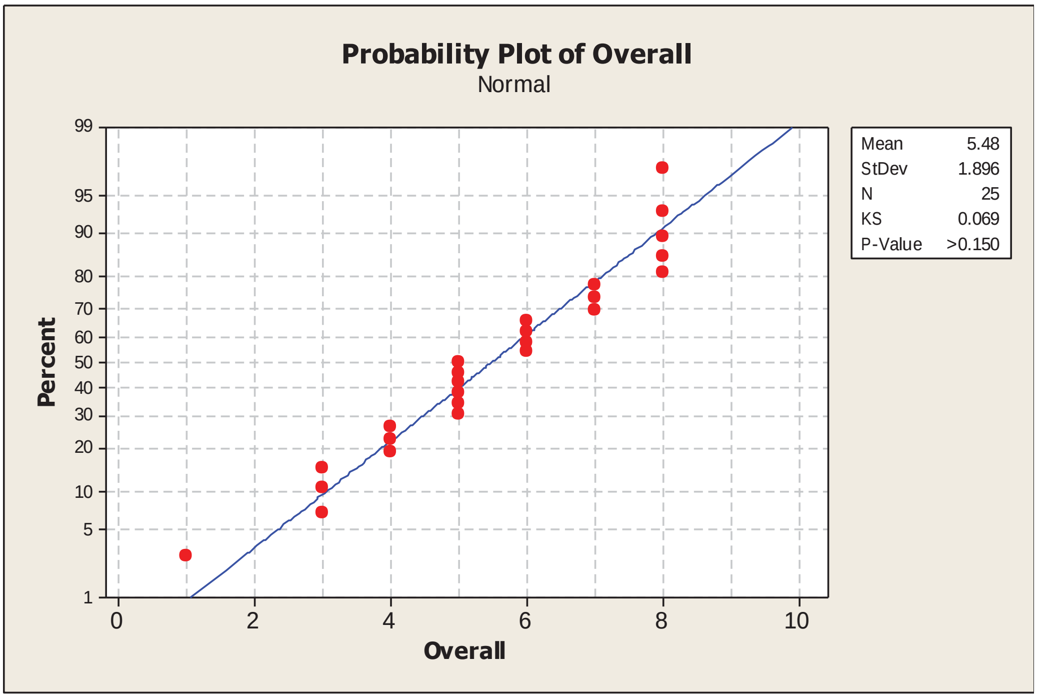
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source  DF      SS     MS     F      P
Sample   4   91.84  22.96  4.99  0.001
Error   95  436.80   4.60
Total   99  528.64

S = 2.144   R-Sq = 17.37%   R-Sq(adj) = 13.89%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  -----+---------+---------+---------+----
126    20  6.200  1.989                (-------*-------)
342    20  4.000  2.828  (-------*-------)
561    20  4.600  2.479       (-------*-------)
799    20  6.000  1.717                 (-------*-------)
853    20  6.400  1.392                    (-------*-------)
                         -----+---------+---------+---------+----
                            3.6       4.8       6.0       7.2

Pooled StDev = 2.144
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source  DF      SS     MS     F      P
Sample   4   58.56  14.64  4.86  0.001
Error   95  286.40   3.01
Total   99  344.96

S = 1.736   R-Sq = 16.98%   R-Sq(adj) = 13.48%


                           Individual 95% CIs For Mean Based on
                           Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    20  6.200  1.642               (-------*-------)
342    20  4.600  1.903  (-------*-------)
561    20  4.600  2.479  (-------*-------)
799    20  5.600  1.231          (-------*-------)
853    20  6.400  1.046                 (-------*-------)
                           --+---------+---------+---------+-------
                           4.0       5.0       6.0       7.0

Pooled StDev = 1.736
```

General Linear Model: Color, Texture, ... versus Sample

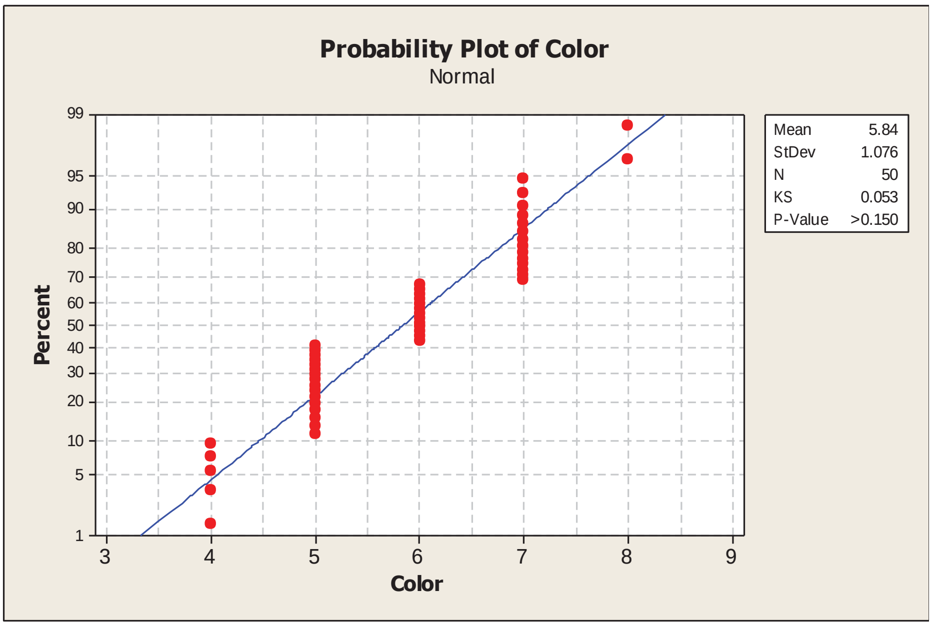
```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   36.160   36.160   9.040  2.89  0.026
Error   95  297.600  297.600   3.133
```

```
Total   99  333.760


S = 1.76992   R-Sq = 10.83%   R-Sq(adj) = 7.08%


Unusual Observations for Color

Obs    Color      Fit   SE Fit  Residual  St Resid
 85  2.00000  6.60000  0.39577  -4.60000     -2.67 R
 90  2.00000  6.60000  0.39577  -4.60000     -2.67 R
 95  2.00000  6.60000  0.39577  -4.60000     -2.67 R
100  2.00000  6.60000  0.39577  -4.60000     -2.67 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   88.640   88.640  22.160  8.49  0.000
Error   95  248.000  248.000   2.611
Total   99  336.640


S = 1.61571   R-Sq = 26.33%   R-Sq(adj) = 23.23%


Analysis of Variance for Flavour, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4  100.160  100.160  25.040  8.35  0.000
Error   95  284.800  284.800   2.998
Total   99  384.960


S = 1.73144   R-Sq = 26.02%   R-Sq(adj) = 22.90%


Analysis of Variance for Freshness, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   91.840   91.840  22.960  4.99  0.001
Error   95  436.800  436.800   4.598
Total   99  528.640


S = 2.14427   R-Sq = 17.37%   R-Sq(adj) = 13.89%


Analysis of Variance for Overall, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   58.560   58.560  14.640  4.86  0.001
Error   95  286.400  286.400   3.015
Total   99  344.960


S = 1.73630   R-Sq = 16.98%   R-Sq(adj) = 13.48%


Unusual Observations for Overall
```

```
Obs  Overall     Fit   SE Fit  Residual  St Resid
 64  8.00000  4.60000  0.38825   3.40000      2.01 R
 69  8.00000  4.60000  0.38825   3.40000      2.01 R
 74  8.00000  4.60000  0.38825   3.40000      2.01 R
 79  8.00000  4.60000  0.38825   3.40000      2.01 R
 84  8.00000  4.60000  0.38825   3.40000      2.01 R
 85  1.00000  4.60000  0.38825  -3.60000     -2.13 R
 89  8.00000  4.60000  0.38825   3.40000      2.01 R
 90  1.00000  4.60000  0.38825  -3.60000     -2.13 R
 94  8.00000  4.60000  0.38825   3.40000      2.01 R
 95  1.00000  4.60000  0.38825  -3.60000     -2.13 R
 99  8.00000  4.60000  0.38825   3.40000      2.01 R
100  1.00000  4.60000  0.38825  -3.60000     -2.13 R

R denotes an observation with a large standardized residual.
```

Week 12

Normality plot for Color

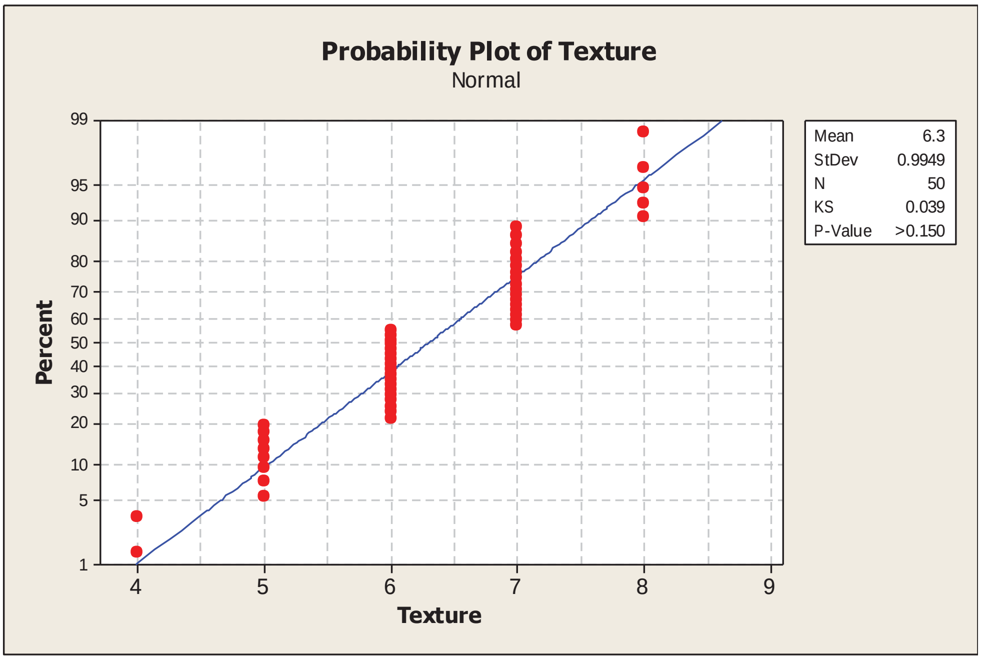


One-way ANOVA: Color versus Sample

```
Source  DF      SS    MS     F      P
Sample   4    3.44  0.86  0.74  0.565
Error   95  110.00  1.16
Total   99  113.44

S = 1.076   R-Sq = 3.03%   R-Sq(adj) = 0.00%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    20  6.100  1.071            (----------*-----------)
342    20  5.800  1.105       (-----------*-----------)
561    20  5.600  1.231  (-----------*-----------)
799    20  6.000  0.918          (-----------*-----------)
```

```
853    20  5.700  1.031      (----------*-----------)
                        --+---------+---------+---------+-------
                        5.20      5.60      6.00      6.40

Pooled StDev = 1.076
```

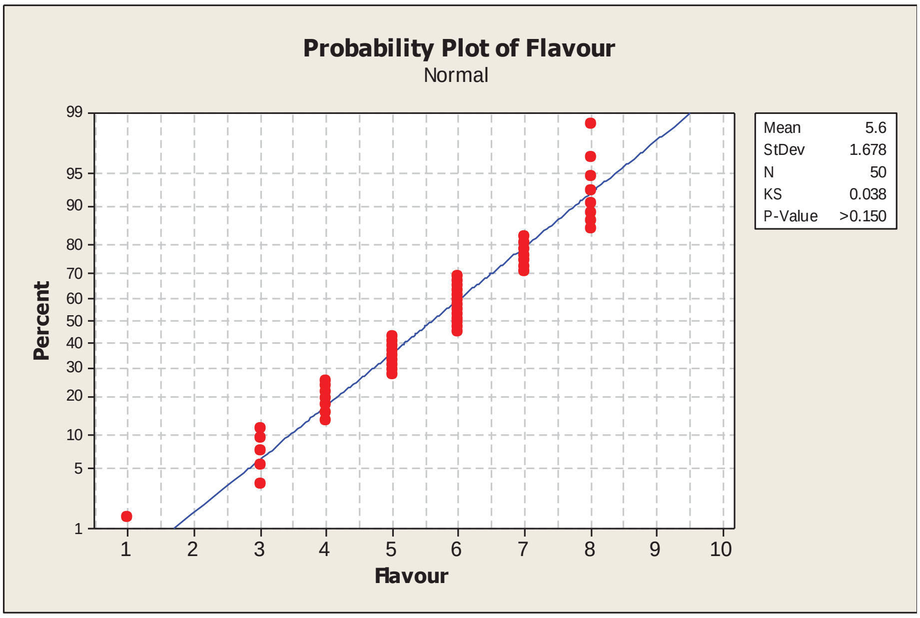
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source  DF      SS     MS     F      P
Sample   4   4.800  1.200  1.24  0.301
Error   95  92.200  0.971
Total   99  97.000

S = 0.9852   R-Sq = 4.95%   R-Sq(adj) = 0.95%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N    Mean   StDev   -+---------+---------+---------+--------
126    20  6.4000  0.8208           (----------*----------)
342    20  6.1000  1.3338      (---------*----------)
561    20  6.4000  1.1425           (----------*----------)
799    20  6.6000  0.8208               (----------*----------)
853    20  6.0000  0.6489   (----------*----------)
                          -+---------+---------+---------+--------
                         5.60      6.00      6.40      6.80

Pooled StDev = 0.9852
```

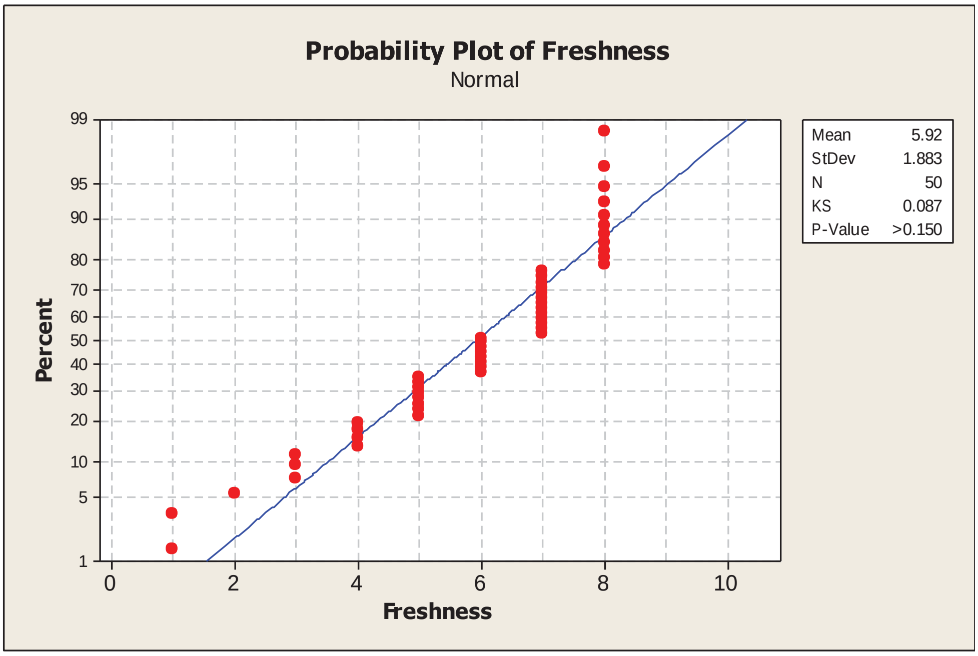
Normality plot for Flavour



One-way ANOVA: Flavour versus Sample

```
Source  DF      SS     MS      F      P
Sample   4  130.40  32.60  21.27  0.000
Error   95  145.60   1.53
Total   99  276.00

S = 1.238   R-Sq = 47.25%   R-Sq(adj) = 45.03%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    20  5.900  1.071                (---*----)
342    20  4.900  1.971         (----*---)
561    20  3.900  0.852  (----*---)
799    20  7.300  0.801                          (----*---)
853    20  6.000  1.124                (----*----)
                         --+---------+---------+---------+-------
                         3.6       4.8       6.0       7.2

Pooled StDev = 1.238
```

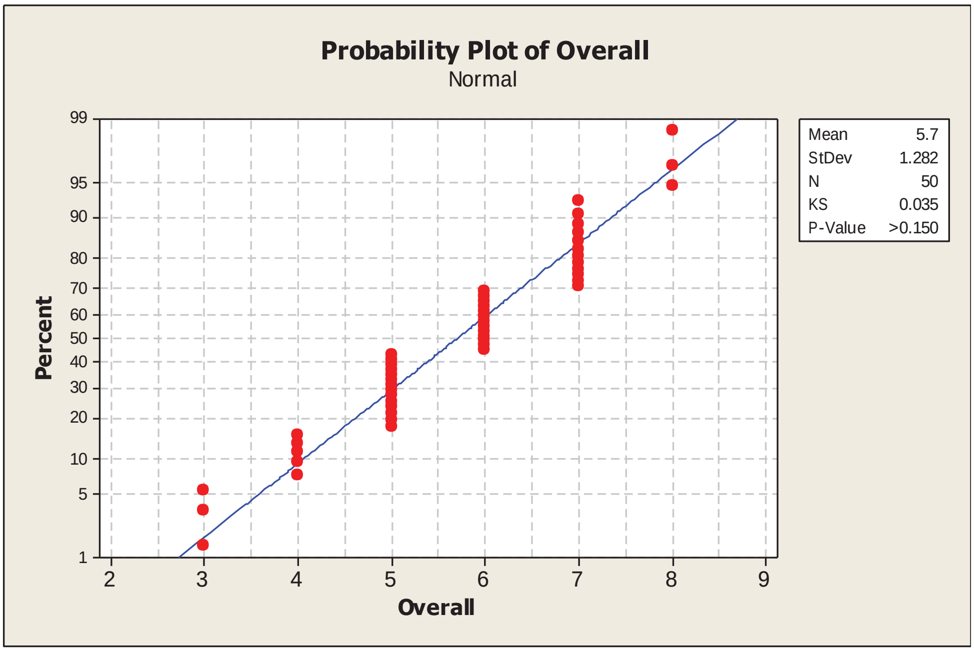
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source  DF      SS     MS      F      P
Sample   4  140.16  35.04  16.07  0.000
Error   95  207.20   2.18
Total   99  347.36

S = 1.477   R-Sq = 40.35%   R-Sq(adj) = 37.84%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  -+---------+---------+---------+--------
126    20  6.700  1.129                 (-----*----)
342    20  5.000  2.103        (-----*----)
561    20  4.100  1.683  (----*-----)
799    20  7.300  1.218                      (-----*----)
853    20  6.500  0.946                (----*-----)
                         -+---------+---------+---------+--------
                         3.6       4.8       6.0       7.2

Pooled StDev = 1.477
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source  DF      SS     MS      F      P
Sample   4   65.20  16.30  16.16  0.000
Error   95   95.80   1.01
Total   99  161.00

S = 1.004   R-Sq = 40.50%   R-Sq(adj) = 37.99%


                        Individual 95% CIs For Mean Based on
                        Pooled StDev
Level   N   Mean  StDev  --+---------+---------+---------+-------
126    20  6.300  1.031                        (-----*----)
342    20  5.400  1.392              (-----*----)
561    20  4.300  0.923  (-----*----)
799    20  6.600  0.821                            (-----*----)
853    20  5.900  0.718                   (-----*----)
                        --+---------+---------+---------+-------
                        4.00      4.80      5.60      6.40

Pooled StDev = 1.004
```

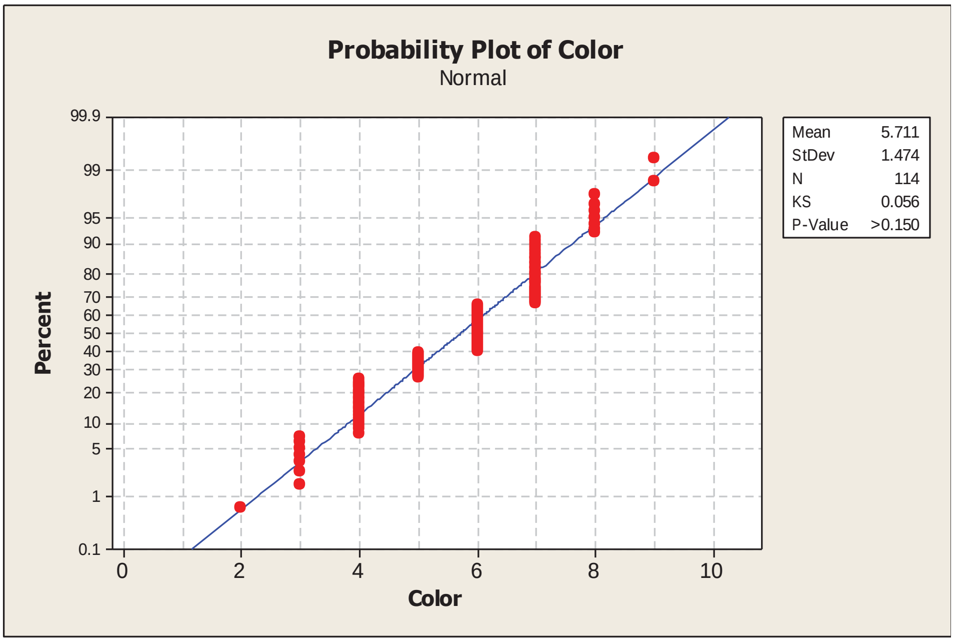
General Linear Model: Color, Texture, ... versus Sample

```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4    3.440    3.440   0.860  0.74  0.565
Error   95  110.000  110.000   1.158
Total   99  113.440
```

```
S = 1.07606   R-Sq = 3.03%   R-Sq(adj) = 0.00%


Unusual Observations for Color

Obs    Color      Fit  SE Fit  Residual  St Resid
 81  8.00000  5.60000  0.24061   2.40000      2.29 R
 91  8.00000  5.60000  0.24061   2.40000      2.29 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   4.8000   4.8000  1.2000  1.24  0.301
Error   95  92.2000  92.2000  0.9705
Total   99  97.0000


S = 0.985153   R-Sq = 4.95%   R-Sq(adj) = 0.95%


Unusual Observations for Texture

Obs  Texture      Fit   SE Fit  Residual  St Resid
 68  4.00000  6.10000  0.22029  -2.10000     -2.19 R
 78  4.00000  6.10000  0.22029  -2.10000     -2.19 R
 83  4.00000  6.40000  0.22029  -2.40000     -2.50 R
 93  4.00000  6.40000  0.22029  -2.40000     -2.50 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavour, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  130.400  130.400  32.600  21.27  0.000
Error   95  145.600  145.600   1.533
Total   99  276.000


S = 1.23799   R-Sq = 47.25%   R-Sq(adj) = 45.03%


Unusual Observations for Flavour

Obs  Flavour      Fit   SE Fit  Residual  St Resid
 62  8.00000  4.90000  0.27682   3.10000      2.57 R
 70  1.00000  4.90000  0.27682  -3.90000     -3.23 R
 72  8.00000  4.90000  0.27682   3.10000      2.57 R
 80  1.00000  4.90000  0.27682  -3.90000     -3.23 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Freshness, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  140.160  140.160  35.040  16.07  0.000
Error   95  207.200  207.200   2.181
Total   99  347.360
```

```
S = 1.47684   R-Sq = 40.35%   R-Sq(adj) = 37.84%


Unusual Observations for Freshness

Obs  Freshness      Fit   SE Fit  Residual  St Resid
  2    4.00000  7.30000  0.33023  -3.30000     -2.29 R
 12    4.00000  7.30000  0.33023  -3.30000     -2.29 R
 62    8.00000  5.00000  0.33023   3.00000      2.08 R
 69    2.00000  5.00000  0.33023  -3.00000     -2.08 R
 70    1.00000  5.00000  0.33023  -4.00000     -2.78 R
 72    8.00000  5.00000  0.33023   3.00000      2.08 R
 79    2.00000  5.00000  0.33023  -3.00000     -2.08 R
 80    1.00000  5.00000  0.33023  -4.00000     -2.78 R
 84    7.00000  4.10000  0.33023   2.90000      2.01 R
 90    1.00000  4.10000  0.33023  -3.10000     -2.15 R
 94    7.00000  4.10000  0.33023   2.90000      2.01 R
100    1.00000  4.10000  0.33023  -3.10000     -2.15 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Overall, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4   65.200   65.200  16.300  16.16  0.000
Error   95   95.800   95.800   1.008
Total   99  161.000


S = 1.00420   R-Sq = 40.50%   R-Sq(adj) = 37.99%


Unusual Observations for Overall

Obs  Overall      Fit   SE Fit  Residual  St Resid
 62  8.00000  5.40000  0.22455   2.60000      2.66 R
 70  3.00000  5.40000  0.22455  -2.40000     -2.45 R
 72  8.00000  5.40000  0.22455   2.60000      2.66 R
 80  3.00000  5.40000  0.22455  -2.40000     -2.45 R

R denotes an observation with a large standardized residual.
```

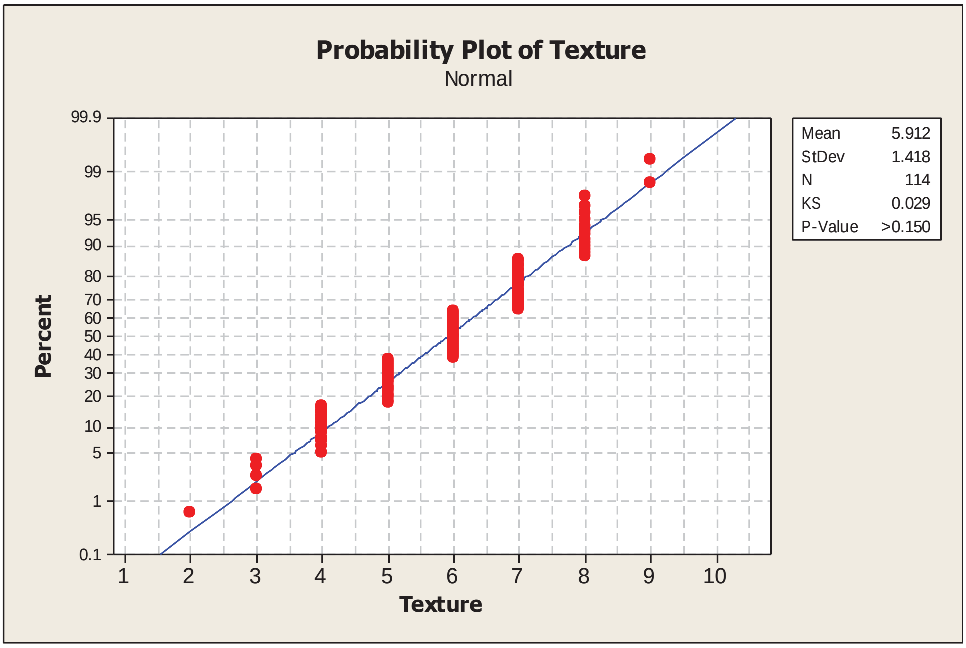
Normality plot for Color



One-way ANOVA: Color versus Sample

```
Source  DF      SS    MS     F      P
Sample   4    7.76  1.94  1.81  0.134
Error   95  102.00  1.07
Total   99  109.76

S = 1.036   R-Sq = 7.07%   R-Sq(adj) = 3.16%


                       Individual 95% CIs For Mean Based on
                       Pooled StDev
Level   N   Mean  StDev  ---+---------+---------+---------+------
126    20  5.800  1.005        (--------*--------)
342    20  5.400  1.142  (--------*--------)
561    20  6.000  1.214           (--------*--------)
799    20  5.900  0.968         (--------*--------)
853    20  5.300  0.801  (--------*--------)
                       ---+---------+---------+---------+------
                        5.00      5.50      6.00      6.50

Pooled StDev = 1.036
```

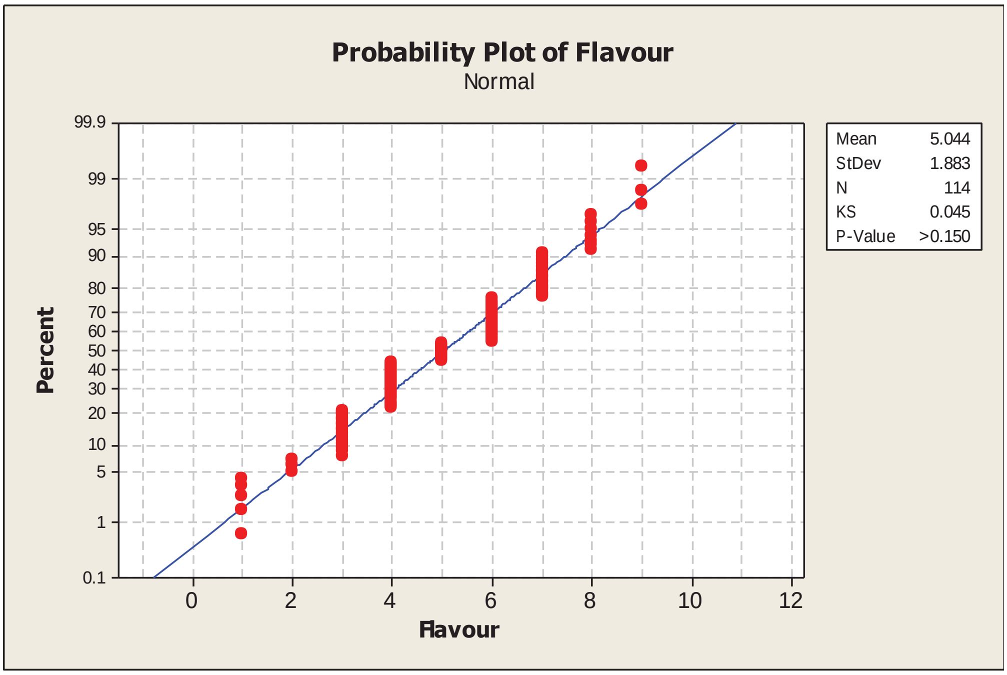
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source  DF      SS     MS     F      P
Sample   4  20.960  5.240  6.69  0.000
Error   95  74.400  0.783
Total   99  95.360

S = 0.8850   R-Sq = 21.98%   R-Sq(adj) = 18.69%


                           Individual 95% CIs For Mean Based on
                           Pooled StDev
Level   N    Mean   StDev  -----+---------+---------+---------+----
126    20  6.3000  0.9234              (------*------)
342    20  5.7000  1.2183    (------*------)
561    20  6.1000  0.7182          (------*-----)
799    20  6.8000  0.8944                        (-----*------)
853    20  5.5000  0.5130  (------*-----)
                           -----+---------+---------+---------+----
                              5.40      6.00      6.60      7.20

Pooled StDev = 0.8850
```

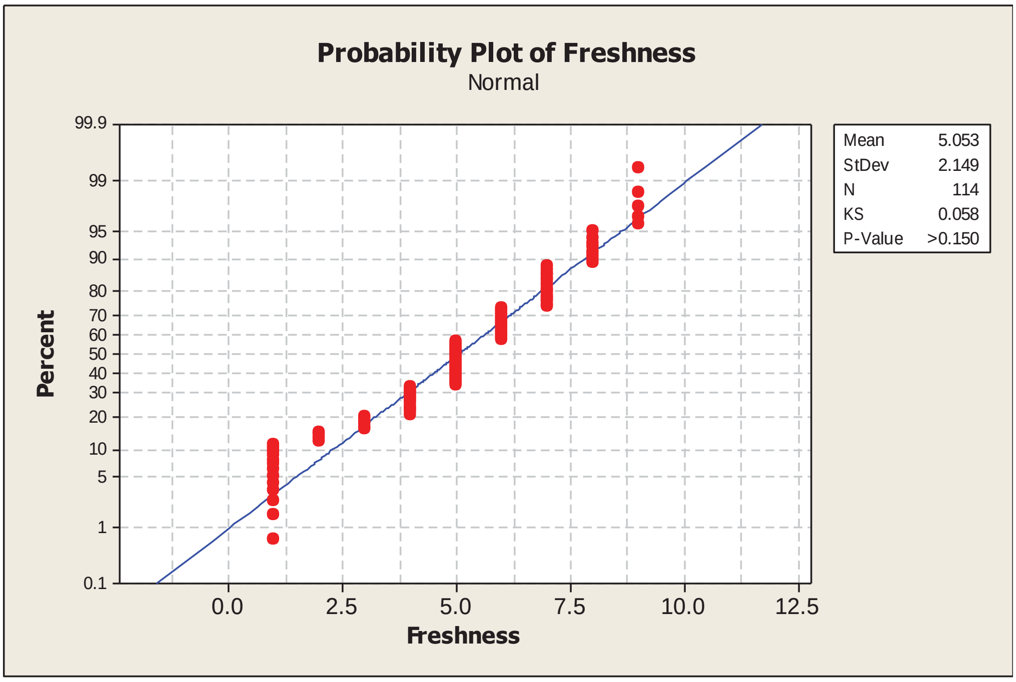
Normality plot Flavour



One-way ANOVA: Flavour versus Sample

```
Source  DF      SS     MS      F      P
Sample   4  207.20  51.80  50.84  0.000
Error   95   96.80   1.02
Total   99  304.00

S = 1.009   R-Sq = 68.16%   R-Sq(adj) = 66.82%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ----+---------+---------+---------+-----
126    20  5.700  1.031                  (--*--)
342    20  3.900  1.252        (--*--)
561    20  2.900  0.968  (--*--)
799    20  7.000  0.795                          (--*--)
853    20  5.500  0.946                 (--*--)
                         ----+---------+---------+---------+-----
                           3.0       4.5       6.0       7.5

Pooled StDev = 1.009
```

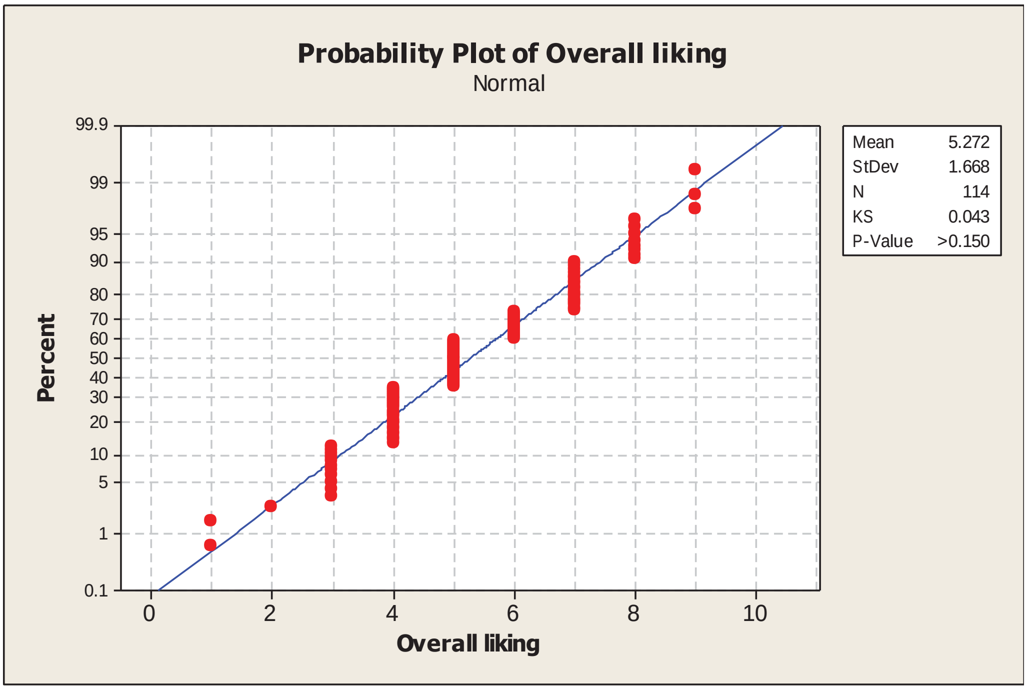
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source  DF      SS     MS      F      P
Sample   4  302.56  75.64  55.45  0.000
Error   95  129.60   1.36
Total   99  432.16

S = 1.168   R-Sq = 70.01%   R-Sq(adj) = 68.75%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ---+---------+---------+---------+------
126    20  6.900  0.968                        (--*--)
342    20  3.300  1.658   (--*--)
561    20  3.100  1.334  (---*--)
799    20  7.100  0.968                          (--*---)
853    20  6.000  0.649                    (--*--)
                         ---+---------+---------+---------+------
                           3.0       4.5       6.0       7.5

Pooled StDev = 1.168
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

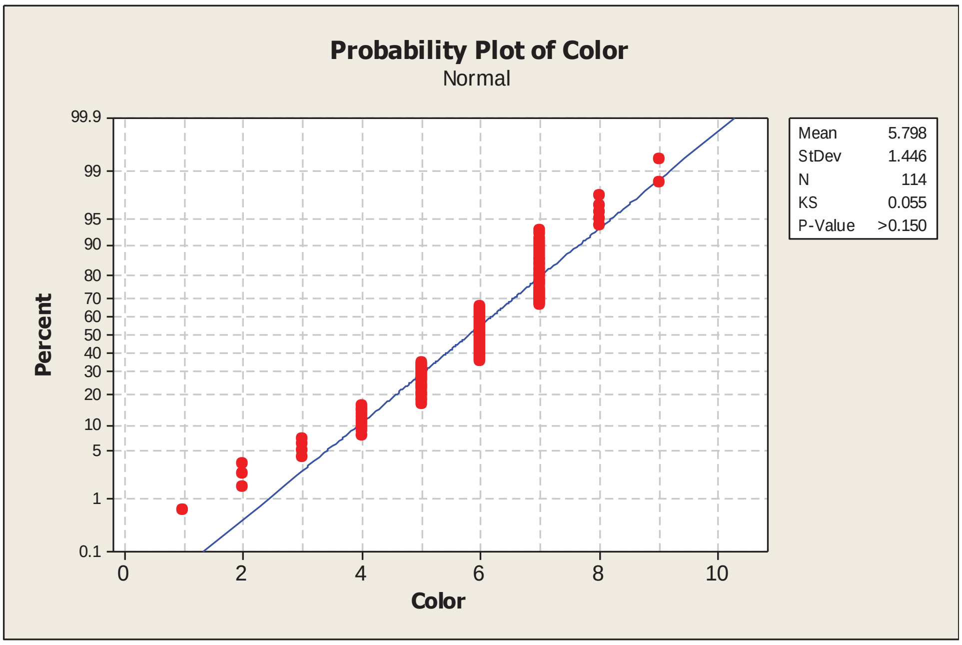
```
Source  DF       SS      MS      F      P
Sample   4   87.440  21.860  39.04  0.000
Error   95   53.200   0.560
Total   99  140.640

S = 0.7483   R-Sq = 62.17%   R-Sq(adj) = 60.58%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N    Mean   StDev  ----+---------+---------+---------+-----
126    20  6.3000  0.6569                        (---*---)
342    20  4.8000  0.8944          (---*---)
561    20  4.0000  1.0260  (---*---)
799    20  6.5000  0.5130                           (---*---)
853    20  5.6000  0.5026                 (---*---)
                          ----+---------+---------+---------+-----
                            4.00      4.80      5.60      6.40

Pooled StDev = 0.7483



Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4    7.760    7.760   1.940  1.81  0.134
Error   95  102.000  102.000   1.074
```

```
Total   99  109.760


S = 1.03619   R-Sq = 7.07%   R-Sq(adj) = 3.16%


Unusual Observations for Color

Obs    Color      Fit   SE Fit  Residual  St Resid
 21  8.00000  5.80000  0.23170   2.20000      2.18 R
 31  8.00000  5.80000  0.23170   2.20000      2.18 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4  20.9600  20.9600  5.2400  6.69  0.000
Error   95  74.4000  74.4000  0.7832
Total   99  95.3600


S = 0.884962   R-Sq = 21.98%   R-Sq(adj) = 18.69%


Unusual Observations for Texture

Obs  Texture      Fit   SE Fit  Residual  St Resid
  3  5.00000  6.80000  0.19788  -1.80000     -2.09 R
 13  5.00000  6.80000  0.19788  -1.80000     -2.09 R
 61  8.00000  5.70000  0.19788   2.30000      2.67 R
 71  8.00000  5.70000  0.19788   2.30000      2.67 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavour, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  207.200  207.200  51.800  50.84  0.000
Error   95   96.800   96.800   1.019
Total   99  304.000


S = 1.00943   R-Sq = 68.16%   R-Sq(adj) = 66.82%


Unusual Observations for Flavour

Obs  Flavour      Fit   SE Fit  Residual  St Resid
 62  6.00000  3.90000  0.22572   2.10000      2.13 R
 70  1.00000  3.90000  0.22572  -2.90000     -2.95 R
 72  6.00000  3.90000  0.22572   2.10000      2.13 R
 80  1.00000  3.90000  0.22572  -2.90000     -2.95 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Freshness, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  302.560  302.560  75.640  55.45  0.000
```

```
Error   95  129.600  129.600   1.364
Total   99  432.160


S = 1.16799   R-Sq = 70.01%   R-Sq(adj) = 68.75%


Unusual Observations for Freshness

Obs  Freshness      Fit   SE Fit  Residual  St Resid
 68    1.00000  3.30000  0.26117  -2.30000     -2.02 R
 69    1.00000  3.30000  0.26117  -2.30000     -2.02 R
 70    1.00000  3.30000  0.26117  -2.30000     -2.02 R
 78    1.00000  3.30000  0.26117  -2.30000     -2.02 R
 79    1.00000  3.30000  0.26117  -2.30000     -2.02 R
 80    1.00000  3.30000  0.26117  -2.30000     -2.02 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Overall, using Adjusted SS for Tests

Source  DF   Seq SS  Adj SS  Adj MS      F      P
Sample   4   87.440  87.440  21.860  39.04  0.000
Error   95   53.200  53.200   0.560
Total   99  140.640


S = 0.748331   R-Sq = 62.17%   R-Sq(adj) = 60.58%


Unusual Observations for Overall

Obs  Overall      Fit   SE Fit  Residual  St Resid
 70  3.00000  4.80000  0.16733  -1.80000     -2.47 R
 80  3.00000  4.80000  0.16733  -1.80000     -2.47 R
 82  2.00000  4.00000  0.16733  -2.00000     -2.74 R
 92  2.00000  4.00000  0.16733  -2.00000     -2.74 R

R denotes an observation with a large standardized residual.
```

Week 16

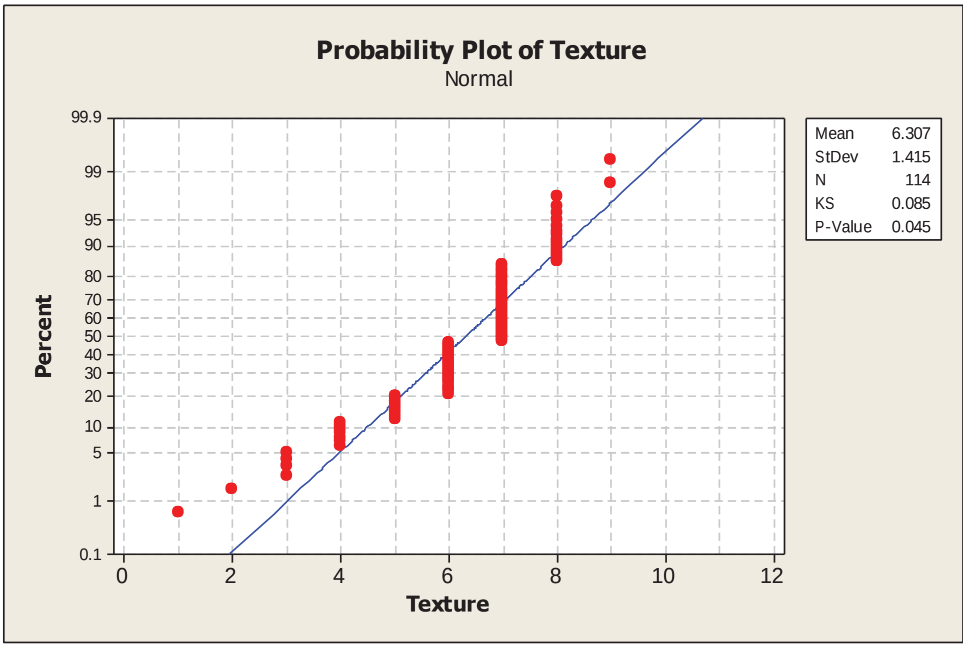
Normality plot for Color



One-way ANOVA: Color versus Sample

```
Source  DF      SS     MS     F      P
Sample   4   4.060  1.015  1.33  0.263
Error   95  72.300  0.761
Total   99  76.360

S = 0.8724   R-Sq = 5.32%   R-Sq(adj) = 1.33%


                          Individual 95% CIs For Mean Based on Pooled StDev
Level   N    Mean   StDev   +---------+---------+---------+---------
126    20  5.6500  0.7452           (----------*----------)
342    20  5.3000  1.0311   (----------*----------)
561    20  5.6000  0.8826          (----------*----------)
799    20  5.9000  0.9679                 (----------*----------)
853    20  5.4500  0.6863       (----------*----------)
                          +---------+---------+---------+---------
                          4.90      5.25      5.60      5.95

Pooled StDev = 0.8724
```

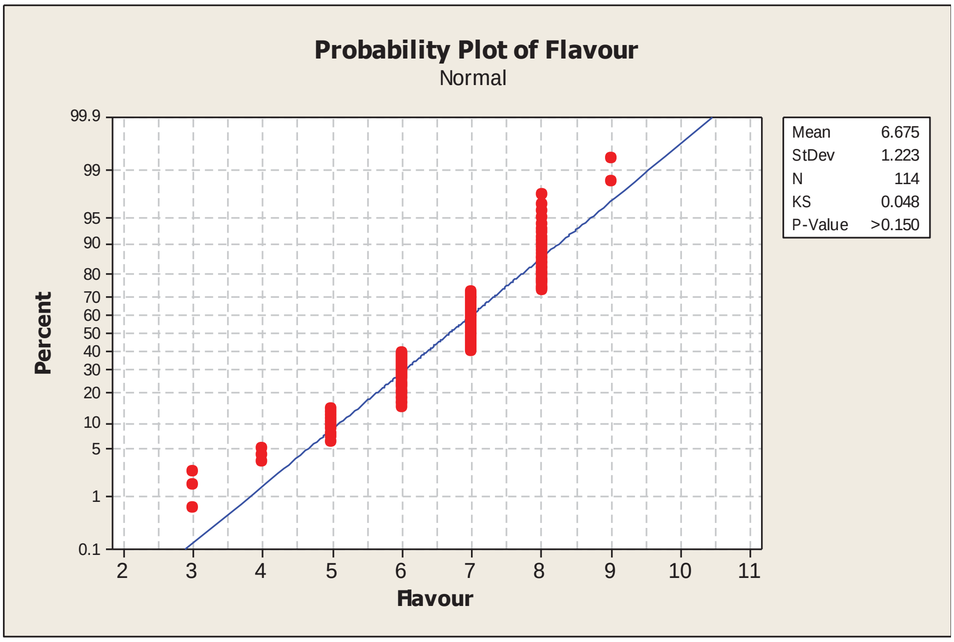
Normality plot for Texture



One-way ANOVA: Texture versus Sample

```
Source  DF      SS     MS     F      P
Sample   4  19.240  4.810  7.91  0.000
Error   95  57.750  0.608
Total   99  76.990

S = 0.7797   R-Sq = 24.99%   R-Sq(adj) = 21.83%


                          Individual 95% CIs For Mean Based on
                          Pooled StDev
Level   N    Mean   StDev  -----+---------+---------+---------+----
126    20  6.2000  0.7678            (-------*------)
342    20  5.8000  1.1050      (------*------)
561    20  5.6000  0.5026  (------*------)
799    20  6.7500  0.8507                     (------*------)
853    20  5.6000  0.5026  (------*------)
                          -----+---------+---------+---------+----
                            5.50      6.00      6.50      7.00

Pooled StDev = 0.7797
```

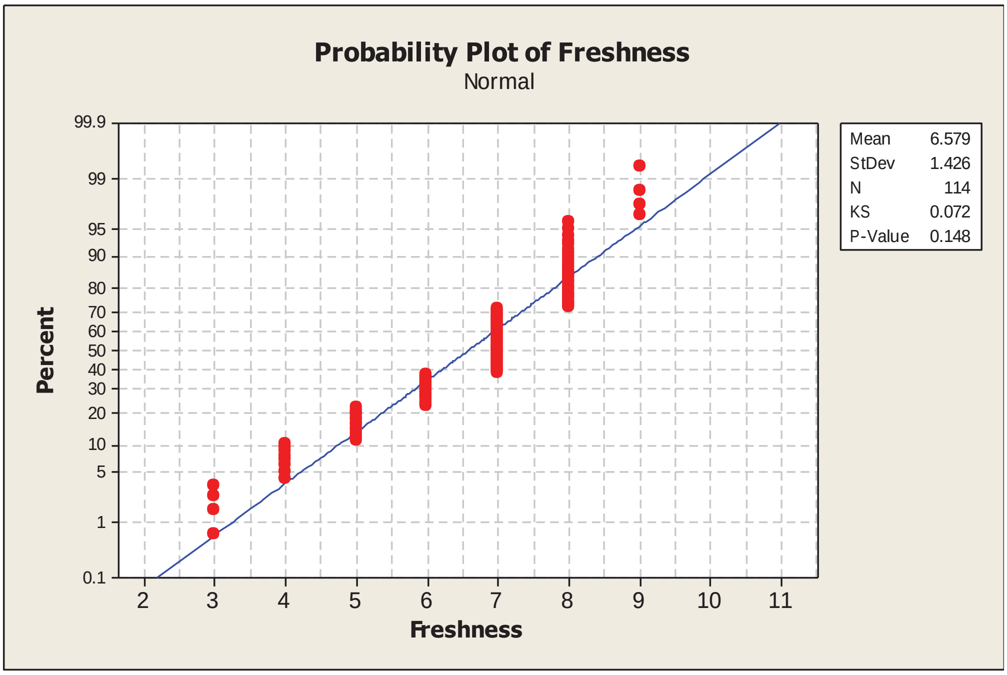
Normality plot for Flavour



One-way ANOVA: Flavour versus Sample

```
Source  DF       SS      MS      F      P
Sample   4  243.860  60.965  72.85  0.000
Error   95   79.500   0.837
Total   99  323.360

S = 0.9148   R-Sq = 75.41%   R-Sq(adj) = 74.38%


                           Individual 95% CIs For Mean Based on
                           Pooled StDev
Level   N    Mean   StDev  ---+---------+---------+---------+------
126    20  5.7500  0.9105                   (-*--)
342    20  3.2000  1.0052    (-*--)
561    20  3.0000  1.0260  (--*--)
799    20  7.0000  0.7947                          (--*-)
853    20  5.6500  0.8127                  (--*-)
                           ---+---------+---------+---------+------
                             3.0      4.5      6.0      7.5

Pooled StDev = 0.9148
```

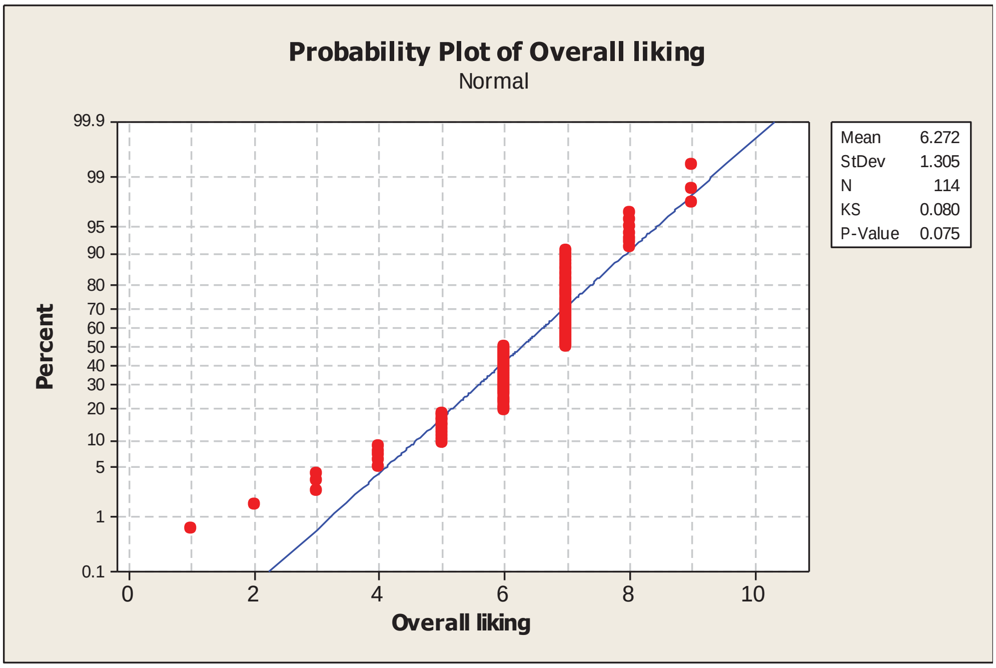
Normality plot for Freshness



One-way ANOVA: Freshness versus Sample

```
Source  DF      SS     MS      F      P
Sample   4  364.76  91.19  81.31  0.000
Error   95  106.55   1.12
Total   99  471.31

S = 1.059   R-Sq = 77.39%   R-Sq(adj) = 76.44%


                         Individual 95% CIs For Mean Based on
                         Pooled StDev
Level   N   Mean  StDev  ----+---------+---------+---------+-----
126    20  6.800  0.894                      (--*--)
342    20  2.800  1.609  (--*--)
561    20  2.850  1.040  (--*--)
799    20  7.100  0.852                        (--*--)
853    20  6.100  0.641                  (--*--)
                         ----+---------+---------+---------+-----
                           3.0       4.5       6.0       7.5

Pooled StDev = 1.059
```

Normality plot for Overall liking



One-way ANOVA: Overall versus Sample

```
Source  DF       SS      MS      F      P
Sample   4  151.740  37.935  93.61  0.000
Error   95   38.500   0.405
Total   99  190.240

S = 0.6366   R-Sq = 79.76%   R-Sq(adj) = 78.91%


                        Individual 95% CIs For Mean Based on
                        Pooled StDev
Level   N    Mean   StDev  ------+---------+---------+---------+---
126    20  6.4500  0.6048                        (-*--)
342    20  3.9000  0.7881    (--*--)
561    20  3.6500  0.7452  (-*--)
799    20  6.5000  0.5130                        (--*--)
853    20  5.7000  0.4702                   (--*--)
                        ------+---------+---------+---------+---
                            4.0       5.0       6.0       7.0

Pooled StDev = 0.6366
```

General Linear Model: Color, Texture, ... versus Sample

```
Factor  Type   Levels  Values
Sample  fixed       5  126, 342, 561, 799, 853


Analysis of Variance for Color, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4   4.0600   4.0600  1.0150  1.33  0.263
```

```
Error   95  72.3000  72.3000  0.7611
Total   99  76.3600


S = 0.872383   R-Sq = 5.32%   R-Sq(adj) = 1.33%


Unusual Observations for Color

Obs    Color      Fit   SE Fit  Residual  St Resid
  3  4.00000  5.90000  0.19507  -1.90000     -2.23 R
 13  4.00000  5.90000  0.19507  -1.90000     -2.23 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Texture, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS     F      P
Sample   4  19.2400  19.2400  4.8100  7.91  0.000
Error   95  57.7500  57.7500  0.6079
Total   99  76.9900


S = 0.779676   R-Sq = 24.99%   R-Sq(adj) = 21.83%


Unusual Observations for Texture

Obs  Texture      Fit   SE Fit  Residual  St Resid
  3  5.00000  6.75000  0.17434  -1.75000     -2.30 R
 13  5.00000  6.75000  0.17434  -1.75000     -2.30 R
 61  8.00000  5.80000  0.17434   2.20000      2.89 R
 71  8.00000  5.80000  0.17434   2.20000      2.89 R
 79  4.00000  5.80000  0.17434  -1.80000     -2.37 R
 80  4.00000  5.80000  0.17434  -1.80000     -2.37 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Flavour, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  243.860  243.860  60.965  72.85  0.000
Error   95   79.500   79.500   0.837
Total   99  323.360


S = 0.914791   R-Sq = 75.41%   R-Sq(adj) = 74.38%


Unusual Observations for Flavour

Obs  Flavour      Fit   SE Fit  Residual  St Resid
 62  5.00000  3.20000  0.20455   1.80000      2.02 R
 70  1.00000  3.20000  0.20455  -2.20000     -2.47 R
 80  1.00000  3.20000  0.20455  -2.20000     -2.47 R
 83  1.00000  3.00000  0.20455  -2.00000     -2.24 R
 96  5.00000  3.00000  0.20455   2.00000      2.24 R
 98  5.00000  3.00000  0.20455   2.00000      2.24 R

R denotes an observation with a large standardized residual.
```

```
Analysis of Variance for Freshness, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  364.760  364.760  91.190  81.31  0.000
Error   95  106.550  106.550   1.122
Total   99  471.310


S = 1.05905   R-Sq = 77.39%   R-Sq(adj) = 76.44%


Unusual Observations for Freshness

Obs  Freshness      Fit   SE Fit  Residual  St Resid
  6    5.00000  7.10000  0.23681  -2.10000     -2.03 R
 62    5.00000  2.80000  0.23681   2.20000      2.13 R
 64    5.00000  2.80000  0.23681   2.20000      2.13 R
 74    5.00000  2.80000  0.23681   2.20000      2.13 R

R denotes an observation with a large standardized residual.


Analysis of Variance for Overall, using Adjusted SS for Tests

Source  DF   Seq SS   Adj SS  Adj MS      F      P
Sample   4  151.740  151.740  37.935  93.61  0.000
Error   95   38.500   38.500   0.405
Total   99  190.240


S = 0.636603   R-Sq = 79.76%   R-Sq(adj) = 78.91%


Unusual Observations for Overall

Obs  Overall      Fit   SE Fit  Residual  St Resid
 27  5.00000  6.45000  0.14235  -1.45000     -2.34 R
 70  2.00000  3.90000  0.14235  -1.90000     -3.06 R
 82  2.00000  3.65000  0.14235  -1.65000     -2.66 R
 92  2.00000  3.65000  0.14235  -1.65000     -2.66 R
100  5.00000  3.65000  0.14235   1.35000      2.18 R

R denotes an observation with a large standardized residual.
```

8.4 Survival analysis for shelf-life estimation of the selected treatments using R

The R code and its output:

```
> ## --- Fit code
>
> ## --- Load survival library
> # install.packages("survival")
> library("survival")
Loading required package: splines
>
> local({pkg <- select.list(sort(.packages(all.available = TRUE)),graphics=TRUE)
+ if(nchar(pkg)) library(pkg, character.only=TRUE)})
> ## Read data
> Dat <- read.table (file="data1.txt", header=T)
> Dat <- Dat[Dat$TimePoint<17,]
> ## Transform data
> ## Sample size
> N <- dim(Dat)[1]
## Indices
> Like       <- Dat$OverallLiking  > 5
> NotLike    <- Dat$OverallLiking <= 5
> ## Initialise lower and upper bounds
> Time1 <- rep (0, N)
> Time2 <- rep (0, N)
> ## --- People like the food
> Time1[Like] <- Dat$TimePoint[Like]
```

```
> Time2[Like] <- NA
> ## --- People don't like the food
> Time1[NotLike] <- Dat$TimePoint[NotLike] - 2  ## All time points two apart
> Time2[NotLike] <- Dat$TimePoint[NotLike]
> ## --- Fix zeros (weibull can't be exactly zero)
> Time1[Time1 == 0] <- 0.01
> ## --- Create data set
> Survival <- Surv (time=Time1, time2=Time2, type="interval2")
> SDat     <- data.frame (Survival, Sample=factor(Dat$Sample))
> ## Shelf life function
> ShelfLife <- function (M, p=0.5) { exp(M$icoef)[1] * ( log(1/p) )^( exp(M$icoef)[2]) }
>
> ## Fit weibull models separately
> M1 <- survreg(Survival ~ 1, subset=Sample==levels(SDat$Sample)[1], data=SDat,
dist="weibull")
> M2 <- survreg(Survival ~ 1, subset=Sample==levels(SDat$Sample)[2], data=SDat,
dist="weibull")
> M3 <- survreg(Survival ~ 1, subset=Sample==levels(SDat$Sample)[3], data=SDat,
dist="weibull")
> M4 <- survreg(Survival ~ 1, subset=Sample==levels(SDat$Sample)[4], data=SDat,
dist="weibull")
> M5 <- survreg(Survival ~ 1, subset=Sample==levels(SDat$Sample)[5], data=SDat,
dist="weibull")
> ## Shelf lifes
> ShelfLife (M1, p=0.5)
(Intercept)
  20.57613
```

```
> ShelfLife (M2, p=0.5)
(Intercept)
  10.17278
> ShelfLife (M3, p=0.5)
(Intercept)
  9.256992
> ShelfLife (M4, p=0.5)
(Intercept)
  33.60168
> ShelfLife (M5, p=0.5)
(Intercept)
  15.42418
> ## Display coefficients
> Coef <- data.frame(rbind(M1$icoef, M2$icoef, M3$icoef, M4$icoef, M5$icoef))
> names(Coef) <- c("Intercept", "Slope")
> Weibull.Shape <- exp (-Coef$Slope)
> Weibull.Scale <- exp (Coef$Intercept)
>
> Par <- cbind (Weibull.Shape, Weibull.Scale)
> print (Par)
    Weibull.Shape Weibull.Scale
[1,]    1.1699088     28.14627
[2,]    2.3269118     11.90819
[3,]    2.0178833     11.10074
[4,]    0.9263245     49.91092
```

```
[5,]     1.3820482      20.10828

## Shelf lifes

> ShelfLife (M1, p=0.25)

(Intercept)

   37.21126

> ShelfLife (M2, p=0.25)

(Intercept)

   13.70278

> ShelfLife (M3, p=0.25)

(Intercept)

   13.05122

> ShelfLife (M4, p=0.25)

(Intercept)

   71.01229

> ShelfLife (M5, p=0.25)

(Intercept)

   25.46927

> ## Display coefficients

> Coef <- data.frame(rbind(M1$icoef, M2$icoef, M3$icoef, M4$icoef, M5$icoef))

> names(Coef) <- c("Intercept", "Slope")

> Weibull.Shape <- exp (-Coef$Slope)

> Weibull.Scale <- exp (Coef$Intercept)

>

> Par <- cbind (Weibull.Shape, Weibull.Scale)

> print (Par)
```

```
     Weibull.Shape Weibull.Scale
[1,]     1.1699088      28.14627
[2,]     2.3269118      11.90819
[3,]     2.0178833      11.10074
[4,]     0.9263245      49.91092
[5,]     1.3820482      20.10828
```

APPENDIX 9

9.1 Material Specifications

Globus Group Pty Ltd
ABN 94 000 070 155

1 Hartzell Place
Bankstown NSW 2200
Box VU
Bankstown Airport NSW 2
Australia
Tel: +61 (2) 8700 1700
Fax: +61 (2) 8700 1790

Tegel Foods Ltd

PACKAGING MATERIAL SPECIFICATION (F13)

VARIETY: Colour Case Brown 5 NPC (no soy) 165 x 20m

Tegel Foods Code :

Globus Part No :

DESCRIPTION: **Pre-shirred, Colour Casing**

Globus (NZ)Quality Management System **Certified to AS/NZS ISO9001-2000**
Auditor: Telarc SAI Ltd Client ID: 612

ISSUE No : 1 **DATE ISSUED: 12.5.09**

Prepared by : **Gordon Greenaway**

Globus Internal References : 95, 215, 243, 255

SUPERSEDES :

REASON FOR CHANGE:

Packaging Machine :

MATERIAL DATA

1. From external layer to internal layer
   (a) PE

   (b) Nylon

   (c) PE

   (d) Cellophane

   (e) Colour

2. Total gauge = 70µm ± 10%

3. Nominal Barrier Properties

   OTR: 30 - 50 cc/m^2.24hr.atm, measured at 23° and 60% RH

   WVTR: 9 - 20 g/m^2.24hr, measured at 40° and 90% RH

4. Thermal Shrinkage (MD/TD) % @ 80C for 5min. = 5 / 7

CASINGS DATA

1. Casing per stick	20m
2. Sticks per carton	12
3. Casing per carton	240m

4.	Layflat width	165mm
5.	Fill diameter (estimate)	105mm

PACKAGING/DELIVERY

1. Cartons

Each carton to be labeled with the following data:

- Supplier
- Globus Part Number
- Customer Part Number
- Description
- Quantity
- Batch Number
- Pack Date
- Customer Name
- Customer Product ID
- Globus O/N

SHELF LIFE

Shelf life is one year stored, unopened, in a cool and dark location.

COMPLIANCE

FDA No. & E.No.

Layer:

NPC: The film laminate is US FDA approved for use as food packaging.

Colour Transfer:

Brown 5: Sorghum Husk Extract, Annatto Extract (160b), Shellac (904), Glycerol (422), Methyl Cellulose (461), Cellulose (460ii), Sodium Alginate (401) and Sucrose esters of fatty acid (473) (The norbixin component of the Annatto is at a resultant level of 68.5mg/sqm of casing).

.

Note: Number in parenthesis is INS (International Numbering System) No. which equals to FDA No. and E. No. of EU Food Law. In case of E.No., attach E in front of each number.

Material without a number is categorised as food, not as additives.

GMO Information

Not containing any GMO products.

Allergen Information

Apart from those stated above the products are free from other major allergens defined by FDA and EU Food Law i.e.

FDA – 8 major allergens. Colour Case NPC is free of milk, eggs, fish, crustacean shellfish, tree nuts, soy bean, peanut and wheat.

EU – 12 major allergens. Colour Case NPC is free of milk, eggs, fish, crustacean shellfish, tree nuts, soy bean, peanut, cereals, celery, mustard, sesame, sulphur dioxide and sulphites (more than 10mg/kg).

Disclaimer : All information provided is correct to the best of our knowledge. Due to individual variations in product application, Globus Group does not accept responsibility for the ultimate products, their effectiveness, legality or infringement of patents.

FOOD PACKAGING
AND EQUIPMENT

GLOBUS GROUP NZ LTD
67-69 Seaview Rd
PO Box 30562
Lower Hutt, 5040
New Zealand
Tel: +64 4 568 3929
Fax: +64 4 568 4701

GLOBUS GROUP PTY LTD
ABN 94 000 070 155
1 Hartzell Place
Bankstown NSW 2200
PO Box 135
Georges Hall NSW 2198
Australia
Tel: +61 2 8700 1700
Fax: +61 2 8700 1790

GLOBUS MATERIAL SAFETY DATA SHEET (MSDS)

No. : 100

Dated : 5/8/09 (Globus Internal
Reference Nos. : 300)

Colour Case NPC – BW5 – No Soy

Refer No. 200907-054 written and/or revised on: July 21, 2009

1. Product	Product	: NPC - BW5-NS
2 Composition	Single material or Composite	: Composite
	Chemical name or General name,	: Package with color/food additives inside to be transferred
	FSANZ code, Elements and Contents	: Sorghum Husk Extract, Annatto Extract(160b), Shellac(904), Glycerol(422), Methyl Cellulose(461), Cellulose(460 ii), Sucrose esters of fatty acid (473) NPC film : cellophane (regenerated cellulose

			($C_6H_{10}O_5$) $_n$) /Polyethylene (ethylene homopolymer, $(C_2H_4)_{m,}$ CAS No.9002-88-4) / Nylon (Polyamid-6, Poly(ε-caprolactam), CAS No.25038-54-4)
3	Hazards and Toxicity	Classification	: not applicable
		Hazards	: not applicable
		Toxicity	: not applicable
4	Emergency & first aid	Eyes	: if colors was put in eyes, wash with clean water at least 15 minutes
		Skin	: if color was stuck to allergic skin, take the material off and wash with clean &/or warm water
		Inhalation	: ink is edible and not poisonous however in case of breathing being effected, it is recommended to move victim into open place with fresh air
		Swallow	: exhale/discharge the material and clean the mouth with water
		After above first aids , contact a doctor for further treatment	
5	Fire	Extinguishant	: water, sand, powders, carbon dioxide, air foams and so on
		Unsuitable Extinguishant	: nothing
		Extinguishing method	: extinguish fire with extinguisher and check spread of fire. If movable, carry goods in safe place. If immovable, spray water on surrounding area for cooling down.

		Protection while extinguishing	：use protectors when extinguishing fire
6	Accidental release measures	Color fastness	：Colorant coated on surface inside of tube is edible and will not easily get off under usual environment of air, water and warm water.
		Disposal	：After production by using the product, it’s recommendable to collect and handle the used product in same way as usual waste for burning.
7	Storage and handling	Handling for safety	：keep away from heat and flame. ：refer to 4.emergency and first aid & article 8 protectors if unusual contact, inhalation and ingestion etc happens.
		Storage	：remaining product should be well closed in bag to avoid air and humidity. ：keep in cool and dry conditions.

8 Protective measures	Protectors	protection is not necessary in usual conditions of usage. Those who have allergy against plastics and/or natural colorants, it’s recommendable to wear protectors.
	Respiratory	basically not necessary
	Hands	wear gloves
	Eyes	wear safety glasses
	Skin and Body	wear protective wear
9 Physical and Chemical Characteristics	Physical conditions	
	-Form and shape	sheet or heat-sealed tube with colorants
	-Color	brownish color
	-Smell	of certain odor (not hazardous)
	-Transition point in temperature	N.A.
	Explosively	
	Solubility	
10 Stability and Reactivity	Stability	stable under standard conditions
	Reactivity	stable under standard conditions
	Conditions to avoid	high temp high humidity, low temp low humidity
	Hazardous/toxic degradated products	N.A.
11 Hazard information	Acute toxic	not hazardous in standard application
	Local influence	no influence in normal usage

		In case stuck to allergenic skin or colorants come into eyes, follow section4 of first aid
12 Environmental Information	Environmental Information	The product is not hazardous material but it’s recommendable to dispose following local and ,state regulations
13 Disposal	waste	：burning is recommendable main components of off gas is water and Carbon dioxide. follow local and state regulations.
14 Transportation	International Regulations	：avoid making goods wet and rough handling so
	Handling	that packages will not be damaged
15 Regulatory information	Food Sanitation Act	：packaging material with coating of food additives and colorants
	Fire Service Law	：not applicable
	Poisonous and Deleterious Substances Control Law	：not applicable
	Industrial Safety and Health Law	：not applicable
	PRTR	：not applicable
	Waste Management Law	：waste plastics

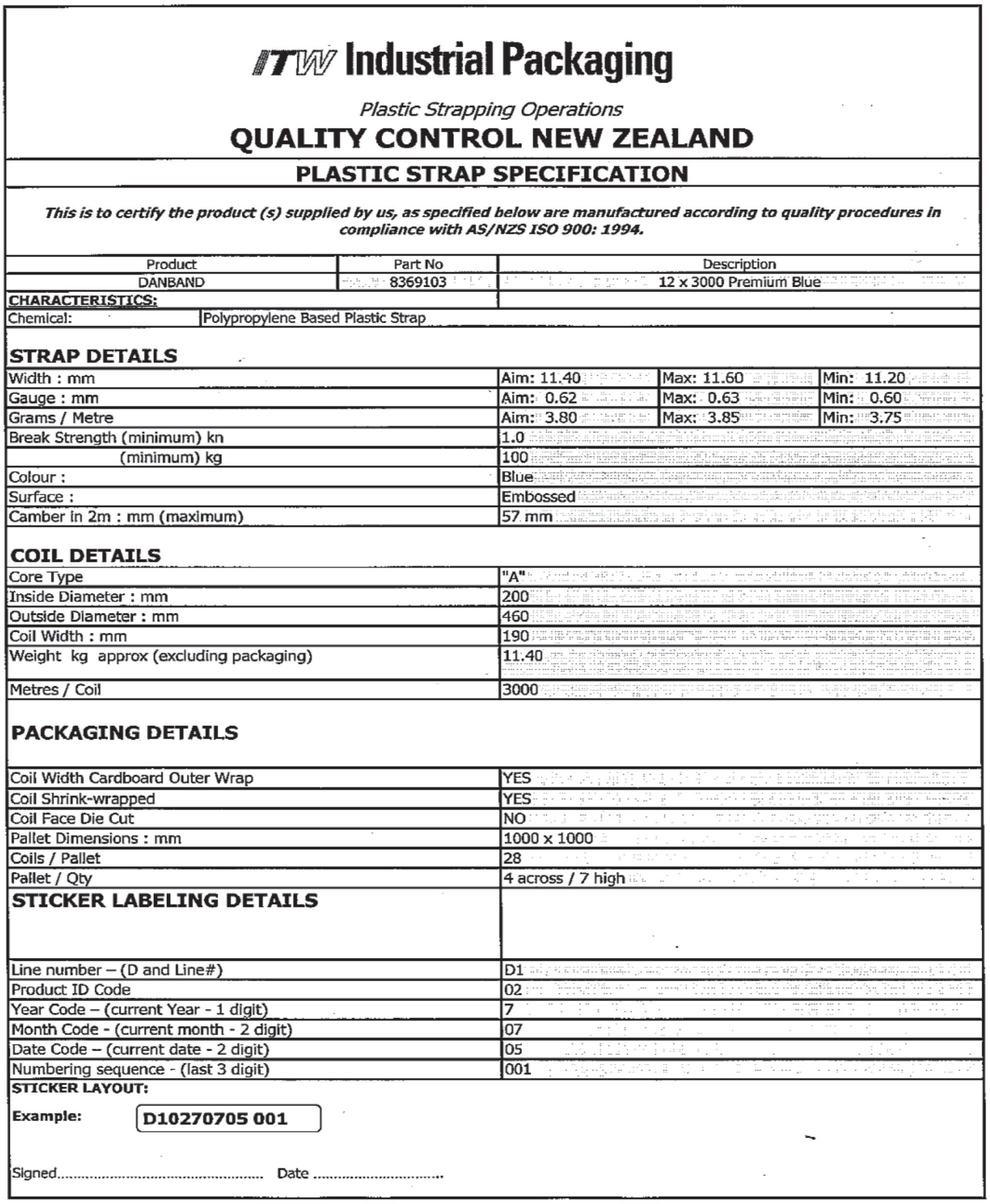
16 Others

This MSDS summarizes at the date of issue our best knowledge of the health and safety hazard information of the product, and in particular of safe handling and use of this product for application of casings of ham and sausage in your workplace. If new information comes in future, this MSDS will be revised according to necessity.

If clarification or further information is needed to ensure safe usage, users are requested to contact our sales representative, Globus Group.

Disclaimer : All information provided is correct to the best of our knowledge. Due to individual variations in product application, Globus Group does not accept responsibility for the ultimate products, their effectiveness, legality or infringement of patents.

Specifications for the Plastic Strap



ITW Industrial Packaging

Plastic Strapping Operations

QUALITY CONTROL NEW ZEALAND

PLASTIC STRAP SPECIFICATION

This is to certify the product (s) supplied by us, as specified below are manufactured according to quality procedures in compliance with AS/NZS ISO 900: 1994.

Product	Part No	Description
DANBAND	8369103	12 x 3000 Premium Blue

CHARACTERISTICS:	
Chemical:	Polypropylene Based Plastic Strap

STRAP DETAILS

Width : mm	Aim: 11.40	Max: 11.60	Min: 11.20
Gauge : mm	Aim: 0.62	Max: 0.63	Min: 0.60
Grams / Metre	Aim: 3.80	Max: 3.85	Min: 3.75
Break Strength (minimum) kn	1.0		
(minimum) kg	100		
Colour :	Blue		
Surface :	Embossed		
Camber in 2m : mm (maximum)	57 mm		

COIL DETAILS

Core Type	"A"
Inside Diameter : mm	200
Outside Diameter : mm	460
Coil Width : mm	190
Weight kg approx (excluding packaging)	11.40
Metres / Coil	3000

PACKAGING DETAILS

Coil Width Cardboard Outer Wrap	YES
Coil Shrink-wrapped	YES
Coil Face Die Cut	NO
Pallet Dimensions : mm	1000 x 1000
Coils / Pallet	28
Pallet / Qty	4 across / 7 high

STICKER LABELING DETAILS

Line number – (D and Line#)	D1
Product ID Code	02
Year Code – (current Year - 1 digit)	7
Month Code - (current month - 2 digit)	07
Date Code – (current date - 2 digit)	05
Numbering sequence - (last 3 digit)	001

STICKER LAYOUT:

Example: D10270705 001

Signed.. Date ....

Tegel item code 420281
Supplier item code 8369103

***ITW* Plastic Strapping Operations**
298 Neilson Street, Te Papapa

Auckland

NON METALLIC STRAPPING SPECIFICATIONS

PRODUCT DESCRIPTION

Type: Polypropylene Strapping (PP)
Brand: Danband Premium
Size: 12.00mm X 0.60mm
Part Number: **8269103 Blue**
83691A5 Clear
Characteristics: Embossed Surface
Blue or Clear Colour
Principal Applications: General Industries
i.e. Typically automated equipment

MECHANICAL PROPERTIES

Cross Section (min): 11.50mm x 0.55mm
Length Per Roll: 3000 metres
Breaking Load (minimum): 1.2 kilo Newtons (122kg)

STANDARD COILS AND PALLETS

Inside Diameter: 200mm
Outside Diameter: 210mm
Coil Width: 190mm
Coils Per Pallet: 24

AS/NZS ISO 9002