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Functional properties of microwave pasteurised and oil coated whole shell eggs

by

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DECLARATION

I hereby declare that this dissertation submitted to the University of Pretoria for the MInst Agrar degree is my work and has not previously been submitted by me for a degree at any other University or institution of higher education.

Muvhulawa Sylvia Mudau

DEDICATION

I would like to dedicate this dissertation to my children Orinea and Gundo Neluheni, who were born during the course of this study.

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Firstly, I thank God for He is the reason I am able to do any of this under his guidance throughout my studies.

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ABSTRACT

Functional properties of microwave pasteurised and oil coated whole shell eggs

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Department: Food Science

Degree: M InstAgrar (Food Production and Processing)

Food borne infections due to *Salmonella enterica* serovar Enteritidis (*S.* Enteritidis) has shown a dramatic increase in many countries. Different egg pasteurisation treaments have been developed in the past but are still not providing practical or optimal solutions. A method is required that ensure that eggs are microbiologically safe, that does not affect the functional quality and possibly also extend the egg shelf life. This research project formed part of a larger study, "Project 32438: The development of a novel microwave system for the pasteurisation of raw whole shell eggs" funded by the National Research Foundation Innovation Fund and conducted by a consortium consisting of the Council for Scientific and Industrial Research (CSIR), Delphius Technologies, Eggbert Eggs and University of Pretoria. One of the phases in the development of the microwave system was an evaluation of the effectiveness of applying different microwave power levels (250W and 300W) on eliminating or reducing *S.* Enteritidis as well as evaluating the effect on the functional properties of the eggs. The microbiological tests were conducted by the CSIR while the latter evaluation was the focus of the study reported here.

Microwave pasteurised eggs had lower foaming capacity but higher Haugh values than control (unpasteurised) eggs. Albumin foam stability did not differ between control and microwave pasteurised eggs and the pH of the albumin was almost similar. The yolk pH of pasteurised eggs was higher than that of unpasteurised eggs. Significant differences were found for the sensory properties of broken out eggs as evaluated by a trained sensory panel. At 300W, pasteurised eggs collected from the left side of the oven had partially coagulated albumin that was not clear. The visual appearance of

pasteurised eggs at 300W from left side was more adversely affected than the eggs collected from the right side oven position and all eggs pasteurised at 250W. The albumin foaming capacity, visual appearance and sensory properties of raw eggs pasteurised at 250W were slightly affected by microwave heating. A triangle taste test showed that there was a significant difference between control and pasteurised (300W) eggs. A home use consumer test showed that control and microwave pasteurised (250W) eggs were equally acceptable.

Pasteurisation could extend the shelf life of whole shell eggs (WSE) by reducing or destroying spoilage microorganisms. Another phase of the project therefore focused on obtaining background data pertaining to the shelf life of eggs. The effect of coating of egg shells with mineral oil on the functional properties and shelf life of WSE stored at 16ºC (58 % RH); 25ºC during the day and 15ºC at night (55% RH) and 32ºC (32 % RH) for a period of six weeks, were evaluated. These conditions were selected to reflect typical temperatures that are used for storing eggs in South Africa. Haugh units of eggs decreased with storage time at all storage conditions but for coated eggs it decreased at a slower rate. The pH of both the yolk and albumin of coated shell eggs was lower than that of uncoated shell eggs. Coating did not have an influence on the foam stability of egg albumen. Foaming capacity of albumen was negatively affected by oil coating. Coated shell eggs stored at the three conditions had a prolonged shelf life compared to uncoated eggs stored in the same manner.

If the prototype microwave oven can be optimised to ensure even distribution of microwaves, microwave pasteurisation of shell eggs has potential to become a significant break through in the poultry industry.

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CHAPTER 1

1. INTRODUCTION

The incidence of food borne infections caused by *Salmonella enterica* serovar Enteritidis (*S.* Enteritidis) has shown a dramatic increase in many countries (Nastasi, Mammina, Fantasia & Pontello, 1997). This pathogen is widely distributed in egg laying flocks and infects the ovaries of the hen (Suzuki, 1994). Consumption of contaminated shell eggs with *S.* Enteritidis causes food borne disease in people (Radkowski, 2001). The contents of eggs become infected at the time of lay before the shell is formed through transovarian transmission (Cox, Berrang & Cason, 2000). Although there are several methods of preservation and sanitation of shell eggs such as washing, rapid chilling, irradiation and ultrasonic treatment, these do not destroy *S.* Enteritidis, which is harboured inside shell eggs (Catalano & Knabel, 1994).

Heat treatments are used to pasteurise shell eggs (James, Lechevalier & Ketteringham, 2002) so that the *S.* Enteritidis which is inside the eggs could be eradicated (Hou, Singh, Muriana & Stadelman, 1996). Hou *et al*. (1996) found that heating whole shell eggs (WSE), using a water bath at 57°C, for long enough to destroy Salmonellae i.e. more than 30 min, denatures albumin proteins. Egg proteins are responsible for many functional properties such as foaming, coagulation, viscosity building and emulsification in many types of foods (Chang & Chen, 2000). Pasteurisation methods such as hot air, hot water immersion (Himathongkham, Riemann & Ernst, 1999; Mermelstein, 2001) atmospheric steam, and irradiation (Ma, 1996; Ferreira & Del Mastro, 1998) have also been applied to pasteurise WSE. These methods are inadequate and adversely affect functional properties and sensory qualities of eggs. Therefore, suitable pasteurisation methods are required to make the egg bacteriologically safe; possibly also extend the shelf life, but it should maintain functional and sensory qualities (Ferreira & Del Mastro, 1998).

Another method that has potential of reducing the level of *S.* Enteritidis is microwave heating. Microwaves are non-ionizing energy capable of generating heat deep inside the penetrated medium by the molecular friction in an alternating electromagnetic field (Lewandowicz, Fornal & Walkowski, 1998). Microwave heating can possibly help to eradicate *S.* Enteritidis located inside shell eggs with less damage to the functional and

sensory properties. Microwaves penetrate the foods rapidly and cause friction heat by interacting with polarized molecules such as water, fat, amino acids, and proteins (Decareau, 1985; Food and Drug Administration, 2000; Bennion & Scheule, 2004).

This research project formed part of a larger study, " Project 32438: The development of a novel microwave system for the pasteurisation of raw whole shell eggs" funded by the NRF Innovation Fund and conducted by a consortium consisting of CSIR, Delphius Technologies, Eggbert Eggs and University of Pretoria. The purpose of this part of the study was to evaluate the effects of pasteurising WSE in order to eliminate *S.* Enteritidis which may be inside eggs, on the functional and sensory properties. To accomplish this purpose, microwave heating as an alternative method of pasteurising WSE was used in this study. This study also focused on the effects of oil coating and storage conditions on the functional properties and shelf life of WSE. Limited information exists in literature on the effects of microwave pasteurisation and oil coating on the functional properties and shelf life of WSE.

CHAPTER 2

2. LITERATURE REVIEW

2.1 Contamination of eggs by *S.* Enteritidis

Studies indicate that when contamination occurs by transovarian transmission, *S.* Enteritidis is introduced into the egg from infected ovaries or oviduct tissue prior to shell deposition (Whiting *et al.*, according to Latimer, Jaykus, Morales, Cowen & Crawford-Brown, 2002; Gast & Holt, 2000; Cox *et al.*, 2000). *S.* Enteritidis is widely distributed in the egg environment especially in laying flocks (Suzuki, 1994). The trans-shell transmission is usually derived from the feacal contamination on the surface of the eggshell, although it may also include contamination through environmental vectors such as humans, pets, and rodents (Fig.1) (Guard-Petter, 2001).

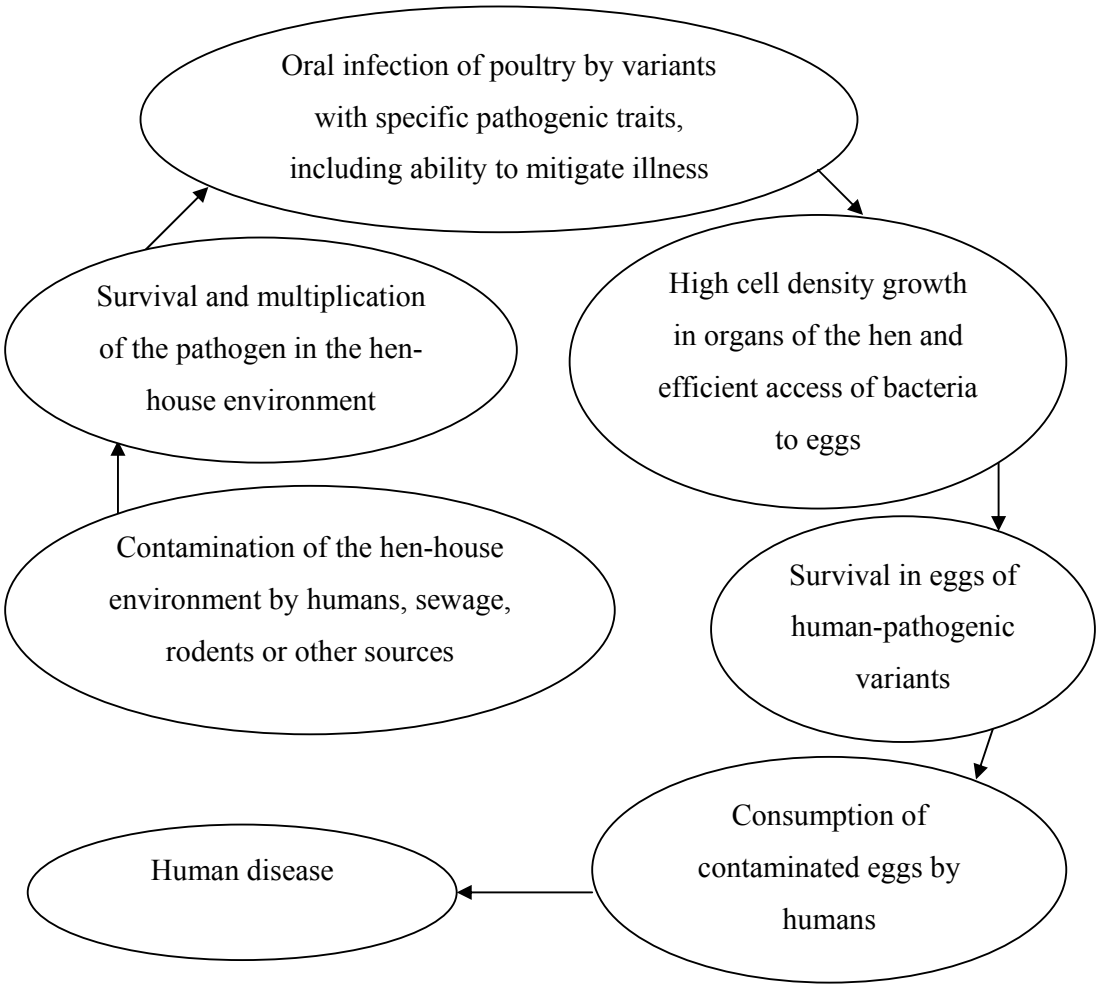

Figure 1. The route to human infection by *S.* Enteritidis after consuming infected eggs (Guard-Petter, 2001)

Consumer mass market shell eggs are said to be contaminated rather than infected, as they are not fertile and do not contain living avian cells. *S.* Enteritidis on eggshells can contaminate egg contents by migration through the shell and associated membranes. For epidemiological purposes, the route is primarily linear (Fig.1), with humans as the end host; however, transmission of infection from humans to chickens suggests that the route can be cyclical (Guard-Petter, 2001).

The bacteria on the surface of the shell are able to pass through the pores of the shell to contaminate the interior of the egg, even though the shell has some physical barriers (Wang & Slavik, 1998). Mayes & Takeballi found that albumin gets contaminated when the cuticle, shell and shell membranes fail to prevent microbial invasion (according to Messens, Duboccage, Grijspeerdt, Heyndrickx & Herman, 2004). Ebel & Schlosser reported that there is confusion between transovarian and trans-shell routes of contamination regarding which one of these is more prevalent in the commercial egg industry (according Latimer *et al*., 2002).

The *S.* Enteritidis is pandemic and involves interaction of the pathogen with multiple environments, including the hen house, the birds, the egg, as well as the human host (Fig.1) (Humphrey, 1994; Guard-Petter, 2001).

2.2 Outbreaks associated with *S.* Enteritidis

S. Enteritidis is the cause of food borne illness in humans (Guard-Petter, 2001). Salmonellosis in humans is an intestinal infection with *Salmonella* and is characterized by headache, abdominal pain, fever, nausea, and vomiting (Guard-Petter, 2001). The infection can spread to the blood stream and other areas of the body, leading to severe fatal illness in people with weakened immune systems (Potter, 1999). Schroeter *et al.* reported that poultry meat, mayonnaise, sauces, and other food products containing raw eggs have been implicated with salmonellosis (according to Maré, Van der Walt & Dicks, 2000).

Epidemiological investigations in Hungary, the United Kingdom, the United States and Germany confirmed that the food that is mostly responsible for food poisoning was egg (Ullmann & Scholtze, according to Guard-Petter, 2001). Fresh shell eggs that look clean and uncracked may contain *S.* Enteritidis that can cause food borne illness (Food Safety Focus and Inspection Services, 2003). Lee (2000) stated that the occurrence of

food poisoning from eggs has increased four fold in the United States and fourty fold in Europe. The role of chicken eggs in the transmission of *S.* Enteritidis to the human population in Europe (Lee, 2000), Canada (Todd, 1996) and Poland (Radkowski, 2001) has also been reported. The incidence of salmonellosis in humans in England and Wales has been reported (Guard-Petter, 2001). The cases of salmonellosis in connection with *S.* Enteritidis and other serovars such as *S.* typhimurium were increasing as from 1981 to 1998 (Guard-Petter, 2001).

2.3 Structure and composition of hens' eggs

2.3.1 Whole shell egg

The albumin, yolk, shell membranes and the shell are the major parts of a hen's egg (Linden & Lorient, 1999; Fennema, 1985). Fig 2. illustrates the morphology of a hen's egg. Yolk and albumin vary in their physical tolerances, composition, and capacity for microbial growth (Humphrey, 1994). The colour of the shell does not affect the quality, flavour, cooking characteristics and nutritional value of the egg contents because the colour could be white or brown depending on the breed of the laying hen. There are two membranes inside the shell, the outer membrane is attached to the shell and the inner is attached to the albumin. These membranes supply protective barriers against microbial infection (Vaclavik, 1999).

2.3.2 Egg yolk

The egg yolk is spherical in shape and surrounded by a colourless membrane. The yolk contains cholesterol and almost all of the fat. The yolk absorbs more energy and cooks faster due to the higher amount of fat. Fats have low specific heats compared with water; hence, foods that are high in fat heat more quickly than foods high in water (Bennion & Scheule, 2004). Egg yolk has a higher nutrient density (e.g. lipoproteins) compared to the albumin, containing most of the vitamins with the exception of vitamin C. The lipoproteins are classified as high density lipoprotein (HDL), low density lipoprotein (LDL), and very-low density lipoprotein (VLDL). Density is determined by the percentage of lipid material in the molecule. The HDL fraction of the egg yolk consists of phosvitin and the livetins, α-, β-, and γ-livetin. Livetin is a water soluble plasma protein that is composed of a globular protein fraction. Phosvitin contains no lipid material and is a phosphoprotein (Caballero, Trugo & Finglas, 2003).

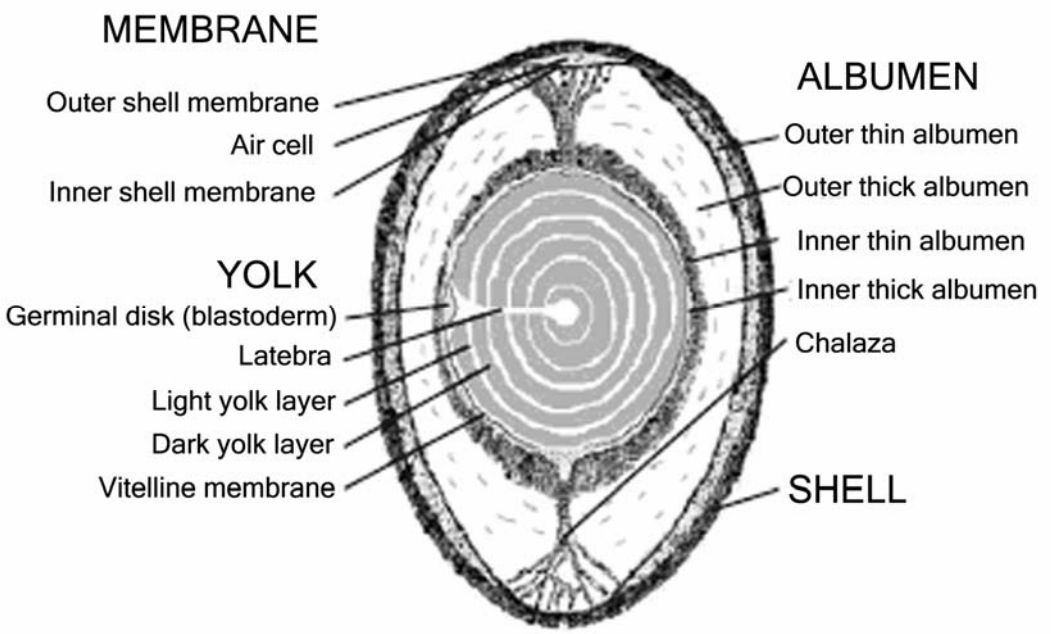

Figure 2. Schematic of hen egg morphology (Laghi, Cremonini, Placucci, Sykora, Wright & Hills, 2005)

The major yolk proteins are lipoproteins, which include vitellin and vitellinin, and are responsible for the excellent emulsifying properties of egg yolk when used in products such as mayonnaise. The yolk colour depends on the type of diet or feed given to a laying hen, specifically linked to yellow pigments such as carotenoids in the diet (Vaclavik, 1999).

2.3.3 Albumin

Albumin is the major constituent of the egg (Mine, 1995). The water content of albumin is between 87 to 89 % and is dependent on the strain and age of the hens (Powrie, 1977). Albumin is composed of four distinct layers: two thick whites separated by inner and outer thin whites. Eggs contain a very high concentration of complete protein with all essential amino acids in a well-balanced proportion. The predominant protein in egg albumin is ovalbumin, a protein that denatures easily by heat. Other proteins that are found in albumin include avidin, conalbumin, ovomucoid and globulins including lysozyme. Ovomucin, which is present in a significant amount, has a great effect on the consistency of thick albumin (Fennema, 1985; Vaclavik, 1999). Carbohydrates exist either in a free form or combined with the proteins. Albumin also contains a high percentage of protein and low percentage of lipid

compared to the egg yolk. In addition, biotin, niacin, and riboflavin are vitamins that are found in the egg albumin. Minerals contained in the albumin include magnesium and potassium (Vaclavick, 1999).

2.3.3.1 Albumin proteins

2.3.3.1.1 Ovalbumin

Ovalbumin is a major protein in albumin. It is classified as a phosphoglycoprotein, since carbohydrates are attached to the polypeptide (Li-Chan, Powrie & Nakai, 1995). The peptide chain of ovalbumin contains four free sulfhydryl groups and one disulfide group and during storage of shell eggs native ovalbumin is converted to *S*-ovalbumin, a more heat stable protein (Linden & Lorient, 1999). The *S*-form is different from native ovalbumin only in its greater stability, compactness and hydrophobicity (Nakamura & Ishimashu, 1981). The appearance of *S*-ovalbumin occurs at the same time as the loss of the food value of eggs since eggs with high *S*-ovalbumin content have runny whites. Ovalbumin is the only albumin protein to contain free sulfhydryl groups (Mine, 1995).

2.3.3.1.2 Ovotransferrin

Ovotransferrin is also known as conalbumin (Li-Chan *et al*., 1995). It is a glycoprotein that binds iron (Table 1) and inhibits bacteria (Mine, 1995).

Table 1. Composition and some physico-chemical properties of the major albumin proteins (Mine, 1995)

Protein	Albumin (% mass)	Isoelectric points (pI)	Molecular weight (kDa)	Denaturation temperature (T_d)	Characteristics
Ovalbumin	54.0	4.5	44.5	84.0	Phosphoglycoprotein
Ovotransferrin	12.0	6.1	77.7	61.0	Binds metallic ions
Ovomucoid	11.0	4.1	28.0	77.0	Inhibits trypsin
Ovomucin	3.5	4.5-5.0	$5.5\text{-}8.3 \times 10^3$	-	Sialoprotein; viscous
Lysozyme	3.4	10.7	14.3	75.0	Lyses some bacteria
G2 Globulin	4.0	5.5	49.0	92.5	-
G3 Globulin	4.0	5.8	49.0	-	-
Avidin	0.05	10.0	68.3	-	Binds biotin

Ovalbumin is more heat resistant than ovotransferrin and less vulnerable to surface denaturation. It is reported that this protein is stable at 57°C for 10 minutes at pH 9. It consists of a single oligosaccharide unit composed of four mannose residues and eight N-acetylglucosamine residues that have the capacity to bind bi- and trivalent metal ions into a complex. Two atoms of Fe^{3+}, Al^{3+}, Cu^{2+}, or Zn^{2+} per molecule of protein form a heat stable complex at pH 6. These complexes are red, colourless, yellow, and colourless respectively. Therefore, conalbumin metallic ion complexes are resistant to thermal denaturation and proteolytic attack (Linden & Lorient, 1999; Fennema, 1985).

2.3.3.1.3 Ovomucoid

Ovomucoid is a heat resistant glycoprotein that coagulates at temperatures above 60°C in acidic conditions (Fennema, 1985). Hen ovomucoid has nine disulfides and no free sulfydryl groups. Ovomucoid gives the albumin its high viscosity that is important for foam stability (Coultate, 2002).

2.3.3.1.4 Lysozyme

Lysozyme is an enzyme in albumin, which is capable of lysing bacterial cells. It is much more heat sensitive in egg albumin than when present alone in a phosphate buffer between pH 7 and 9. Lysozyme is inactivated at pH above 7 (Fennema, 1985 & Li-Chan *et al.,* 1995).

2.3.3.1.5 Ovomucin

Ovomucin comprises 3% of the total albumin. Hen albumin ovomucin is a glycoprotein characterized by a high molecular weight and it strongly affects the viscosity properties of albumin (Powrie & Nakai, 1986). Two different types of ovomucin complexes have been reported in hen albumin: an insoluble ovomucin complex formed from whole thick albumin and a soluble one formed from both thick and thin albumin. Ovomucin and lysozyme can interact to form a water-soluble complex over the pH range of 7.2 to 10.4 (Fennema, 1985; Mine, 1995). A complex has been reported to be stable at 99°C for 2 hours at pH values between 7.1 and 9.4.

2.3.3.1.6 Globulin (Ovoglobulins)

The presence of three globulins, G1, G2, G3, in albumin was reported in 1940 (Mine, 1995). The G1 was identified as lysosome and is well characterized. The globulins (G2

and G3) each comprise 4 % of the albumin proteins. The globulins are considered to be important in the foaming of albumin.

2.3.3.1.7 Avidin

Avidin is a strongly basic glycoprotein synthesized in the hen oviduct and deposited in the albumin fraction of eggs. Avidin is a tetrameric protein, consisting of subunits of identical amino acid composition and sequence (15.6 kDa and 128 amino acids each). Avidin is believed to be a trace component (0.05 %) of albumin. It has been well studied because of its ability to tightly and specifically bind biotin, one of vitamin B group (Davies & Reeves, 2002).

2.4 Methods used to pasteurise whole shell eggs

Pasteurisation is a process whereby foods are heated below 100˚C for a definite time to inactivate enzymes, pathogenic and non-pathogenic organisms and to extend the shelf life (Fellows, 2000).

2.4.1 Water bath (Hot water immersion)

In the water bath, heat is transferred from hot water to the eggshell, through convection and then by conductive heat transfer from outside to inside the egg. The shell eggs are conveyed through a hot water bath until the center of the yolk reaches the desired temperature for the desired length of time. Hou *et al*. (1996) found that the use of a water bath to pasteurise WSE at 57ºC for 30min does not affect the functional qualities of albumin proteins. These conditions also gave a 3-log cycle reduction in count of *S.* Enteritidis*.* The albumin proteins denatured when processing time exceeded 30 min. Himathongkham *et al*., (1999) reported that dipping eggs for 3 seconds in boiling water resulted in complete destruction of *S.* Enteritidis on shells and membranes, but sometimes caused eggs to crack. According to Davidson (2004), the use of waterbath to pasteurised shell eggs reduced Salmonella by 4.8 logs.

2.4.2 Hot air pasteurisation

The hot air oven heats the surface of the egg by convection and through conduction within the egg. The oven was set at 55ºC for 3 hours to pasteurise shell eggs and a maximum of 5-log reduction in *S.* Enteritidis was achieved with little damage to the functionality of albumin proteins (Hou *et al*., 1996).

2.4.3 Irradiation

Irradiation is a rapid technology and destroys the DNA of bacteria (Mermelstein, 2001). Much research have been carried out world wide on irradiation and it was found that irradiation is effective in destroying food borne pathogens like *S.* Enteritidis that contaminate poultry meat and eggs. Ferreira & Del Mastro (1998) reported that Co_{60} gamma irradiation with 5kGy doses can reduce the bacterial count on eggs to non-detective levels. It has been reported that irradiation of eggs influence the physical appearance of eggs such as lowering viscosity of albumin, and weaken the yolk membrane that may have a negative influence on consumers (Ma, 1996; Wong & Kitts, 2003). In addition, irradiation has a negative influence on the flavour, aroma, whippability and freshness of eggs (Mermelstein, 2001).

2.5 Functional properties of eggs

Functional properties of eggs refer to the attributes of eggs that make them a useful ingredient in food products. Coagulation, foaming, emulsifying, colouring, flavouring and anti crystallizing properties as well as providing essential nutrients are the most important functions of eggs (Yang & Baldwin, 1995). The functional properties of chicken eggs are affected by temperature and storage (Reddy, Reddy & Siddique, 1992). When eggs are stored, the conversion of ovalbumin into S-ovalbumin and the dissociation of the ovomucin-lysozyme complex (with destruction of the ovomucin gel) are major reactions from a technological point of view. They result in decreased gelling and foaming properties and liquefaction of albumen. These reactions are due to a rise in the pH (which can be accelerated by an increase in temperature). An increase in pH and its detrimental consequences can be minimized by storing shell eggs at 90 % relative humidity in order to reduce loss of water by evaporation (Linden & Lorient, 1999). The functional properties of eggs can be significantly affected by the nature and concentration of dietary vegetable lipids used to feed the laying hens (Tallarico, Sirrri, Meluzzi, Pittia, Parpinello & Franchini, 2002).

2.5.1 Foaming properties

Globulins, ovomucin and ovalbumin are proteins in albumin that are responsible for the foaming properties of the albumin. These proteins are particularly sensitive and may be damaged by heat processing (Davidson, 2003). Foaming capacity as well as stability are important criteria of foaming properties (Mine, 1995). Albumin is an excellent

foaming agent that contributes to the lightness of baked foods such as fluffy omellets, meringues, soufflés and candies (Kirunda & McKee, 2000). Damodaran (1997) described three important properties a protein should exhibit to be a good foaming agent; a protein should have the ability to rapidly adsorp protein at the air-water interface during whipping; conformational or structural change of protein at the interface should be rapid and reduction or lowering of surface tension should be fast; and the protein should form a high tensile strength, cohesive (viscoelastic) film through intermolecular interactions. Albumin proteins are mainly responsible for foam formation (Damodaran, 1997). Foaming ability of albumin depends on the quality of its proteins (Kirunda & McKee, 2000). Egg proteins produce a large volume of stable foams, which coagulate during heating. The foam of the egg proteins provides leavening and contributes lightness and volume to food products such as angel food cakes, sponge and chiffon (Lee & Chen, 2002; Du Preez, 2000).

Foams can be produced by beating the egg contents (Fennema, 1985). The rate and motion of beaters may influence the incorporation of air, while the pretreatments like blending, processing temperature, and the addition of ingredients will also influence egg foams (Du Preez, 2000). The volume of foam increases with the beating time to a maximum and gradually decreases (Fennema, 1985). The ability of proteins to form and stabilize foams depends on the source of proteins and degree of denaturation, the presence of calcium ions, the size and flexibility of proteins, pH, temperature and whipping methods and time (Cheftel, Cuq & Lorient, 1985). If proteins are damaged, then foam volume will decrease and the liquid drainage from the whipped foam will increase (Davidson, 2003).

2.5.1.1 Factors that influence the foaming ability of albumin proteins

2.5.1.1.1 pH

Linden & Lorient (1999) reported that globulin, ovalbumin and ovomucin showed excellent foaming performances at their native pH 8 to 9. As the foams were formed, the number of sulfhydryl groups decreased showing disulfide interchanges during foaming (Penfield & Campbell, 1990).

2.5.1.1.2 Whipping time

Mechanical actions, such as excessive beating, influence foaming ability of albumin. Beating albumin for more than 10 minutes causes a partial coagulation of proteins at

the water interface. These proteins cannot be dissolved and properly adsorbed at the interface and fail to form a coherent interfacial film. In addition, the viscosity of the liquid lamellae is not sufficient to make the foam stable (Stadelman & Cotterill, 1973).

2.5.1.1.3 Sodium chloride (NaCl) and Calcium ions (Ca+)

NaCl enhances foaming capacity of a protein solution but at low concentration. It usually minimizes surface viscosity and rigidity of protein films but increase spreading rate, thereby weakening interpeptide attractions and increasing foam volume for certain proteins. However, a high concentration of NaCl will decrease foaming (Oshodi & Ojokan, 1997). Ca^{+} can improve foam stability by forming bridges between the carboxylic groups of the protein.

2.5.1.1.4 Copper

Sagis, de Groot-Mostert, Prins & Van der Linden (2001) found that the foams prepared with copper ions in eggs took more time to foam, and were also more stable than fresh eggs. Copper ions form complexes with conalbumin and thus contribute to the stability of albumin foams (Warner & Trans, according to Sagis *et al*., 2001). The conalbumin copper complex is more stable against surface denaturation than native conalbumin. The formation of the copper conalbumin complex results in the binding of two positively charged copper ions per conalbumin molecule, effectively reducing the charge of the conalbumin molecule, and thus reducing the electrostatic repulsion between the film interfaces (Sagis *et al*., 2001).

2.5.1.1.5 Carbohydrates

The presence of carbohydrates reduces the foaming capacity but enhance foam stability (Linden & Lorient, 1999). The glycoprotein of the albumin particularly ovalbumin is related to its capacity to retain water in the lamellae. Foam expansion is minimized by the addition of sucrose and other sugars but the foam stability is improved, because they increase the bulk viscosity (Cheftel *et al*., 1985).

2.5.1.1.6 Fat

Significant concentrations of lipids adversely damage the foaming properties of proteins by placing themselves at the air/water interface, hence preventing competitive adsorption, the most favourable conformation of the protein films (Cheftel *et al.,* 1985; Linden & Lorient, 1999).

2.5.1.1.7 Dilution

Yang & Baldwin (1995) found that the addition of small quantities of water prior to whipping could increase the volume of foams. These foams are almost as stable as those made from albumins without water.

2.5.2 Coagulation and gelling capacity

Coagulation is a change from liquid (fluid) state to a solid or semisolid state (Yang & Baldwin, 1995). Egg proteins coagulate or thicken when heated. They have the ability to attract and hold large quantities of liquid, thus forming a gel or coagulum. A coagulum is also formed from egg protein molecules, as a result of mechanical action, acid, alkalies and other reagents such as urea (Yang & Baldwin, 1995). This quality makes eggs useful as a thickening agent in food products such as cakes, custards, sauces, scrambled, hard cooked and fried eggs (Mine, 1995). Heat coagulation of albumin is accompanied by changes in the secondary structure of proteins. The denaturation decreases the solubility of proteins in solution.

Pasteurisation generates heat, which brings about thermal coagulation. Thermal coagulation takes place at temperatures above 57°C in the case of albumin and in the case of the yolk it coagulates at temperatures above 65°C (Muriana, 1997). Ovalbumin and conalbumin are important for gelling properties. Lipoproteins such as phosvitin and livetins are heat stable and they are not susceptible to thermal coagulation but other proteins in the yolk are affected by heat (Linden & Lorient, 1999). The gelling properties of the yolk proteins depend on the lipoproteins. Donoval *et al*., according to Mine (1995) reported the denaturation temperatures of three major proteins in albumin ovotransferrin (61°C), lysozyme (75°C) and ovalbumin (84°C) at pH 7 as shown (Table 1).

2.5.3 Emulsifying properties of the yolk

Egg yolk is an important emulsifying ingredient in the manufacture of mayonnaise, salad dressing and cakes (Fennema, 1985). The components of the yolk responsible for emulsification are phospholipids and lipoproteins. Proteins such as lipovitellin and lipovitelinin help to reduce surface tension and facilitate the formation of the emulsion, but do not influence stability (Linden & Lorient, 1999).

2.5.4 Flavours and colours

Colour is the first visual impression of the acceptability of many foods. The colour of the yolk determines the attraction and acceptability of the eggs for consumers. The egg yolk imparts flavour in food products. The flavours are bound on the lipids of the yolk and consist of volatile compounds (Linden & Lorient, 1999).

2.5.5 Binding capacity

Egg yolk has excellent binding properties, with water holding, lipid retention and adhesion properties. In addition, albumin proteins have anti crystallisation capacity of sucrose in saturated solution, improve the homogeneity and the texture of confectionery products (Linden & Lorient, 1999).

2.6 Factors that influence the physico-chemical properties of albumin proteins

From egg laying to consumption, chemical, physical and biological changes occur in the egg (Stadelman & Cotterill, 1995). These changes depend on the conditions of storage of the eggs, which include time, temperature and relative humidity of the storage environment (Berardinelli, Donati, Giunchi, Guarnieri & Ragni, 2003).

2.6.1 Heat damage

Pasteurisation temperature (57°C) reduces the foaming capacity of albumins but does not affect emulsifying capacity of the yolk (Linden & Lorient, 1999). The appearance of albumin heated to 60°C begins to change becoming slightly opaque. The increase in opaque appearance of albumin has been observed as the temperature increases from 63-72°C, as a result a coagulum has formed (Woodward & Cotterill, 1983). Heat damage to the protein increases whipping time and decrease cake volume. The damaged albumin protein produces lower foam volume and therefore is less desirable in making meringues. If proteins are damaged, then foam volume will decrease and the liquid drainage from the whipped foam will increase (Davidson, 2004). According to Schuman, Sheldon, Vandepopuliere & Ball (1997) the Haugh unit values increased while the yolk index and albumin pH values were not affected during heat processing of shell eggs (57°C).

The thermal coagulation of albumin proteins into a gel is shown in Fig. 3. When albumin proteins are heated they change in protein conformation from the native state

to a partially unfolded state. Ovalbumin, ovotransferrin and lysozyme form intermolecular-β sheets. The formation of a stable intermolecular - β sheet structure is essential in the thermal denaturation and aggregation of albumin (Mine, 1995).

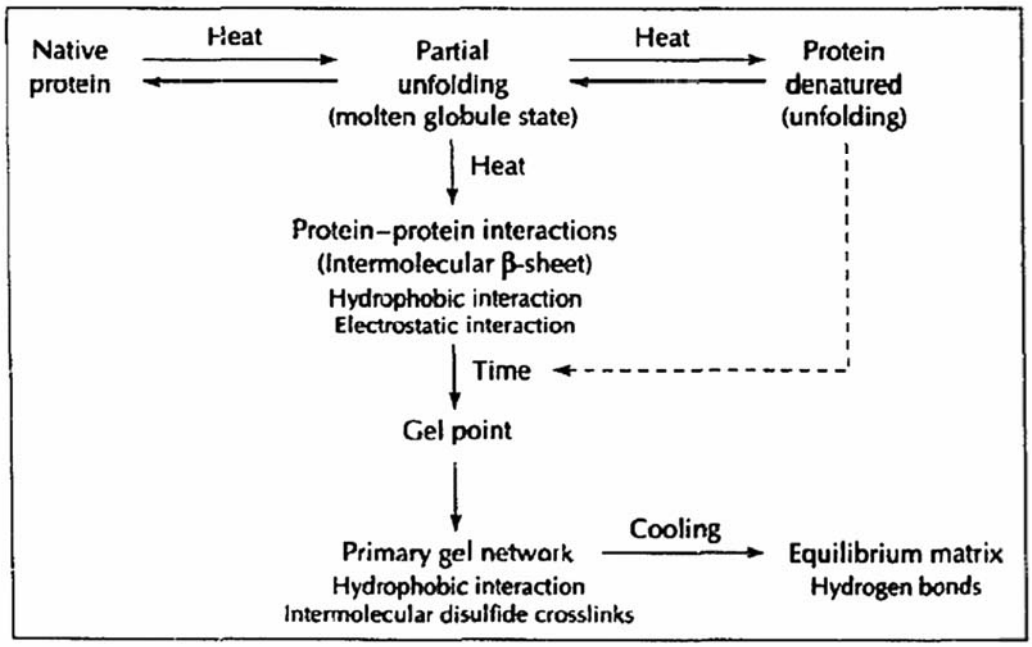

Figure 3. Flowchart of heat induced gelation of albumin proteins (Mine, 1995)

During this transition, functional groups that are involved in intermolecular hydrogen-bonding and electrostatic and hydrophobic interaction in the native state become available for intermolecular interactions under favourable conditions, resulting in a gel network. Intermolecular disulphide cross-links are not necessary for gelation but can produce more stable gels. The formation of disulphide bonds in albumin contributes to the structure of the formed gel and increases the gel strength (Mine, 1997).

2.6.2 Age of eggs

Generally, the average quality of albumin decreases with age and this may explain the changes in egg structure (Solomon, 1997). As the egg ages, egg quality deteriorates and the rate of deterioration is increased with higher storage temperatures (Stadelman & Cotterill, 1995). Aging influences the vitelline membrane and causes it to lose weight. Eggs lose weight through loss of water and carbon dioxide (CO_2), which volatilize through the membranes and the porous shell during storage. At the same time, the air cell size enlarges and the physicochemical properties of the albumin change; albumin gradually becomes clear and less viscous and shows a rise in pH value (Messens, Duboccage, Grijspeerdt, Heyndrickx & Herman, 2004; Solomon, 1997). Degradation of

one of the major structural glycoproteins and the disulfide bonds of the ovomucin causes a loss in membrane integrity over time (Kato *et al*, according to Kirunda & McKee, 2000). Elasticity of the vitelline membrane decreases with age. During aging, water is transferred from the albumin to the yolk, and result in a change in viscosity and egg yolk weight. Aged eggs exhibit higher albumin and yolk pH compared to fresh eggs. These changes are accelerated by a rise in temperature, which leads to an increase in pH of the albumin to 9.26 (Messens *et al*., 2004), and from 7.6 to 9.7 (Vaclavick, 1999). A decrease in Haugh units as a consequence of albumin thinning due to aging has been observed (Kirunda & McKee, 2000). According to Li-Chan *et al.* (1995) the formation of the heat stable form of ovalbumin known as S-ovalbumin occurs during storage of shell eggs. In addition, the dissociation of the ovomucin-lysosyme complex, with destruction of the ovomucin gel results in a partial loss of gelling and foaming properties.

2.6.3 Coating of shell eggs with oil

The use of coating agents such as vegetable oils (mineral oils), water glass, paraffin, acrylic acid resins, polyvinyl alcohol and sodium carboxymethyl cellulose have been reported in the literature (Hisil & Otles, 1997). Coating shell eggs closes the pores that are found on the shell surface to reduce evaporation and CO_2 escape therefore more CO_2 is retained and the albumin pH increases more slowly (Hisil & Otles, 1997). By using these treatments, loss of egg weight can also be minimized (Wells & Belyavin, 1987). Bakalinova (1998) found that coating of shell eggs with grease before storage maintains their functional properties as compared to uncoated shell eggs. In addition, oil coating extends the shelf life of shell eggs in weeks if they are stored at refrigeration temperature (Australian Centre for International Agricultural Research, 1997).

2.7 Microwave heating

Microwave heating is the conversion transfer of electromagnetic energy to thermal energy through direct interaction of the incident radiation with the molecules of the target material. Substances that contain polar molecules like water, protein, some carbohydrates and fat absorb microwaves whereas metal surfaces reflect it. Microwaves cause polar molecules to vibrate at a high frequency, hence producing a friction heating effect. Zhang *et al*. (according to Food and Drug Administration, 2000) reported some critical factors such as the volume, surface area, composition and shape of food that need to be considered when processing using microwaves. Microwave energy is used to

process the following products: ready prepared meals, granola, fresh pasta, and yogurt. The tempering and thawing of meat and fish, precooking of bacon, and sausage cooking represent the largest uses of microwave processing by the food industry (Bennion & Scheule, 2004).

2.7.1 Mechanism of microwave heating

Heating with microwave frequency involves primarily two mechanisms; dielectric and ionic. The primary component responsible for dielectric heating is water in the food. Due to its dipolar nature, water molecules have a tendency to follow the electric field associated with electromagnetic radiation. Heat is produced by oscillations of the water molecules. The second most important mechanism in heating using microwaves and radio frequencies is through the oscillatory migration of ions present in the food, generating heat under the influence of the oscillating electric field. The energy absorbed from the microwave frequency increases the temperature of food high enough to inactivate micro-organisms for effective pasteurisation (Food and Drug Administration, 2000).

2.7.1.1 Dipole rotation mechanism

Dipole rotation is the basic physical phenomena, which is responsible for the heating of food materials at microwave frequencies of 2450MHz (Funebo & Ohlsson, 1998). Water is the most common polar molecule in foods responsible for dielectric heating (Decareau & Peterson, 1986; Food and Drug Administration, 2000). The dipole rotation mechanism relies on the fact that water molecules have positive and negative ends thus it is a dipole. This mechanism depends on the existence of polar molecules and electrical conductivity of ions. When the dipole is subjected to a microwave field that quickly changes direction, the dipoles try to align their dipoles with the direction of the electrical field as shown in Fig.4.

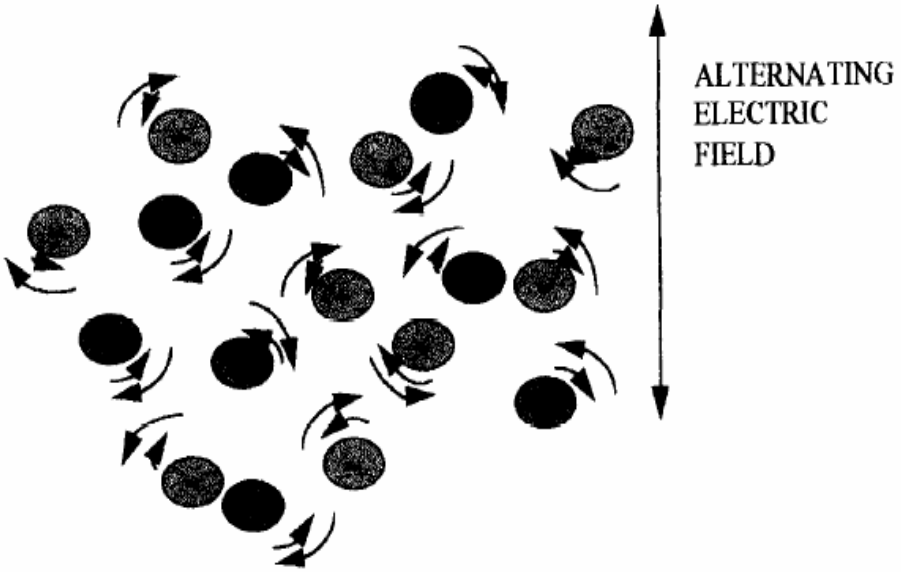

Figure 4. Dipole rotation mechanism (Khraisheh, Cooper & Magee, 1997)

The polar molecules move in the rapidly alternating electric field in a microwave oven. When the molecules rotate, the kinetic energy is dissipated as heat (Funebo & Ohlsson, 1998). According to Decareau (1985) the heat generated within the food is due to molecular friction primarily by the disruption of weak hydrogen bonds associated with the dipole rotation of free water molecules. When the field is removed it provides energy for the water molecules to rotate into alignment (Khraisheh *et al.*, 1997; Decareau & Peterson, 1986).

2.7.1.2 Ionic heating mechanism

Ionic heating involves the movement of ions in the food that generates heat under the influence of the oscillating electric fields (Decareau & Peterson, 1986; Food and Drug Administration, 2000). Ions hold an electric charge and are accelerated by the electric field. Kinetic energy is provided by the field to the ions, and collides with other ions, converting kinetic energy into heat that is given off (Decareau & Peterson, 1986). A high concentration of ions in foods will increase the absorption of energy at 2450MHz (Funebo & Ohlsson, 1998).

Hydrated ions, such as sodium and chloride from table salt, move in the direction of the electric field, and the electric resistance heating is achieved. The ions are surrounded by water molecules and randomly transfer energy to the water molecules during their movement. At higher temperatures they are very mobile and not tightly bound to ions.

Therefore the ions can then freely adsorb and dissipate a lot of energy. The conductive heating due to dissolved ions increases with a rise in temperature (Ohlsson & Bengtsson, 2001).

2.7.2 Factors influencing microwave processing of foods

2.7.2.1 Food composition

The rate of microwave power absorption in most materials is proportional to its water content. In microwave heating, the heat is not conducted through but is generated inside the body (material). Microwave heating reduces the time required for heating a body to a uniform temperature (Harlfinger, according to Nott & Hall, 1999). The dielectric properties of food products are primarily determined by the chemical composition, and to a lesser extent, by their physical structure. The molecular structure also affects the ability of the microwaves to interact with materials and transfer energy (Venkatesh & Raghavan, 2004; Ryynänen, 1995). The influence of water and salt (ash) content depends to a large extent on the manner in which they are bound or restricted in their movement by the other food components. The composition of a food influences the rate of heating. For instance, water, carbon, and food with high water content are good microwave absorbers (Decareau & Peterson, 1986). Fats and sugars have low specific heats compared to water; hence, foods that are high in fat or sugar heat more quickly than foods high in water. Foods with low density also heat more rapidly than high density foods when the same weights of these products are heated. Dense foods limit the depth of penetration of the microwaves. For example, a dense brownie batter heats much more slowly than a light, porous cake (Bennion & Scheule, 2004).

2.7.2.2 Shape and density of the material

The shape of the food material is important to obtain uniformity of heating. Food materials that have different shapes lead to local heating; similarly, sharp edges and corners cause nonuniform heating (Singh & Heldman, 2001). Heating patterns are a function of material variations such as sample shape and size as well as the water, salt, and sugar contents and processing variations (heating time and power delivery) (Nott & Hall, 1999).

2.7.2.3 Dielectric properties

Dielectric properties describe how materials interact with electromagnetic radiation. Natural biological materials absorb only the electric part of the electromagnetic field. Food materials are practically non magnetic, as they contain only tiny amounts of magnetic material, such as iron and cobalt (Mudgett, 1995).

Dielectric properties are properties that permit the passage of the lines of force of an electrostatic field but do not conduct the current (Bennion & Scheule, 2004). Dielectric properties are one of the most important factors in food affecting microwave heating performance, and describe how a material interacts with microwaves; that is the ability to absorb, transmit and reflect electromagnetic energy (Ryynänen, 1995). Different foods have different dielectric properties, uneven heating may occur in meals with several different components (Bennion & Scheule, 2004).

Temperature, salt content, moisture content and the state of moisture (frozen, free or bound) are the major factors that influence dielectric properties of agricultural and biological materials (Venkatesh & Raghavan, 2004). The dielectric properties also depend on the frequency of the applied alternating electric field, temperature of the material and on the density, composition and structure of the material. The bulk density of the air particle mixture in granular materials also influences their dielectric properties. The dielectric properties of materials depend on their chemical composition and particularly on the permanent dipole moments with water and any other molecules making up the material of interest. Polarisation arising from the orientation with the imposed electric field of molecules, which have permanent dipole moments, contributes to the frequency dependence of the dielectric properties (Venkatesh & Raghavan, 2004).

Dielectric constant is defined as the ability to store energy in the material. Dielectric loss factor indicates the energy losses. The influence of the dielectric properties on the heating of materials by absorption of energy through radio frequency dielectric heating, whether at high frequencies or microwave frequencies has been well known for a long time and many potential applications have been investigated. The concepts of dielectric heating have become more popular in industrial microwave heating (Venkatesh & Raghavan, 2004).

2.7.3 Effect of microwaves on survival of micro-organisms

The effect of microwaves on microoganisms present in foods are influenced by the intrinsic characteristics of the products being processed (pH, humidity, antibodies present, biological structures, chemical composition, amount and shape of the food) and extrinsic factors such as temperature, humidity, ambient gases, frequency and intensity of the radiation, time of exposure, and position of the foods in relation to the effective radiation field. Chemical and physical composition of the microorganisms being irradiated, their stage of development (vegetative cell, spore or development phase, wet or dry, etc.) and their initial amount are also influential (Valsechi, Horii & De Angelis, 2004). The energy absorption from microwaves increases the temperature of the food high enough to inactivate microorganisms for effective pasteurisation or sterilisation. Kindle, Busse, Kampa, Meyer-konog & Daschner (1996) found that microwaves of 2450MHz frequency and a power of 600W reduce colony counts of species such as *Escherichia coli, Enterobacter sakazakii, Klebsiella pneumoniae, Staphyllococcus aureus, Candida albicans* and *Poliomyelitis* vaccine virus in milk (150 ml) that was exposed for 85-100 seconds depending on milk type. At these conditions, *Pseudomonas aeruginosa* and *Mycobacterium terrae* were totally destroyed. Microwave heating has been reported to inactivate pathogenic micoorganisms including *Escherichia coli*, *Enterococcus*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Salmonella* and bacteria *Bacillus cereus*, *Campylobacter jejuni*, and *Clostridium perfringens* in poultry, beef, fish, pork products, milk, and eggs (Heddleson *et al*. according to Food and Drug Administration, 2000).

2.7.4 Inactivation mechanisms of micro-organisms by microwaves

The first mechanism for inactivation by microwaves involves thermal effects. Microwaves inactivate microorganisms entirely by heat through mechanisms comparable to other biophysical processes induced by heat, such as denaturation of enzymes, proteins, nucleic acids, and other vital components as well as disruption of membranes (Heddleson & Doores, according to Food and Drug Administration, 2000; Datta & Davidson, 2000). The microbial destruction is the result of the penetration of electromagnetic waves into a biological wet material, heating up the intra and extracellular fluids by the transfer of energy from polar water molecules and dissolved ions. This results in the generation of heat within the material itself due to molecular activity (Senize, according to Valsechi *et al*., 2004).

The second mechanism for inactivation by microwaves involves non-thermal effects. According to Yaghmaee & Durance (2005), Woo and others studied the effect of microwave irradiation on *E. coli* and *Bacillus subtilis* and reported protein and DNA leakage, adverse damage on the surface of cells and cell walls and appearance of dark spots in bacterial cells as a result of microwave treatment. There are four dominant theories used to explain non-thermal inactivation by microwaves: selective heating, electroporation, rupture of the cell membrane, and magnetic field coupling (Kozempel *et al*., according to Food and Drug Administration, 2000). The selective heating theory states that micro-organisms are heated more effectively by microwaves than the surrounding medium and therefore killed without difficulty. Electroporation is caused when pores form in the membrane of the microorganisms due to electrical potential across the membrane, resulting in leakage. Cell membrane rupture is related to electroporation in that the voltage drop across the membrane causes it to rupture. In the fourth theory, cell lysis occurs due to absorption of electromagnetic energy with critical molecules within the cells, disrupting internal components of the cell (Food and Drug Administration, 2000).

CHAPTER 3

HYPOTHESES AND OBJECTIVES

3.1 Hypotheses

- Microwave pasteurisation of WSE will not affect the functional properties and acceptability of the appearance of raw broken out eggs, as well as the flavour of cooked eggs adversely because the internal egg temperature will not be high enough to denature proteins in the egg albumin.

- Oil coating will prolong the shelf life of WSE stored at different conditions. The oil will seal the pores on the surface of the shell and this will reduce the evaporation of moisture and CO_2 from eggs.

3.2 Objectives

- To determine the effect of microwave pasteurisation of WSE on the pH, Haugh units, foaming and sensory properties, as well as consumer acceptance of WSE.

- To determine the effects of oil coating and different storage conditions [16ºC (58 % RH); 25ºC during the day and 15ºC at night (55 % RH) and 32ºC (32 % RH)] on the functional properties and shelf life of WSE.

CHAPTER 4

EFFECT OF MICROWAVE PASTEURISATION ON THE FUNCTIONAL PROPERTIES OF RAW WHOLE SHELL EGGS

4.1 Abstract

The effect of microwave pasteurisation at 250W and 300W on the functional properties of raw whole shell eggs (WSE) was evaluated. WSE obtained from Eggbert Eggs (Pty) Ltd were pasteurised in a first prototype pilot-plant microwave oven at 250W and 300W respectively, and eggs were collected from the left and right hand sides of the oven conveyor belt. Microwave pasteurised eggs had significantly ($p<0.001$) lower foaming capacity than control (unpasteurised) eggs and had higher Haugh values due to protein coagulation. There was no significant difference ($p>0.05$) in the foam stability of control and microwave pasteurised eggs and the pH of the albumin was almost similar. The yolk pH of pasteurised eggs was significantly ($p<0.05$) higher than that of unpasteurised eggs. A 16 member trained sensory panel evaluated the sensory properties of broken out raw eggs and found that there were significant differences ($p<0.001$) among the sensory attributes of the treatments. At 300W, pasteurised eggs collected from the left side of the oven had partially coagulated albumin and it was not clear in appearance as compared to unpasteurised eggs. This may be due to the formation of localized energy hot spots resulting from angles at which microwaves were directed inside the oven. The visual appearance of microwave-pasteurised eggs at 300W from left side was more adversely affected than the eggs pasteurised at 250W and collected from the right side oven position. A triangle taste test was conducted to see if a significant difference existed between control scrambled and scrambled eggs prepared from pasteurised eggs. There was a significant difference between control and pasteurised (300W) eggs. A home use test was also conducted to see if consumers will find microwave pasteurised (250W) eggs as acceptable as control eggs. The results showed that control and microwave pasteurised eggs were equally acceptable.

Keywords: Shell eggs; microwave pasteurisation; Haugh value; pH; foaming properties; descriptive sensory panel; Home use test; overall acceptability.

4.2 Introduction

Eggs are potential carriers of pathogenic microbes like *S.* Enteritidis due to their rich nutritive value. More than 90% of food borne salmonellosis cases result from consumption of shell eggs (Woodward, Khakhria & Johnson, 1997). *S.* Enteritidis is a causative agent of salmonellosis (Todd, 1996). Several methods such as fumigation, disinfection, egg washing and grading have been used to control this pathogen but they are only effective against post laying contamination (Tellez, Trejo, Sanchez, Ceniceros, Luna, Zazua & Hargis, 1995), they do not destroy salmonella which is harboured inside shell eggs (Catalano & Knabel, 1994).

Pasteurisation is aimed to eliminate *S.* Enteritidis (Hou *et al*., 1996). According to Tellez *et al*. (1995), pasteurisation is the only control measure against internally infected eggs. Heat pasteurisation is predominantly used in the food industry for its efficiency (Lado & Yousef, 2002), safety and relatively low cost. Pasteurisation is a process whereby food materials are heated below 100°C for a specific time to inactivate pathogenic and non-pathogenic organisms and to extend the shelf life (Fellows, 2000). However, heat treatment has an effect on the functional properties of proteins (De Witt, 1981). Excessive heat may cause undesirable protein denaturation (Lado & Yousef, 2002) more especially on the albumin that is more sensitive to heat (Stadelman & Cotterill, 1973). *S.* Enteritidis needs to be destroyed without damage to the egg proteins.

This phase evaluated the functional properties of eggs pasteurised using microwave technology described in CSIR patent no: PCT/IB2005/001079 (Erasmus & Rossouw, 2005). The purpose of this study was to determine the effect of microwave pasteurisation at 250W and 300W on the functional properties such as foaming capacity and foaming stability of WSE. The magnetron of the first prototype pilot plant microwave oven used for this study was unable to direct microwaves uniformly in the oven cavity leading to oven position, either left or right, being identified as an experimental variable. Due to the inability to uniformly distribute heat in this prototype microwave oven, the formation of hot spots in areas where internal egg temperatures differed occurred. The microwave oven used in this study was only a first prototype build. The results of this and other studies have shown that improvements in the microwave cavity design were needed and those were subsequently implemented into

the follow-up prototypes. The current unit at Arendsnes is giving quite consistent homogeneous heating throughout the rows after re-designing of the cavities long after this project trial was done. The value of this study lies in the proof of concept, even though the machine was not perfect as it was a world first prototype of its kind. Another objective was to determine the effect of microwave pasteurisation on the sensory properties of raw broken out eggs. A triangle test was used to determine if a significant difference existed between control and microwave pasteurised (300W) eggs. This test was done on cooked scrambled eggs. Lastly, a consumer home use test was also used to determine the acceptability of microwave pasteurised (250W) eggs by consumers.

4.3 Materials and methods

4.3.1 Pasteurisation of eggs

Eggs were obtained from a commercial egg packing plant, Arendsnes (Eggbert Eggs (Pty) Ltd, South Africa). Two day old eggs between 60g and 65g were collected. They were graded and packed in trays containing 30 eggs and then stored in an incubator at 30°C for two days. One hundred and twenty eggs from the incubator were selected randomly and served as controls. The microwave oven, a small-scale pilot plant prototype (Fig 5) was custom build by Delphius Technologies, as described in CSIR patent no: PCT/IB2005/001079 (Erasmus & Rossouw, 2005), was switched on for 60 min during processing. A total of 480 eggs were also selected randomly, labelled and transferred from the 30-egg trays to 6-egg trays. Half of the eggs were pasteurised at 300W for 40-45 min, and the other half was pasteurised at 250W for the same time period. At the end of the cavity, eggs were randomly selected at regular intervals to determine the internal temperatures on the top and on the bottom of individual eggs by making a tiny hole on the surface of the shell and inserting a temperature probe inside the egg. On the left hand side of the oven the recorded average egg temperature was 57°C and on the right side 48°C. After pasteurisation, the eggs were unloaded and packed in paper ptrays holding 30 eggs. These were stacked in boxes and stored at 25°C. One day after pasteurisation, eggs were transferred to a room where they were stored at 18°C. Sensory and functional analyses were conducted within a 7 day period. The experiment was repeated three times over a 3 week period. After selection and processing, there were 5 treatment groups in this experiment: Control, 300W/Right, 300W/Left, 250W/Right, and 250W/Left. One week after analyses, the remaining eggs were transferred to a cold room (4°C) and then stored for 3 weeks prior to the home use and triangle tests.

Figure 5. Prototype pilot-plant microwave oven unit. The conveyor belt is visible in the front, and two rows of eggs were packed in this continuous system**.**

4.3.2 Haugh units

The Haugh unit values of six randomly selected eggs per treatment group were determined once for each replicate. The WSE were individually broken on a glass plate and the height (mm) of the albumin was measured at six areas using a micrometer (Technical Services and Supplies, England).

The Haugh unit values were calculated using the following formula (Monira, Salahuddin, & Miah, 2003),

Haugh Unit =100 log ($H-1.7W^{0.37}+7.6$)

Where H = Average height of albumin (mm); W = Mass of whole shell egg (g)

4.3.3 pH and foaming properties of albumin

The albumin and yolk pH as well as albumin foaming properties for each treatment group were measured in duplicate for each replicate. Per replicate, three shell eggs from each treatment were individually broken using a knife. A plastic egg separator placed on top of a 600 ml glass beaker was used to separate the yolk from the albumin. The yolk pools of the three eggs were collected in a stomacher bag and then homogenised

for 30s using a stomacher lab blender (Art Medical Equipment (Pty) Ltd, Johannesburg). The liquid albumin was transferred to a stomacher bag, homogenized for 30s and then returned to the beaker. After homogenisation, the height (mm) in a 600 ml beaker was measured. The pH and temperature of the homogenized albumin and yolk, respectively were measured, using a Hanna pH 211 Microprocessor. A Braun Multiquick ® professional MR 55550 M CA mixer was immersed in the albumin mixture in the beaker and whipped at speed 1 for 3 min. Note that the temperature of albumin ranged between 18.7 ºC and 24.8 ºC due to day time temperature fluctuation. This was an unfortunate situation as it probably influenced the results. It should have been better controlled. The height of foam was measured as well as the drainage height after 60 min. The following formulas were used to calculate the foaming capacity and foam stability (adapted from Chang & Chen, 2000).

Foam capacity (%) = {Foam height (mm)}/{Unwhipped albumin height (mm)} X 100
Foam stability (%) = {[Foam height (mm)-Drained liquid height*(mm)] / Foam height (mm)} X 100

* Drained liquid height (mm) = Height of liquid phase after 60min (mm)-Height of liquid phase after 30s (mm)

4.3.4 Descriptive sensory evaluation

Potential panellists were recruited on the University of Pretoria main campus. The poster used to recruit panellists for descriptive sensory evaluation is shown in Appendix A. Individuals that were interested were invited to attend an information session. The session was held in the lecture theatre (Room 2-35) of the Department of Food Science. The details about availability, promptness, and expectations of panellists were discussed during this session as well as the reward system. A screening questionnaire (Appendix B) containing questions on general health, chronic health conditions, food allergies and availability were also completed during this session.

Two screening sessions were conducted over two days to select the final panel of 16 that participated in the evaluation. On each screening day, two sessions (10h30 and 12h30) were conducted and panellists were asked to attend one of these sessions on each screening day in the sensory laboratory. During the first screening session the potential panellists evaluated ten sets of egg samples. Each set consisted of five broken out eggs, the one sample was coded as the control sample, while the other four samples

were blind coded with randomly selected three digit codes. The panellists were asked to compare the visual appearance of each blind-coded egg sample with the appearance of the control (fresh) eggs, using the following scale:

1=Same as control

2=Slightly different to control

3=Moderately different to control

4=Very different to control

5=Extremely different to control

A copy of the evaluation form for the first screening day is shown in Appendix C. The different sets consisted of fresh eggs according to sell by date on packaging (served as control, bought from Woolworths, Garsfontein), slightly damaged eggs (collected from CSIR), severely damaged eggs (collected from CSIR) and old eggs (stored for more than 3 months). The slightly damaged and severely damaged egg samples were pasteurised eggs but with different degrees of coagulation of the albumin. The eggs were broken out of the shell onto square Styrofoam plates (130 x 180mm) (70/120 Atlantic Black), evaluated by panellists and then classified according to how similar or different to the control they were. If the blind coded samples were found to be different from the control sample, the panellists were prompted for comments to describe the differences.

During the second session of screening, triangle tests (Meilgaard, Civille & Carr, 1987) were used to evaluate ten sets of eggs varying in degrees of albumin denaturation and storage age. In a triangle test, three samples are served to the panellist. Two samples are the same and one is different or odd (Meilgaard *et al*., 1987; Lawless & Heymann, 1998). The evaluation form for the triangle tests is shown in Appendix D. The results of the two screening sessions were used to choose the final panel of 16 members. The panel consisted of thirteen females and three males between the ages of 20 and 34 years. The ability of the panellists to describe the differences was also judged by considering the ability to verbally express visual appearance of the eggs.

To visually evaluate the quality of raw eggs and specifically in terms of heat damage, the panel was trained using generic descriptive analysis (Einstein, 1991). The training included exposure to a wide range of eggs representing difference in quality, mainly related to effects of storage age and degree of heat coagulation. Training consisted of a total of 6 hours, two hour sessions per day over a period of 3 days. Sensory descriptors

were developed, defined (Table 2) and agreed upon by the panellists to differentiate control and pasteurised eggs (250W and 300W). Good sensory practices in preparation of samples, sample coding, order of presentation, and use of individual booths were used as described by Lawless & Heymann (1998). To prepare samples, sixteen eggs from each treatment (16 panellists x 5 treatments) were broken out onto square Styrofoam plates (130 x 180mm) (70/120 Atlantic Black). The plates were coded with randomly selected three digit codes. The egg samples were presented to individual panellists to assess the visual appearance of broken out raw eggs and rate the intensity of each sensory attribute. One person evaluated 5 eggs per session. Each person was given five individual eggs to look at. The panel rated all the attributes for each treatment on a numbered (9-point) scale anchored at both sides with sensory descriptors (Table 2 and Appendix E). When evaluating the colour of yolk, colour fans (Roche-Switzerland) with 15 colour references were used representing a 15-point scale. There were two 1-hour sessions per day in the morning at 10h30 and in the afternoon at 12h30. The evaluation of egg samples was conducted once per week including two sessions over a period of three weeks. The sensory evaluation of eggs was done in a sensory laboratory equipped with 16 individual booths.

Table 2. Sensory attributes and rating scale anchors used by the trained sensory panel to describe broken out raw eggs using 9 point category scales.

Sensory attributes of eggs	Scale anchors
Yolk size	1=Small; 9=Large
Yolk height	1=Low; 9=High
Brightness of yolk colour	1=Dull yellow; 9=Bright yellow
Yolk blemishes	1=Not blemished ; 9=Very blemished
Albumin firmness	1=Not firm and compact; 9=Very firm and compact
Albumin coagulation	1=Not coagulated; 9= Very coagulated
Prominence of chalaza	1=Not prominent; 9=Very prominent
Albumin clearness	1=Not clear; 9=Very clear
Yolk colour [1]	1=Light yellow; 15=Orange

[1] Roche egg colour fan (Roche-Switzerland) with 15 colour references (15-point scale).

4.3.5 Sensory evaluation of cooked eggs

The triangle test (Meilgaard *et al*., 1987; Lawless & Heymann, 1998) was used to determine whether a significant difference existed between scrambled eggs prepared from control and microwave pasteurised eggs (300W). Eggs collected from the left and right hand side of the oven conveyor belt were pooled. The panel was recruited on and around the campus and comprised of 52 panellists consisting of students and staff members who consume eggs regularly (more than once per week). The panel consisted of 42% females and 58% males. Recruitment was done through an advertisement and flyers (Appendix F).

4.3.5.1 Sample preparation and serving

Fresh batches of scrambled eggs were prepared from control and microwave pasteurised (300W) eggs as follows: fifteen WSE stored at 18°C for 3 weeks and then at 4°C for 3 weeks were broken into a stainless steel bowl. The mixture of eggs was whipped to homogenize the yolk and albumin at speed 3 for 2 minutes (Peckham & Freeland-graves, 1987) using a mixer (Braun Multiquick ® professional MR 55550 M CA). Excella cooking oil (75ml) (Continental Oil Mills (Pty) Ltd, South Africa) was used to coat the base of a Pineware electric non-stick round frying pan (30mm diameter). The pan was set at maximum heat (setting 9) and heated for one minute. The mixture of eggs was poured in and cooked for 4 minutes while stirring using a plastic spatula. Scrambled eggs from the frying pan were transferred into a glass bowl. About 15g of egg was scooped into clear glass ramekins and covered with aluminium foil squares (100 mm x 100 mm) and then placed in a pre-heated AEG oven (50°C) to keep warm. For each panellist, a set of 3 samples of scrambled eggs were placed on a white tray in the order of presentation and coded with randomised three digit numbers. The consumers were presented with the samples, an evaluation form (Appendix G), forks, and serviettes. Evaluation of eggs also took place in the sensory laboratory. There was three sessions scheduled per day at 9h30, 11h00 and at 12h30 respectively.

4.3.6 Consumer sensory evaluation

A consumer home use test was done to determine the acceptability of microwave pasteurised eggs by consumers. Fifty-four panellists (n=54) were recruited around the campus of the University of Pretoria through campus e-mail. To participate in the home use evaluation, potential panellists were selected and screened telephonically. The

selection was based on the following criteria: egg consumption, willingness to participate, availability and a lack of food allergies.

WSE stored at 18°C for 3 weeks and then at 4°C for 3 weeks were used for the consumer home use test. One control and one pasteurised (250W) egg, either from left or right hand side of oven (coded with random three digit numbers) served as a set. The two eggs were placed in a transparent plastic container with a lid and placed inside a white plastic shopping bag that was labelled with a set number. Each person was given one set together with an evaluation form (Appendix H). Consumers were given the following options: boiling, frying, poaching, and scrambling as cooking methods to prepare the eggs and then asked to indicate cooking method they have used during the evaluation.

4.3.7 Statistical analysis

The results obtained from the Haugh unit, pH and foaming properties measurements were statistically analysed making use of the software STATISTICA 7.1 (Stat Soft, Inc., 2005). Analysis of variance (ANOVA) was performed to determine whether microwave power level (250W and 300W) and oven position (left or right) significantly influenced Haugh units, pH and foaming properties of albumin. The significance of differences was based on the probability of a Type I error set at $p \leq 0.05$. Where necessary, the Least Significant Difference (LSD) test was used to detect significant differences between means of control and pasteurised eggs at 5% level. Data from descriptive sensory evaluation of eggs were obtained from three replicates and were analysed with ANOVA. The LSD test was also used to determine differences among the treatments.

The binomial distribution function (one-sided) in Microsoft ® Excel 2003 was used to analyse the results of the triangle test. For the consumer home use test, data was analysed with one way ANOVA at 5% level, in order to determine the effect of pasteurisation on the acceptability of the visual appearance and overall acceptability of WSE. Microsoft ® Excel 2003 was also used to analyse (descriptive statistics) the data collected from consumer home use questions. Consumers were asked to rank, in order of importance, factors such as brand name, freshness, packaging, price and visual appearance that are used to select eggs to purchase. This information was then analysed using comparison of the differences between rank sum totals using the critical value ($k_{n,\ p<0.05}$) obtained from the Basker table (Basker, 1988).

4.4 Results

The demographic characteristics, egg consumption behaviour and perceptions of consumers that participated in the home use test are summarised in Table 3.

Table 3. Summary of demographic characteristics and egg consumption behaviour of consumers that participated in the home use test (n=54).

		Percentage (%)
Gender	Male	22
	Female	78
Age (Years)	18-25	14
	26-35	35
	36-45	20
	45+	31
On average, how often do you eat eggs ?	Less than once per week	19
	Once per week	17
	2 times per week	29
	3 to 6 times a week	31
	Daily	4
When do you normally eat eggs?	Breakfast time	76
	Lunch time	12
	Dinner/supper time	12
Where do you normally store eggs after purchase?	In the refrigerator	69
	In the cupboard	31
	Other, please specify	-
On average, for how long do you normally store eggs after purchase?	1 week	16
	2 weeks	37
	3 weeks	21
	4 weeks	16
	More than 4 weeks	10

The majority of people who participated in this survey were female. Most people who participated in the survey consume eggs three to six times a week and most at breakfast time. The majority of consumers reported that they normally store eggs in the refrigerator, mostly for two to three weeks after purchase.

4.4.1 Quality of eggs

Microwave pasteurisation had a significant effect ($p<0.01$) on the Haugh units, yolk pH and foaming capacity of albumin (Table 4). Control and 250W/R eggs had significantly lower Haugh units than 250W/L, 300W/R and 300W/L treated eggs. Pasteurised eggs had a slightly higher yolk pH ($p<0.01$) compared to control eggs. Microwave pasteurisation treatment did not increase or decrease albumin pH ($p=0.05$). As shown in Table 4, control eggs had significantly higher ($p<0.01$) foaming capacity. No difference was found between the foam stability of the albumin from control (83% ± 2.3) and pasteurised eggs (87% ± 7.1)-results not shown.

There were significant differences ($p<0.01$) between control and microwave pasteurised eggs for sensory properties such as the yolk size, yolk height, brightness of yolk , yolk blemishes, albumin firmness, albumin coagulation, prominence of chalaza, and albumin clearness (Table 4). Control eggs had significantly smaller yolks than pasteurised eggs. Panellists rated yolks of eggs that were pasteurised at 300W and positioned on the left side of the oven as less intense in yellowness compared to the other eggs. Pasteurisation had a significant effect ($p<0.01$) on yolk blemishes (Table 4).

The albumin of pasteurised eggs was more firm compared to the albumin of unpasteurised eggs. No significance difference ($p>0.01$) was detected in albumin coagulation between control and 250W/R eggs. There was a significant difference ($p<0.01$) amongst 250W/L, 300W/R and 300W/L in albumin coagulation. Panellists perceived 300W/L eggs as much more coagulated compared to 250W/L and 300W/R eggs. There was a significant difference ($p<0.01$) in the prominence of chalaza between control and pasteurised eggs. The chalaza of eggs pasteurised at 300W/L was less prominent compared to all other egg treatments. There was a significant difference ($p<0.01$) in the albumin clearness between control and pasteurised eggs. The albumin of 300W/L eggs was less clear compared to the albumin of 300W/R eggs as seen in Table 4.

Table 4. Mean values ± standard deviation (SD) for sensory and quality attributes of raw broken out eggs microwave pasteurised at two different power levels and collected from either the right (R) or left (L) side of the microwave

Quality attributes	Treatment					
	Control	250W/R	250W/L	300W/R	300W/L	P-value
Haugh unit	57 (± 1.1)^{a}	64 (± 1.3)^{a}	72 (± 2.2)^{b}	75 (± 2.3)^{bc}	82 (± 2.5)^{c}	<0.0001
Yolk pH	6.0 (± 0.2)^{b}	6.5 (± 0.2)^{a}	6.5 (± 0.3)^{a}	6.5 (± 0.2)^{a}	6.4 (± 0.2)^{a}	0.0011
Albumin pH	9.2 (± 0.2)^{a}	9.3 (± 0.1)^{a}	9.3 (± 0.1)^{a}	9.2 (± 0.2)^{a}	9.3 (± 0.1)^{a}	0.138
Foaming capacity %	376 (± 22)^{c}	199 (± 37)^{b}	170 (± 18)^{a}	182 (± 19)^{ab}	178 (± 44)^{ab}	<0.0001
Sensory attributes of eggs						
Yolk size	4.6 (±1.7)^{b}	5.7 (±1.6)^{a}	6.0 (±1.8)^{a}	5.4 (±1.4)^{a}	6.1 (±1.7)^{a}	<0.0001
Yolk height	5.0 (± 1.5)^{b}	4.8 (±1.5)^{a}	4.8 (± 1.6)^{a}	5.1 (± 1.6)^{a}	6.2 (± 1.7)^{a}	<0.001
Brightness of yolk	5.5 (± 1.7)^{a}	5.8 (± 1.4)^{a}	5.5 (± 1.4)^{ab}	5.9 (± 1.6)^{ab}	5.1 (± 1.6)^{b}	0.0038
Yolk blemishes	1.4 (± 1.0)^{b}	1.3 (±0.9)^{ab}	1.3 (±1.0)^{ab}	1.4 (± 1.2)^{a}	1.4 (± 1.0)^{ab}	0.0096
Albumin firmness	5.2 (±2.1)^{b}	5.0 (±2.2)^{c}	4.8 (±2.6)^{c}	5.5 (±2.2)^{a}	5.4 (±2.2)^{a}	<0.0001
Albumin coagulation	2.3 (±2.2)^{c}	2.6 (±2.4)^{a}	2.8 (±2.5)^{b}	3.6 (±2.6)^{a}	4.7 (±2.9)^{d}	<0.0001
Prominence of chalaza	6.1 (± 2.6)^{a}	5.4 (± 2.4)^{bc}	5.5 (± 2.5)^{ab}	4.5 (± 2.8)^{d}	4.9 (± 2.6)^{c}	<0.0001
Albumin clearness	7.2 (±2.7)^{b}	6.3 (±2.9)^{d}	6.1 (±2.7)^{c}	5.3 (±2.9)^{e}	4.2 (±2.8)^{a}	<0.0001

^{abcde} Means within a row with different superscripts are significantly different ($p \leq 0.01$)

Yolk size: 1=Small; 9=Large,
Yolk height: 1=Low; 9=High,
Brightness of yolk : 1=Not intense; 9=Very intense,
Yolk blemishes: 1=Not blemished; 9=Very blemished,
Albumin firmness: 1=Not firm and compact; 9=Very firm and compact,
Albumin coagulation: 1=Not coagulated; 9= Very coagulated,
Prominence of chalaza: 1=Not prominent; 9=Very prominent,
Albumin clearness: 1=Not clear; 9=Very clear

A significant difference ($p<0.05$) was observed between replicates for albumin firmness, albumin coagulation, albumin clearness, yolk pH, albumin pH, foam capacity and stability (Table 5). During replicate 3, albumin firmness was rated as significantly ($p<0.01$) higher than albumin firmness in replicates 1 and 2. Albumin was more coagulated during replicates 2 and 3 compared to replicate 1. Albumin was significantly clearer during replicate 1 compared to replicates 2 and 3. Replicates 2 and 3 had significantly ($p<0.01$) lower yolk pH than the yolk pH in replicate 1. Replicate had a significant effect ($p<0.01$) on the albumin pH. Albumin pH was significantly ($p<0.05$) lower during replicate 2 compared to replicates 1 and 3. Replicate had an influence on the foaming capacity of albumin. The foaming capacity during replicate 2 was significantly ($p<0.01$) lower compared to replicates 1 and 3.

Table 5. The effect of replicate on the sensory and quality attributes of raw and pasteurised broken out eggs

Sensory attributes	Replicate 1	Replicate 2	Replicate 3	p-value
Albumin firmness	4.7 (±2.2)^{a}	5.0 (±2.4)^{a}	5.9 (±2.2)^{b}	<0.0001
Albumin coagulation	2.3 (±2.2)^{a}	3.7 (±2.8)^{b}	3.7 (±2.7)^{b}	<0.0001
Albumin clearness	6.8 (±2.6)^{b}	5.0 (±3.0)^{a}	5.4 (±3.0)^{a}	<0.0001
Yolk pH	6.6 (±0.3)^{b}	6.3 (±0.3)^{a}	6.3 (±0.2)^{a}	0.004
pH of albumin	9.5 (±0.03)^{c}	9.1 (±0.06)^{a}	9.2 (±0.02)^{b}	<0.0001
Foam capacity (%)	231 (±77)^{b}	201 (±101)^{a}	232 (±77)^{b}	0.01
Foam stability (%)	90 (±4.9)^{b}	86 (±6.2)^{b}	80 (±5.84)^{a}	0.003

^{abc} Means with different superscripts are significantly different ($p<0.05$)

Albumin firmness: 1=Not firm and compact; 9=Very firm and compact
Albumin coagulation: 1=Not coagulated; 9= Very coagulated
Albumin clearness: 1=Not clear; 9=Very clear

4.4.2 Sensory evaluation of cooked eggs

There was a significant difference ($p<0.05$) in the sensory properties of scrambled eggs from control and pasteurised (300W) samples (Table 6).

Table 6. The effect of pasteurisation of raw whole shell eggs at 300W on the sensory properties of scrambled eggs using a triangle test

Samples	Test
Number of respondents that had the test incorrect	25
Number of respondents that had the test correct	27
Total number of judges	52
Significance (p-value)	0.004

4.4.3 Consumer home use test

4.4.3.1 Visual appearance and overall acceptability of eggs as evaluated by consumers

No significant difference ($p>0.05$) was detected in the acceptability of the visual appearance of control and microwave pasteurised (250W) eggs (Table 7). There was no significant difference ($p>0.05$) found for the overall acceptability of the eggs. Cooking methods used by consumers are given in Table 8.

Table 7. Average consumer ratings (± standard deviations) for the visual appearance and overall acceptability of eggs (1=Dislike extremely; 9=Like extremely) (n=54)

Attributes rating	Control	Pasteurised (250W)	P-value
Visual appearance	7.00 (± 1.61)	6.31 (± 2.03)	0.064
Overall acceptability	7.10 (± 1.53)	6.69 (± 1.95)	0.252

Table 8. Cooking methods used by consumers to cook the eggs during the home use test

Cooking method	Percentage (%)
Boiling	26 %
Frying	42 %
Poaching	1 %
Scrambling	31 %

Most participants fried the eggs during the consumer survey followed by scrambling and boiling. There was a significant difference among factors considered by people when buying eggs ($p<0.05$). The order of importance of factors for consumers when buying eggs are summarised in (Table 9). Consumers selected freshness (sell by date

or expiry date), visual appearance of the shell as well as price as the most important factors when buying eggs. Packaging followed by brand name were the least important factors.

Table 9. The results of the ranking test comparing the factors that influence egg buying preferences of consumers. 1= Most important 5= Least important

Important factors	Rank sum totals
Brand name	163 [c]
Freshness	70 [a]
Packaging	139 [b]
Price	97 [ab]
Visual appearance of the shell	86 [a]

For n=37 panellists and 5 samples, Basker table (Basker, 1988) value for differences among rank sums is 37.1

[abc] Rank sums with different superscript differ significantly ($p \leq 0.05$)

Table 10 is a summary of the sensory attributes used to determine egg freshness by consumers that participated in the home use test survey. In this consumer survey, many people reported that they used taste ("flavour") and smell attributes in order to judge the freshness of cooked eggs while eating eggs. As shown in Table 10, few consumers reported that they used texture when judging the freshness of cooked eggs.

Table 10. Sensory attributes used by consumers when judging the freshness of cooked eggs while eating and the criteria used to determine freshness of raw broken out eggs.

Attributes	Percentage of consumers
Criteria used to judge the freshness of cooked eggs	
Smell	41 %
Texture	10 %
Taste	49 %
Criteria used to determine freshness of a raw broken out eggs	
Appearance of yolk	35 %
Smell	43 %
Colour of albumin and yolk	14 %
Albumin appearance	6 %
Other, specify	2 %

Consumers were asked about safety risks related to eggs in this survey. The level of concern about the safety of eggs, when eating chicken eggs is illustrated in Fig 6. Consumers are not really concerned about the safety risks related to eggs since more than half of the respondents responded that they were slightly or less concerned.

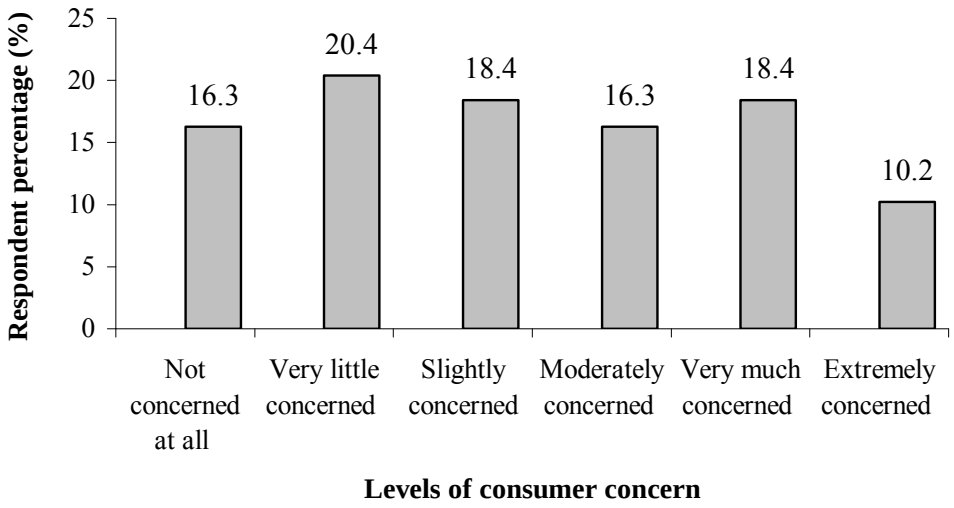

Figure 6. Levels of concern of consumers regarding potential safety risks of eggs.

4.5 Discussion

In the present study, pasteurised eggs had higher Haugh unit values than control eggs. The high Haugh units were more pronounced for eggs pasteurised at 300W compared to 250W eggs. Eggs pasteurised at 300W and located on the left side of the oven were adversely affected due to the angles at which microwaves were directed in the microwave cavity. A high Haugh unit value is generally good because it indicates fresh albumin quality of the eggs (Stadelman, 1986a). According to Schuman *et al*., (1997), an increased Haugh unit as a result of pasteurisation, was due to the impact of the thermal process on the albumin proteins leading to an increased albumin height of egg. This researcher used water bath immersion heat treatment to pasteurise shell eggs. The increase in Haugh unit values could probably be due to albumin coagulation, induced by pasteurisation of WSE. In contrast, Hou *et al*. (1996) conducted a study on the pasteurisation of intact shell egg and found that heating shell eggs at 57°C did not affect the Haugh units. In the present study, pasteurisation did not influence albumin pH of WSE. The age of shell eggs could have been an influence

on pH, as the pH of albumin increased within 3 days after laying. These eggs were pasteurised when the pH had already increased. Hank, Kunkel, Dawson, Acton, and Wardlaw (2001) heated shell eggs for 180 min at 55°C in a hot air oven and observed no changes in the pH of albumin. There was no valid reason provided to explain why albumin pH did not change. In a study done by Schuman *et al*. (1997), the pH of albumin was not affected by heat treatments. Pasteurised eggs had a slightly higher yolk pH compared to control eggs.

The position in the oven had an effect on the quality of eggs that were pasteurised. Quality of eggs that were positioned on the left hand side of the oven and pasteurised at 300W was the most severely affected. The albumin of these eggs was visibly coagulated, not clear in appearance and the chalazae were not prominent as compared to those of eggs pasteurised at 300W but at the right hand side.

Microwave pasteurisation induced changes in the functional properties of eggs. No significant differences in foaming capacity of albumin were observed among eggs pasteurised at 250W and 300W. However a significant difference was found between control and pasteurised eggs. It was reported that high pasteurisation temperature decreases the foaming capacity of the albumin (Linden & Lorient, 1999). High pasteurisation temperature causes the proteins to denature. The decrease in foaming capacity was probably due to denaturation of some albumin proteins during heating. Ponce, Pla, Sendra, Guamis & Mor-Mur (1999) mentioned that heating of eggs resulted in the destruction of covalent bonds and other adverse changes and reduced the quality of the product. Foaming ability of the albumin depends on the quality of its proteins (Kirunda & McKee, 2000). The ability of protein to form and stabilize foams depends on the source of protein and degree of denaturation, other composition effects such as fat, the presence of calcium ions, size and flexibility of proteins, pH, temperature, whipping methods and time (Cheftel *et al*., 1985). Pasteurisation can denature proteins by breaking hydrogen bonds involved in secondary and tertiary protein structure, that exposes the disulfide (SS) linkages (Wong & Kitts, 2003). Breaking of disulfide bonds by reducing agents may change the conformation of a protein molecule (Charley & Weaver, 1998). When the bonds that maintain the tertiary and secondary structures of a protein molecule are disrupted, the polypeptide loses its characteristic spatial arrangement (Charley & Weaver, 1998). Application of

microwave pasteurisation decreased the foam volume of the albumins. Slight denaturation of proteins in albumin could explain the reduced foam capacity of pasteurised eggs (particularly 300W). The most important factors causing denaturation are changes of temperatures, pH value and pressure (Goetz & Koehler, 2004). However, pasteurisation did not significantly ($p>0.05$) affect the foam stability of the albumins. A significant difference was observed between replicates for the foam stability of albumin. Foam stability was higher for replicate 1 compared to replicate 3. Ovomucin is the main component of albumin that forms a film of insoluble material that stabilises the foam (Wong & Kitts, 2003). Therefore, a difference in levels of denaturation of ovomucin may explain the differences in foam stability.

According to Lee & Chen (2002) foaming depends on the ability of proteins to rapidly form cohesive interfacial films that are able to entrap and retain air. The content of free soluble ovomucin in both firm and thin albumin could make the protein more available for adsorption at the film surface of the foam and for lowering the surface tension, thereby stabilising the foam of both firm and thin albumin against liquid drainage.

The pasteurised eggs had firmer textures compared to control eggs and the viscosity of the albumin was also affected. Albumin firmness could probably be due to protein denaturation that occurred during microwave pasteurisation of WSE. Albumin of pasteurised eggs was more firm and compact. The albumin of eggs pasteurised at 300W/L was less clear. Heating caused some changes to the physical properties of WSE. Unlike control eggs, the appearance of albumin was milky white in colour for pasteurised eggs. These results are supported by those of Hou *et al.* (1996) who found that the viscosity of albumin decreased and turbidity increased after heating treatments and suggested that these changes indicated a partial protein denaturation.

Microwave pasteurisation did not affect the yellowness of the yolk as rated by trained panellists. These results agree with the work done by Hou *et al.* (1996) who revealed that heating did not influence the colour of eggs.
There was a significant difference in the texture of cooked pasteurised and control eggs. Consumers found that control eggs were softer than the pasteurised (300W).

Chalazae in eggs that were positioned on the left hand side of the microwave oven and pasteurised at 300W were not as prominent as compared to pasteurised (250W) eggs. Less prominent chalazae was due to albumin coagulation that also changed the colour of albumin from cloudy to white. Smell and visual appearance of yolk are the attributes mostly used by many consumers to determine the freshness of raw or uncooked broken out eggs. For cooked eggs, consumers use sensory attributes such as taste and smell to judge the egg freshness when eating them. Consumer sensory evaluation was done to determine the acceptability of pasteurised shell eggs by consumers. It was found that consumers accepted control and pasteurised (250W) eggs similarly. Oderkirk, according to Lepule (2003) found that for consumers, egg quality includes aspects such as cleanliness of the product and attractive packaging. They are also concerned about cracks, colour, and freshness.

4.6 Conclusions

Microwave pasteurisation reduces the functionality of albumin particularly foaming capacity. The albumin quality of the egg was also affected, as the Haugh unit values increased as a result of the heat applied. This process also influences the physical or visual appearance and sensory properties of raw eggs such as albumin firmness, albumin coagulation, prominence of chalazae and albumin clearness. Eggs pasteurised at 300W were more adversely affected compared to eggs pasteurised at 250W. More severe damage was observed in the eggs pasteurised on the left hand side of the cavity than eggs on the right hand side, because the internal temperature of the eggs on the left side was higher. Hence, eggs pasteurised at 300W on the left hand side can not be used to prepare either sponge cake or meringues, because they will not produce high quality foam. It is better to use 250W to microwave pasteurise eggs using the prototype pilot-plant microwave oven unit. Variations in microwaves directed at the eggs need to be fixed because it causes variation in quality.

Consumers accepted the visual appearance of both pasteurised (250W) and control eggs similarly. Microwave pasteurised (250W) eggs have a potential of being accepted by the consumer market because visual appearance and overall acceptability did not differ from control eggs. Pasteurised (300W) eggs had an influence on some sensory attributes of cooked eggs. Consumers were able to detect the difference between pasteurised (300W) and control eggs.

CHAPTER 5

EFFECT OF OIL COATING ON THE FUNCTIONAL PROPERTIES AND SHELF LIFE OF WHOLE SHELL EGGS (WSE) STORED UNDER DIFFERENT CONDITIONS

5.1 Abstract

The effect of oil coating on the functional properties and shelf life of WSE stored under different conditions were studied. Coated and uncoated WSE that were stored at 16ºC (58% RH); 25ºC during the day and 15ºC at night (55% RH) and 32ºC (32% RH) for a period of six weeks were evaluated. Haugh units, pH of yolk and albumin, foaming capacity, foam stability and descriptive sensory properties of the eggs were measured. Haugh units of eggs decreased with storage time at all storage conditions but for coated eggs it decreased at a slower rate. Coated shell eggs had a significantly lower weight loss over time than uncoated shell eggs. The pH of both the yolk and albumin of coated shell eggs was significantly ($p<0.05$) lower than that of uncoated shell eggs. Uncoated shell eggs had better foaming capacity as compared to coated shell eggs. Coating did not have an influence on the foam stability of egg albumin. Significant differences ($p<0.05$) were found between the visual sensory properties of coated and uncoated broken out eggs that were stored at 16ºC; 58% RH and at 25ºC & 15ºC; 55% RH for seven weeks as assessed by a trained sensory panel. Although there was no significant interaction effects with regard to the height of albumin, position of yolk, firmness of albumin and prominence of chalazae for the broken out raw eggs stored at 32ºC; 32% RH, significant differences were found between coated and uncoated shell eggs. Coated shell eggs stored at the three conditions had a prolonged shelf life compared to uncoated eggs stored in the same manner.

Keywords: Whole shell eggs; Mineral oil; Egg weight; pH; Foaming capacity; Foam stability; Haugh unit; Sensory properties; Shelf life

5.2 Introduction

In South Africa, chicken eggs are generally stored at room temperature (Theron, Venter & Lues, 2003). In the United States eggs are refrigerated as a rule while in South Africa this is not practical. From the time of egg laying to consumption, several chemical, physical and biological changes occur in the egg (Stadelman & Cotterill, 1995). These changes depend on the conditions of storage of the eggs, which include time, temperature and relative humidity of the storage environment (Berardinelli *et al*., 2003). Storage temperatures can also have an influence on the functional properties of the albumin (Hammershøj, Prins & Qvist, 1999). During the storage of shell eggs, the pH of albumin increases from about 7.6 to 9.7 (Vaclavick, 1999). The rise in albumin pH is caused by a loss of CO_2 through the shell and is responsible for the changes involving the albumin (Burley & Vadehra, 1989; Stadelman & Cotterill, 1995). High storage temperatures accelerate CO_2 loss during storage (Linden & Lorient, 1999). The changes in albumin might be due to ovomucin-lysozyme interaction or modification of the structure of ovomucin (Berardinelli *et al.,* 2003). Application of coatings on the shells of eggs reduces weight loss, and may prolong the shelf life by minimizing the loss of CO_2 and thereby maintaining internal quality such as air cell height, Haugh units, yolk index and albumin pH (Imai, 1981). Passmore reported that mineral oil blocks the shell pores and reduces the decline of egg quality (Lepule, 2003). But these researchers did not investigate the effect of storage conditions (e.g. temperature of storage or storage time) on the internal egg quality . In addition, shell coating also prevents the entry of spoilage or pathogenic microorganisms through the egg shell pores. The objective of this study was to determine the effect of oil coating on the functional properties and shelf life of WSE stored at three different storage conditions.

5.3 Materials and methods

5.3.1 Eggs

Two batches of freshly collected whole shell eggs (480 eggs per batch) were randomly selected on two different days at a commercial egg packing plant in Gauteng province, South Africa. The eggs were cleaned, graded (Large, 55-65g), and the one half was individually coated with WOP 30 food grade mineral oil (LeoChem Company; Durban, South Africa) while the other half remained uncoated. All eggs

were packed in commercial fibre trays holding 30 eggs per tray. Shell eggs were stored under three different storage conditions (Table 11) for a period of six weeks.

Table 11. Storage conditions of coated and uncoated whole shell eggs for six weeks

Description of storage condition	Day temperature	Night temperature	Relative humidity (%)
16ºC; 58 % RH	16 ºC	16 ºC	58 %
25ºC & 15ºC; 55 % RH	25 ºC	15 ºC	55 %
32ºC; 32 % RH	32 ºC	32 ºC	32 %

5.3.2 Haugh units

The Haugh unit values of six randomly selected eggs per treatment were determined once a week throughout storage. The whole shell eggs were individually broken on a glass plate and the height (mm) of the albumin was measured at six positions using a micrometer (Technical Services and Supplies, England). The Haugh unit values were calculated using the following formula (Monira *et al*., 2003),

Haugh Unit =100 log ($H-1.7W^{0.37}+7.6$)

H = Average height of albumin (mm); W = Mass of whole shell egg (g)

5.3.3 Changes in egg weight over storage time

The mass (g) of six randomly selected whole shell eggs per treatment was individually measured every week to determine weight loss over time.

5.3.4 pH and foaming properties

The pH of albumin and yolk as well as albumin foaming properties of eggs from each treatment were measured weekly in duplicate. Per replicate, three shell eggs from each treatment were individually broken using a knife. A plastic egg separator placed on top of a 600 ml glass beaker was used to separate the yolk from the albumin. The yolk pools of the three eggs were collected in a stomacher bag and then homogenised for 30s using a stomacher lab blender. The liquid albumin was transferred to a stomacher bag, homogenised for 30s and then returned to the beaker. After homogenisation, the height (mm) in a 600 ml beaker was measured. The pH and temperature of the homogenised albumin and yolk respectively were measured, using a Hanna pH 211 Microprocessor. A Braun Multiquick ® professional MR 55550 M

CA mixer was immersed in the albumin mixture in the beaker and whipped at speed 1 for 3 min. The temperature of albumin ranged between 18.7ºC and 24.8 ºC throughout the trial due to fluctuation in day time temperature. The height of foam was measured as well as the drained liquid height after 60min. Drained liquid height is the height of liquid drained from the foam after a specified period of time (e.g. 60 min).The following formulas were used to calculate the foaming capacity and foam stability (adapted from Chang & Chen, 2000):

Foam capacity (%) = [Foam height (mm)] / [Unwhipped albumin height (mm)] X 100

Foam stability (%) = {[Foam height (mm) - Drained liquid height]*(mm)/ Foam height (mm)} X 100

*Drained liquid height (mm) = Height of liquid phase after 60min (mm)-Height of liquid phase after 30s (mm)

5.3.5 Descriptive sensory evaluation

Sensory evaluation of the eggs, using a trained panel of 10 members was carried out in a sensory evaluation laboratory with individual booths equipped with a direct data collection facility (Compusense ® Five release 4.6, Compusense Inc. Guelph, Canada) at the Department of Food Science (U). Panellists were recruited, screened, selected and trained using generic descriptive analysis (Einstein, 1991). It was the same panel previously trained as described in Chapter 4 (section 4.3.4).The sensory descriptors developed for broken out raw eggs are shown in Table 12.

Table 12. Sensory attributes and rating scale anchors used by the trained sensory panel to describe broken out raw eggs

Sensory attributes of eggs	Scale anchors	Scale value for reference sample [1]
Size of yolk	1=Small; 9=Large	1
Height of yolk	1=Low; 9=High	9
Roundness of yolk	1=Flat; 9=Round	9
Height of albumin	1=Low; 9=High	9
Brightness of yolk colour	1= Dull yellow; 9=Bright yellow	9
Position of yolk	1=Not centered; 9=Centered	9
Firmness of albumin	1=Not firm and compact; 9=Very firm and compact	9
Prominence of chalazae	1=Not prominent; 9=Very prominent	9
Yolk colour [2]	1=Light yellow; 15=Orange	5

[1] Reference sample was a two day old fresh egg

[2] Roche egg colour fan (Roche-Switzerland) with 15 colour references (15-point scale).

Evaluation was done once a week during two 1h sessions. During each session, each panellist was presented with six coded eggs representing the treatments to compare with a reference sample (a two day old fresh egg). The egg contents were served in (70/120 Atlantic Black) Styrofoam square plates (with a size of 130 x 180 mm) coded with randomly selected three digit codes. The serving sequence was randomised over the panel to minimize bias due to order effects. Panellists were instructed to rate each egg according to the nine sensory descriptors (Table 12). Note that the sensory panel evaluated the eggs that were stored for six weeks during the following week (i.e the 7th week). The panel consisting of university students was not available during the 6th week due to a university holiday.

5.3.6 Statistical analysis of data

For each of the storage conditions, a separate analysis of variance (ANOVA) was performed to analyse the main and interaction effects of oil coating and storage time on the egg quality parameters at 95% significance level. All analyses were performed using Statistica (StatSoft Inc., version 7.1 Tulsa, USA). If significant main or interaction effects were found, Fisher's Least Significant Difference (LSD) test was used to detect Gsignificant differences between means ($p<0.05$).

5.4 Results

5.4.1 Haugh units

Significant interaction effects for oil coating and storage time were found for eggs stored at 16ºC;58% RH and 25ºC & 15ºC;55% RH (Figs 7 and 8). Haugh units for coated eggs were significantly higher and decreased at a significantly slower rate compared to uncoated eggs. The Haugh units of coated eggs stored at 16°C; 58% (Fig 7) were generally greater than Haugh units for uncoated eggs stored for a period of six weeks. At storage 25°C & 15°C; 55% RH (Fig 8), Haugh units for coated eggs was 80 during the first week and reduced to 70 at week six; whereas the Haugh units for the uncoated eggs during week 1 was 56 and at week 6 was less than 30. The Haugh units of the uncoated eggs decreased more rapidly than coated eggs (Fig 8).

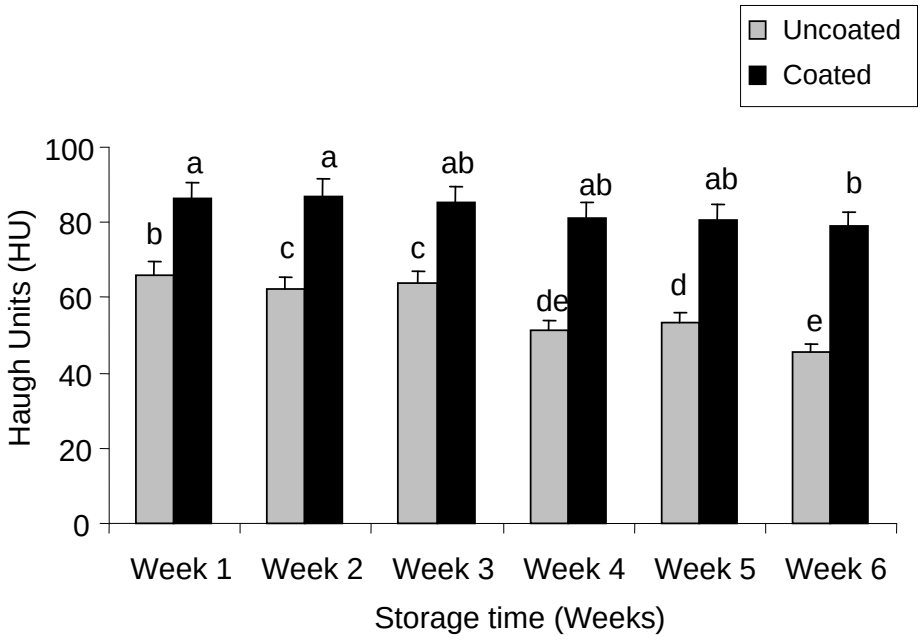

Figure 7. Effect of oil coating and storage time on the Haugh units of whole shell eggs stored at 16ºC; 58% RH

[abcde] Values with different letters differed significantly ($p<0.05$)

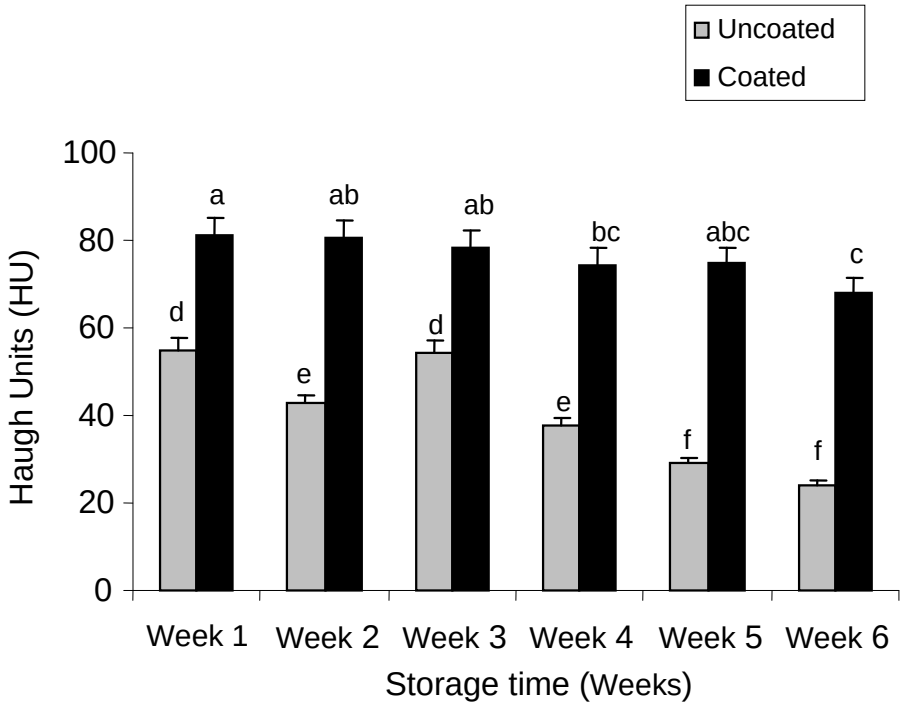

Figure 8. Effect of oil coating and storage time on the Haugh units of whole shell eggs stored at 25ºC & 15ºC;55% RH

[abcdef] Values with different letters differed significantly ($p<0.05$).

Oil coating had a significant effect on Haugh units when eggs were stored at 32ºC; 32% RH. Haugh units of coated eggs were significantly higher (68.9 ± 8.93) than the Haugh units of uncoated eggs (30.1 ± 13.2). Haugh units of eggs stored at 32ºC; 32% RH decreased over time but a significant decrease was clearly seen as from weeks 1, 4 and 6 (Table 13).

Table 13. Effect of storage time on the Haugh units of coated and uncoated whole shell eggs stored at 32°C;32% RH

Storage time (Weeks)	Haugh units (HU) ±Standard deviations (SD)
Week 1	56.7 (± 18.6)[a]
Week 2	54.2 (± 22.9)[ab]
Week 3	49.7 (± 22.2)[bc]
Week 4	48.8 (± 23.2)[c]
Week 5	47.6 (± 21.1)[bc]
Week 6	40.4 (± 25.3)[d]

[abcd] Means with different superscripts are significantly different ($p<0.05$)

5.4.2 Changes in egg weight over storage time

Significant interaction effects for oil coating and storage time were found for the weight loss of eggs stored at all three storage conditions (Table 14). Overall, the weight of coated eggs was mostly higher than the weight of uncoated stored eggs from week 1 to week 6.

5.4.3 pH of albumin and yolk

There were no significant difference for the treatment x storage time interaction effects for pH of albumin, yolk pH and foam stability at all storage conditions. There were significant pH differences ($p<0.05$) for both the yolk and albumin between coated and uncoated eggs (Table 15). The yolk and albumin pH values of coated eggs were significantly lower ($p<0.05$) compared to uncoated eggs when stored at all three storage conditions. Storage time had an effect ($p<0.05$) on the pH of yolk and albumin. After six weeks, the yolk pH of eggs stored at all three storage conditions was significantly higher ($p<0.05$) compared to yolk pH after 1 week and 3 weeks (Table 15). Storage time had a significant influence on the albumin pH of eggs stored at 32ºC; 32% RH (Table 15). At this storage condition, the pH of albumin increased from 9.0 to 9.5 over the storage time.

Table 14. The interaction effect of oil coating and storage time on the average weight (g) ± standard deviation (SD) of whole shell eggs stored at three storage conditions.

Storage time (Weeks)	Average egg mass (g)					
	16ºC; 58% RH		25ºC & 15ºC; 55% RH		32ºC; 32% RH	
	Coated	Uncoated	Coated	Uncoated	Coated	Uncoated
1	65.2 (±11.8)^{d}	61.3(±4.3)^{acd}	61.1 (±3.6)^{c}	59.2(±2.5)^{abc}	61.4 (±3.4)^{a}	60.0 (±4.0)^{ab}
2	60.5 (±3.0)^{ac}	59.3 (±5.0)^{ab}	60.8 (±2.2)^{bc}	57.1(±2.6)^{ad}	62.3 (±3.2)^{a}	57.1 (±4.5)^{b}
3	64.4 (±5.7)^{cd}	57.4 (±2.6)^{ab}	58.7 (±2.4)^{ab}	54.9 (±1.4)^{d}	60.9 (±4.6)^{a}	55.6 (±2.5)^{de}
4	57.1 (±5.0)^{ab}	58.6 (±0.9)^{ab}	59.0(±2.9)^{abc}	58.9(±3.1)^{abc}	59.4 (±1.1)^{ab}	53.2 (±5.8)^{d}
5	57.7(±3.9)^{ab}	59.7(±4.7)^{a}	59.5(±3.5)^{abc}	57.5(±3.8)^{a}	59.4 (±4.1)^{ab}	49.3 (±3.9)^{c}
6	60.1(±4.8)^{a}	55.1(±2.8)^{b}	60.7(±3.7)^{bc}	54.8(±3.8)^{d}	60.1(±3.3)^{ab}	47.4 (±4.1)^{c}

^{abcde} Means in a row for treatment and /or storage time with different superscripts are significantly different ($p<0.05$)

Table 15. Mean pH values ± standard deviation (SD) for albumin and yolk of coated and uncoated whole shell eggs stored at three storage conditions for six weeks

pH								
Storage conditions		Treatment			Storage time			
	Egg portion	Coated [1]	Uncoated [1]	p-value	Week 1[2]	Week 3[2]	Week 6[2]	p-value
16ºC; 58 % RH	Albumin	8.3 (± 0.2)[a]	9.4 (± 0.1)[b]	<0.01	8.9 (±0.5) [a]	8.8 (±0.7) [a]	8.8 (±0.6) [a]	0.32
	Yolk	6.2 (± 0.1)[a]	6.3 (± 0.1)[b]	0.02	6.1 (± 0.1)[a]	6.2 (±0.1)[a]	6.4 (±0.1)[b]	<0.01
25ºC & 15ºC; 55 % RH	Albumin	8.4 (± 0.2)[a]	9.5 (± 0.1)[b]	<0.01	8.9 (±0.5) [a]	8.9 (±0.6) [a]	9.0 (±0.6) [a]	0.27
	Yolk	6.3 (± 0.2)[a]	6.4 (± 0.2)[b]	0.03	6.2 (±0.1)[a]	6.3 (±0.1)[a]	6.5 (±0.2)[b]	<0.01
32ºC; 32 % RH	Albumin	8.6 (± 0.2)[a]	9.8 (± 0.5)[b]	<0.01	9.0 (±0.6)[a]	9.0 (±0.6)[a]	9.5 (±1.0)[b]	0.01
	Yolk	6.3 (± 0.2)[a]	6.6 (± 0.3)[b]	<0.01	6.3 (±0.1)[a]	6.4 (±0.1)[a]	6.8 (±0.3)[b]	<0.01

[ab] When comparing treatments or storage times, means with different superscripts are statistically different ($p<0.05$)

[1] Pooled data for measurements taken at 1, 3 and 6 weeks of storage

[2] Pooled data for coated and uncoated eggs

5.4.4 Albumin foaming capacity

The albumin foaming capacity of coated eggs stored at 16ºC; 58% RH for six weeks (273.7% ± 31.4) was significantly lower ($p<0.05$) compared to uncoated eggs (336.9% ± 28.9) stored in the same manner. There was a significant oil coating and storage time interaction effect for the foaming capacity of eggs stored at 25ºC & 15ºC; 55% RH (Fig 9). There was a significant difference ($p=0.01$) in the foaming capacity of albumin for coated (308.8% ±37.5) and uncoated (350.1% ±37.6) eggs stored at 32ºC; 32 % RH. Foaming capacity of uncoated eggs was high (363.4 % ± 23.2) during week 1 but decreased during week 3 (343.2% ± 9.8), and eventually increased after week 6 of storage (396.4% ± 43.1). For coated eggs, there was a linear decrease in the foaming capacity over time. Coated eggs had lower foaming capacity values compared to uncoated eggs.

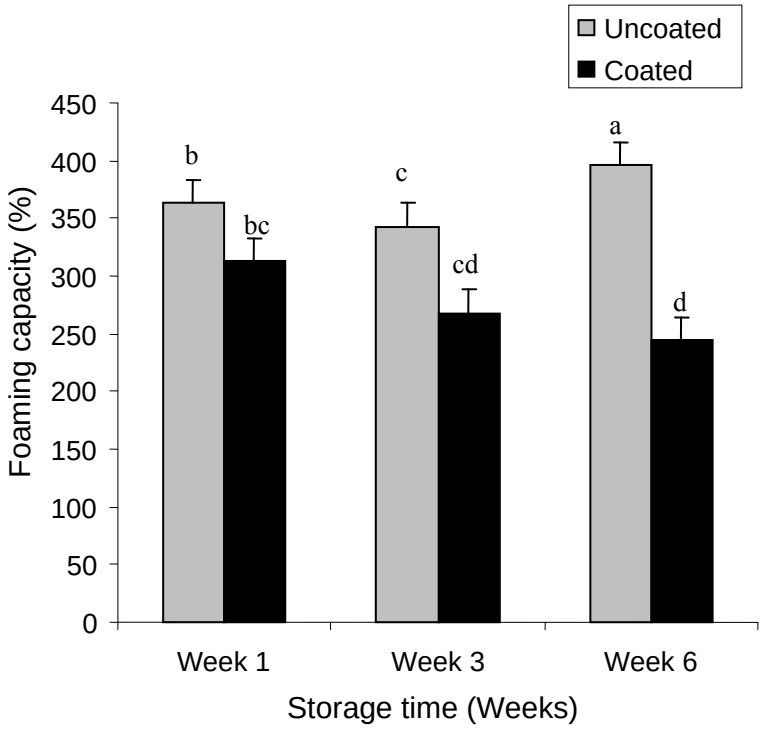

Figure 9. Effect of oil coating and storage time on the foaming capacity of the albumin stored at 25ºC & 15ºC;55% RH

[abcd]Values with different letters differed significantly ($p<0.05$).

5.4.5 Foam stability

The foam stability of coated and uncoated eggs at all storage conditions (p=0.58 at 16ºC; 58% RH, p=0.28 at 25ºC & 15ºC; 55% RH and p=0.81 at 32ºC; 32% RH) was not significantly different. There was no significant difference (p>0.05) in the albumin foam stability of eggs stored both at 16ºC;58% RH and 32ºC; 32% RH for six weeks, while foam stability of albumin from eggs stored at 25ºC & 15ºC; 55% RH was significantly lower (p=0.01) during week 3 compared to weeks 1 and 6 (Table 16).

Table 16. Effect of storage time on the foam stability of egg albumin stored at 25ºC & 15ºC; 55% RH for six weeks.

Storage time (Weeks)	Foam stability (%)
Week 1	337.8 (± 35.7) [a]
Week 3	305.6 (± 43.0) [b]
Week 6	320.7 (± 90.1) [a]

5.4.6 Visual sensory properties of broken out eggs

Significant differences (p<0.05) were found between the sensory properties of coated and uncoated eggs that were stored at 16ºC; 58% RH for seven weeks as assessed by a trained sensory panel (Table 17). The size of yolk of coated eggs stored at 16ºC; 58% RH was significantly smaller compared to uncoated eggs. Coated eggs had a significantly higher upright (heaped) yolk than uncoated eggs. Coated eggs also had rounder shaped yolks while uncoated eggs were flat and irregular in shape. The albumin height of coated eggs was higher than uncoated eggs. At the same storage conditions, the albumin of coated eggs was more firm and compact than uncoated eggs, which in turn were thin and not firm and compact. Chalazae was more prominent (clearly visible) in the coated eggs. There were no significant differences (p>0.05) between coated and uncoated eggs with regard to brightness of yolk, position of yolk and yolk colour intensity.

At storage 16ºC; 58% RH, storage time affected ($p<0.05$) the height and firmness of albumin. There was a decrease in albumin height and firmness after week 2 (Table 17). A significant difference in the size of yolk ($p<0.05$) was found between coated and uncoated eggs stored for six weeks at 25ºC & 15ºC; 55% RH (Table 17). Uncoated eggs had yolks that appeared bigger than that of coated eggs because they were flat. Coated eggs had a significantly higher ($p<0.01$) yolk height compared to uncoated eggs. Uncoated eggs had flattened yolks ($p<0.01$) that were irregular in shape while yolks of coated eggs were heaped, regular and round. Uncoated eggs also had a significantly lower ($p<0.01$) albumin height than coated eggs. No significant differences ($p>0.05$) were found for the brightness of yolk colour and position of yolk between coated and uncoated eggs. Uncoated eggs were less firm ($p<0.01$) and compact compared to coated eggs. The chalazae in coated eggs was more prominent ($p<0.01$). The colour of yolk for coated eggs was lighter ($p<0.05$) than the yolk of uncoated eggs. At this storage condition (25ºC & 15ºC; 55% RH), the size of yolk increased after 2 weeks, while the height of the yolk and height of albumin decreased during storage. No significant differences ($p>0.05$) were found over the storage time for position of yolk, brightness of yolk colour, firmness of albumin, prominence of chalazae and yolk colour.

There was a significant interaction effect of oil coating and storage time ($p<0.05$) for some visual sensory properties of broken out eggs stored at 32ºC; 32% RH (Table 18). Significant differences ($p<0.05$) were found for size of yolk, height of yolk, roundness of yolk, brightness of yolk colour, and yolk colour. The yolk size of uncoated eggs was larger than that of coated eggs at week 2 and week 5. Uncoated eggs had a significantly lower yolk height compared to coated eggs. The yolks of coated eggs were round in shape whereas yolks of uncoated eggs were flattened. During the last week of storage (Week 7), the yolk of coated eggs was more orange compared to the lighter colour of uncoated eggs. Although there was no significant interaction effects with regard to the height of albumin, position of yolk, firmness of albumin and prominence of chalazae for the broken out raw eggs stored at 32ºC; 32% RH, significant differences were found between coated and uncoated shell eggs. At this storage condition, firmness of albumin ($p=0.02$) and prominence of chalazae ($p=0.001$) were affected by storage time.

Table 17. Mean ratings (± standard deviations) for the descriptive visual sensory properties [1] of broken out eggs stored at different storage conditions

Storage conditions	Sensory properties of eggs [1]	Treatment			Storage time			
		Coated[2]	Uncoated[2]	p-value	Week 2[3]	Week 5[3]	Week 7[3]	p-value
16ºC;58 % RH	Size of yolk	4.5 (±1.3)[a]	5.0 (±1.3)[b]	0.02	4.7 (±1.1)[a]	4.6 (±1.5)[a]	4.9(±1.4)[a]	0.58
	Height of yolk	7.2 (±1.3)[b]	6.0 (±1.4)[a]	<0.01	6.6 (±1.4)[a]	6.8 (±1.6)[a]	6.3(±1.5)[a]	0.28
	Roundness of yolk	7.6 (±1.1)[b]	6.5 (±1.6)[a]	<0.01	7.3 (±1.3)[a]	7.1 (±1.4)[a]	6.6(±1.7)[a]	0.10
	Height of albumin	6.5 (±1.8)[b]	4.0 (±1.6)[a]	<0.01	6.0 (±1.8)[b]	5.1 (±2.0)[a]	4.7 (±2.2)[a]	<0.01
	Brightness of yolk colour	5.9 (±1.4)[a]	5.5 (±1.5)[a]	0.16	5.3 (±1.6)[a]	5.7 (±1.3)[a]	6.1(±1.4)[a]	0.05
	Position of yolk	4.0 (±2.7)[a]	4.4 (±2.7)[a]	0.39	4.4 (±2.6)[a]	4.3 (±2.5)[a]	3.8(±2.9)[a]	0.64
	Firmness of albumin	6.7 (±1.6)[b]	3.6 (±1.9)[a]	<0.01	5.8 (±2.0)[b]	4.9 (±2.5)[a]	4.5 (±2.3)[a]	<0.01
	Prominence of chalazae	7.4 (±2.0)[b]	5.2 (±2.5)[a]	<0.01	6.3 (±2.6)[a]	6.5 (±2.4)[a]	6.1(±2.7)[a]	0.84
	Yolk colour	5.9 (±1.4)[a]	5.5 (±1.4)[a]	0.19	5.7 (±1.7)[a]	5.7 (±1.3)[a]	5.7(±1.3)[a]	0.97
25ºC & 15ºC;	Size of yolk	4.7 (±1.4)[a]	6.5 (±1.4)[b]	<0.01	5.0 (±1.5)[b]	5.6 (±1.7)[a]	6.2 (±1.6)[a]	<0.01
55 % RH	Height of yolk	6.7 (±1.4)[b]	4.6 (±1.5)[a]	<0.01	6.2 (±1.7)[b]	5.6 (±1.8)[a]	5.1 (±1.8)[a]	<0.01
	Roundness of yolk	6.9 (±1.3)[b]	4.8 (±2.2)[a]	<0.01	6.8 (±1.8)[b]	5.6 (±2.1)[a]	5.1 (±2.1)[a]	<0.01
	Height of albumin	5.8 (±1.6)[b]	3.5 (±1.8)[a]	<0.01	5.2 (±1.9)[b]	4.4 (±1.8)[a]	4.3 (±2.3)[a]	0.03
	Brightness of yolk colour	5.8 (±1.3)[a]	5.6 (±1.7)[a]	0.50	5.6 (±1.3)[a]	5.6 (±1.7)[a]	5.8(±1.5)[a]	0.92
	Position of yolk	4.2 (±2.7)[a]	3.8 (±2.6)[a]	0.43	4.7 (±2.8)[a]	3.6 (±2.5)[a]	3.6(±2.6)[a]	0.15
	Firmness of albumin	6.0 (±1.7)[b]	2.3 (±1.6)[a]	<0.01	4.6 (±2.5)[a]	3.8 (±2.5)[a]	4.1(±2.5)[a]	0.13
	Prominence of chalazae	6.8 (±2.1)[b]	4.5 (±2.4)[a]	<0.01	5.8 (±2.4)[a]	5.7 (±2.7)[a]	5.4(±2.6)[a]	0.77
	Yolk colour	6.1 (±1.6)[b]	5.5 (±1.3)[a]	0.02	5.7 (±1.6)[a]	5.6 (±1.4)[a]	6.1(±1.3)[a]	0.29

[ab] Means in a row for treatment and /or storage time with different superscripts are significantly different ($p<0.05$)

[1] Refer to Table 2 for details of the rating scales.

[2] Pooled data for measurements taken at 2, 5 and 7 weeks of storage

[3] Pooled data for coated and uncoated eggs

Table 18. Mean ratings (± standard deviations) for the descriptive sensory properties[1] of raw whole shell eggs stored at 32ºC (32% RH) for a period of seven weeks

	Week 2		Week 5		Week 7		
Sensory properties[1]	Coated	Uncoated	Coated	Uncoated	Coated	Uncoated	p-value
Size of yolk	5.0 (± 1.6)^{a}	7.2 (± 1.2)^{bc}	5.3 (± 1.6)^{a}	8.2 (± 0.9)^{c}	5.8 (± 1.8)^{a}	6.2 (±3.5)^{ab}	0.01
Height of yolk	6.3 (± 1.3)^{a}	3.2 (± 1.3)^{c}	6.6 (± 1.2)^{a}	1.9 (± 1.3)^{d}	5.4 (± 1.4)^{b}	1.1 (± 0.2)^{e}	0.01
Roundness of yolk	6.8 (± 1.9)^{a}	4.4 (± 2.8)^{b}	6.8 (± 1.0)^{a}	2.9 (± 2.6)^{d}	5.6 (± 1.6)^{ab}	1.0 (± 0.0)^{c}	0.04
Height of albumin	5.9 (± 1.7) ^{a}	3.1 (± 2.3) ^{a}	5.9 (± 1.6) ^{a}	2.4 (± 1.5) ^{a}	4.9 (± 1.8) ^{a}	2.1 (± 2.5) ^{a}	0.64
Brightness of yolk colour	5.7 (± 1.2)^{a}	5.7 (± 1.6)^{a}	6.1 (±1.3)^{a}	5.6 (±1.9)^{a}	6.5 (± 1.4)^{a}	3.6 (± 2.3)^{b}	<0.01
Position of the yolk	4.0 (± 2.5) ^{a}	3.0 (± 2.3) ^{a}	3.1 (± 2.3) ^{a}	2.6 (± 1.8) ^{a}	3.7 (± 2.5) ^{a}	1.7 (± 1.7) ^{a}	0.38
Firmness of albumin	5.7 (± 1.9) ^{a}	2.1 (± 1.7) ^{a}	5.9 (± 1.8) ^{a}	1.6 (± 1.2) ^{a}	4.8 (± 1.4) ^{a}	1.1 (± 0.3) ^{a}	0.53
Prominence of chalazae	6.8 (± 2.4) ^{a}	4.2 (± 2.4) ^{a}	6.6 (± 2.2) ^{a}	3.4 (± 2.5) ^{a}	5.5 (± 2.4) ^{a}	3.2 (± 1.0) ^{a}	0.45
Yolk colour	5.7 (± 1.4)^{ab}	5.7 (± 1.5)^{ab}	6.2 (± 1.3)^{a}	6.2 (± 1.6)^{a}	6.5 (± 0.9)^{a}	4.7 (± 0.7)^{b}	0.03

[abcd] Values in the same row with different superscripts are significantly different ($p \leq 0.05$)

[1] Refer to Table 2 for details of the scales.

5.5 Discussion

Oil coating reduced the foaming capacity of albumin in all treatments. In addition, the foaming capacity of the coated eggs decreased over time. The mineral oil probably penetrated through the pores and contaminated the egg contents particularly the albumin. Significant amounts of lipids adversely affect the foaming properties of albumin proteins. Damodaran (1996) explained that lipids are more surface active than proteins; they adsorb rapidly at the air-water interface and inhibit adsorption of proteins during foam formation. Since lipid films do not possess the cohesive and viscoelastic properties necessary to withstand the internal pressure of the foam bubbles, the bubbles rapidly expand, then collapse during whipping (Damodaran, 1996). Therefore, the ability of albumin proteins to produce foams is minimised by the presence of oil. In order to produce quality foams, the surface tension needs to be lowered at the interface. Uncoated eggs foamed better than coated eggs throughout storage. A dissociation of the ovomucin–lysozyme complex occurs when shell eggs are stored (Hayakawa, Kondo, Nakamura & Sato, 1983). As a result, the content of free, soluble ovomucin in both firm and thin albumin may rise, which could make the protein more available for adsorption at the film surface of the foam and for lowering the surface tension, thereby stabilising the foam of both firm and thin albumin against liquid drainage (Hammershøj *et al*., 1999). The availability of soluble ovomucin probably increases over time. Hammershøj (2000) observed that disulfide bond reduction agents or mechanical homogenisation of egg albumin contributed to an increased foaming capacity. By rupture of disulfide bonds the flexibility of the proteins increase to allow for unfolding at the film surface. This leads to an increase in surface hydrophobicity and enhanced foam formation.

Egg quality deterioration is a factor of storage time, temperature, humidity and handling during storage. Over time, egg quality deteriorates and the rate of deterioration is increased with higher storage temperatures (Stadelman & Cotterill, 1995). Factors including storage temperature, atmospheric or relative humidity, concentration of CO_2 and genetic factors also influence the internal components of the egg. These factors affect egg components such as the yolk, albumin, vitelline membrane, and shell (Burley & Vadehra, 1989). An increase in albumin pH is due to a loss of CO_2 through the open pores in the shell (Oosterwoud, 1987; Burley & Vadehra, 1989). Loss of water from the albumin also causes a gradual increase in the

volume of the air space (air cell size) in the egg (Burley & Vadehra, 1989; Stadelman & Cotterill, 1995; Vaclavick, 1999; Jacob, Miles & Mather, 2000; Messens *et al*., 2004; Laghi *et al.,* 2005). The quantity of firm albumin decreases while there is an increase in thin albumin concentration during storage. Changes in pH of albumin influence the functional properties (Penfield & Campbell, 1990; Linden & Lorient, 1999). Oil coating preserved the quality and some functional properties (e.g. Haugh units, pH and sensory properties) of eggs stored for seven weeks. Oil coating closes the pores on the eggshell surface that allow moisture and CO_2 to evaporate from the egg. As a result, when these pores are sealed it is not easy to lose moisture and CO_2 rapidly. Therefore, oil coating slows down the rate of evaporation of water and escape of CO_2 (Wong, Herald, Hachmeister, 1996) as well as biological and physical changes. Due to this phenomenon, coated eggs had lower pH, high Haugh unit values, strong vitelline membrane, and heaped small size yolks, high, firm albumin, and less changes in the visual appearance of eggs. Uncoated whole shell eggs were stored at 16ºC; 58% RH for three weeks without noticeable changes in their visual sensory properties. Due to the low temperature, the conversion of thick albumin into thin albumin was very slow. Low temperature also reduces the loss of CO_2 from the albumin and minimises the flow of water from the albumin to the yolk. It is the reason why yolks would appear larger in older eggs. Therefore, quality of eggs will deteriorate at a slower rate compared to eggs stored at higher temperatures.

The quality of yolk is described by its appearance (colour, shape and size), firmness and smell. Freshly laid eggs have round, heaped yolks and high yolk heights. During storage, the yolk size of uncoated eggs enlarged and flattened due to diffusion of water from the albumin to the yolk through the vitelline membrane (Lucisano, Hidalgo, Comelli & Rossi, 1996; Jacob *et al*., 2000; Silversides & Scott, 2001; Bhale, No, Prinyawiwatkul, Farr, Nadarajah & Meyers, 2003). As a result the yolk becomes weak and flat in shape (Charley & Weaver, 1998). At the same time, the strength of the vitelline membrane decreases (Gaman & Sherrington, 1981; Oosterwoud, 1987; Kralik, Komendonović. & Petričević, 1990), because the yolk is surrounded by the vitelline membrane of which the permeability increases with time allowing water to diffuse from the albumin into the yolk during the first few days of storage (Laghi *et al.,* 2005). The vitelline membrane weakens, stretches and loses fibrous (network) material from the outer surface and ruptures more readily when the egg is opened

(Burley & Vadehra, 1989; Kirunda & McKee, 2000). Kido *et al.*, according to Kirunda & McKee (2000) found that degradation of one of the major structural glycoproteins, glycoprotein II, in the vitelline membrane and the breakage of disulfide bonds of the ovomucin cause a loss in vitelline membrane integrity over time.

The yolk colour of both coated and uncoated eggs stored at 16°C; 58% RH and at 25°C &15°C; 55% RH was no longer light yellow in colour. After 5 weeks of storage, the yolk colour of the eggs that were stored at 32°C; 32 % RH became dark yellow. In a study done by Bhale *et al*. (2003) on chitosan coated eggs, it was found that the yolk colour became darker, less reddish and more yellowish, shifting the hue more toward yellow on eggs that were stored for 5 weeks at 25ºC. The yolk colour preference differs with geographical location and customer but a fairly light colour is generally preferred (Bhale *et al*., 2003). Colour depends on the yolk carotenoids (e.g. lutein, zeaxanthin, and β-cryptoxanthin content). During storage, carotenoids can be degraded by oxidative processes, resulting in a change in yolk pigmentation (Caner, 2005). Uncoated eggs had flatter yolks and less round yolks compared to coated eggs. A weakened vitelline membrane is responsible for the yolk flattening and a decrease in the yolk height with an increase in the yolk size. The phenomenon occurs faster at high storage temperature (Jacob *et al*., 2000). Uncoated eggs had lower albumin heights compared to coated eggs. The albumin of uncoated eggs was not firm, it was thin, fluid or watery especially those eggs that were stored at 32°C; 32% RH. For such eggs, the albumin was spreading considerably over the plate and occupied a large area. Watery albumin is clear and this is an indication of ageing albumin. In addition, the albumin may eventually become yellow during storage. Fresh eggs are characterised by cloudy white albumin due to dissolved CO_2 (Oosterwoud, 1987). During storage, the middle thick albumin layer disintegrated and the proportion of the external liquid layer increased. It was emphasized that in egg albumin, under influence of triptical proteinase, proteolysis occurs, and eventually the mucinic threads split and release fixed water (Kralik, according to Kralik *et al.,* 1990). The chalazae of uncoated eggs also were less prominent. Height of albumin, firmness of albumin, brightness of yolk colour, roundness of yolk and height of yolk decreased during storage time. The rate of decrease in height of yolk and roundness of yolk of uncoated eggs was temperature dependent due to accelerated CO_2 loss at higher

temperatures. Hence, coating the eggshell with oil could delay the deterioration of egg internal quality.

A decline in Haugh units is associated with a reduction in egg quality. The higher the Haugh unit values, the better the albumin quality of the eggs (Stadelman, 1986a). Overall, there was a decrease over time in Haugh unit values, both in coated and uncoated shell eggs at all storage conditions. A decrease in Haugh units was induced by egg albumin thinning due to aging (Stadelman & Cotterill, 1995). Thinning, as stated earlier, was due to a destruction of the lysozyme-ovomucin complex when loss of CO_2 resulted in a higher pH. Furthermore, Heath (1977) found that sulfhydryl content accumulates with an increase in pH and suggested that this increase in sulfhydryl content and the associated albumin thinning were the result of the uncoiling of albumin proteins. Hence, the temperature and storage conditions play an essential part in the albumin thinning process. The decrease in Haugh units is followed by an increase in degree of oldness and content of inorganic phosphorus in albumin (Kralik *et al.*, 1990). The α-ovomucin-lysozyme complex covered with β-ovomucin in thick albumin can form a gel structure (Hayakawa *et al.*, 1983). Kato *et al* (according to Hayakawa *et al*., 1983) suggested that α-ovomucin in the ovomucin gel does not solubilise during long-term storage because it is polymerised by disulfide bonds. According to Hayakawa *et al*., (1983) disulphide bonds in lysozyme may covalently link the α-ovomucin, and this complex is protected by coverage of β-ovomucin. The β-ovomucin was reported to dissociate gradually from the ovomucin complex and to solubilise into the liquid fraction of thick albumin, while α-ovomucin remains insoluble during the natural thinning of albumin by ovomucin-lysozyme interaction. Hammershøj & Qvist (2001) suggested that during the thinning of egg albumin, the structure of the firm albumin fraction is broken down, a part of ovomucin, which is bound in a complex is released. Haugh units of coated eggs were higher compared to uncoated eggs stored at 16°C; 58% RH and 25°C & 15°C; 55% RH. Coated shell eggs stored at 16°C; 58% RH for six weeks looked almost similar to fresh eggs stored for two days. Data from this study indicated that Haugh units of uncoated eggs decreased much faster over time. There are several thousand small pores in the surface of the egg shell and the positive pressure of the egg content, at higher temperature, opens these pores and allows more CO_2 and water to escape rapidly (Hisil & Ötles, 1997). Hence, coating makes the shell less permeable to water vapour and CO_2 and so delays the deterioration in quality of eggs during storage. In

this study, the Haugh unit values decreased over time in all storage conditions but differed in the rate of decrease. Alleoni & Antunes (2001) also found that the Haugh unit scores decreased with storage time at room temperature (25ºC).

The weight of coated shell eggs was decreasing at a slower rate compared to uncoated eggs. The rate of weight loss was temperature dependent. Shell eggs lose weight due to water evaporation and loss of CO_2 from the albumin through the pores during storage (Stadelman, 1986b; Caner, 2005). The rate of evaporation increases with the difference in relative humidity inside and outside of the egg. As long as the relative humidity in the atmosphere around the shell eggs is less than 99.6%, water escapes through the pores on the shell surface (Jacob *et al.*, 2000).

In this study, the Haugh unit values of coated eggs stored at or below 25°C were always higher than 72. This value is considered as the minimum value for very fresh and high quality eggs. In the United States egg grading system, a Haugh unit of 72 or higher is associated with Grade AA quality, which is regarded as First grade (United States Department of Agriculture, 2000). Second grade (Grade A) eggs have Haugh units of between 60 and 72. Grade B eggs have Haugh unit values of less than 60. Hence, coated eggs that were stored at 16°C; 58% RH, still looked like fresh eggs with a Haugh unit value of 72 after 6 weeks of storage. For coated eggs that were stored at room temperature (25°C & 15°C; 55% RH), the Haugh units were 70 during week 6 (USA, Grade A). At these storage conditions, uncoated eggs had Haugh units of less than 60 during week 1, which means the eggs had already deteriorated to Grade B. The Haugh unit value of coated and uncoated eggs stored at 32°C; 32% RH were 68 and 30 respectively.

Albumin pH was lower for all coated eggs than uncoated eggs. Uncoated eggs lose CO_2 and moisture in the albumin through the pores more rapidly than coated eggs. The pH of albumin increased from 9.0 to 9.5 for eggs stored at 32°C; 32% RH for six weeks. The pH of albumin is an important parameter in quality retention. Initially, the egg contains about 30 ml of dissolved CO_2 appearing in the carbonate form, all in the albumin (Caner, 2005). The pH of fresh albumin is almost 7.4 but as the egg ages and CO_2 escapes through the pores of shell, increasing alkalinity, the pH eventually increases to 9.6, depending on storage temperature (Alais & Linden, 1991; Silversides

& Scott, 2001; Laghi *et al*., 2005). CO_2 comes from the bicarbonates in the egg and dissolved in the albumin during shell calcification (Alais & Linden, 1991). As a result of low content of CO_2 in the environment eggs lose it in a few days after laying. CO_2 evaporates more or less rapidly depending on the temperature of storage (Alais & Linden, 1991). The albumin pH of coated eggs (8.4) was lower than that of uncoated eggs (9.5) stored at room temperature. Coating shell eggs with mineral oil reduces CO_2 permeability through the egg shell. If eggs are sealed with oil much CO_2 is preserved because rate of evaporation is very slow and the increase in pH is minimal. The mineral oil coating acts as a barrier because it minimises diffusion of gases through the shell. According to Li-Chan *et al.,* (1995), Brooks & Pace stated that the pH of albumin depends on the equilibria among dissolved CO_2, bicarbonate ions (HCO_3^-), carbonate ions (CO_3^{2-}) and proteins. The concentrations of HCO_3^- and CO_3^{2-} ions are controlled by the partial pressure of CO_2 in the outside environment. The rise in the concentration of CO_2 in the environment causes the concentration of HCO_3^- to increase as the CO_3^{2-} concentration decreases (Li-Chan *et al.,* 1995). Low concentration of CO_2 in the environment causes a decrease in the concentration of CO_2 inside the egg. A dynamic equilibrium is the result of molecules moving from a region of higher concentration to a region of lower concentration. High concentration of CO_2 inside the albumin means that the egg is more acidic. In alkaline conditions, most of the CO_2 and hydrogen ions have been lost and hydroxide ions are dominant. Uncoated eggs always had higher yolk pH than coated eggs stored at all three different storage conditions (Li-Chan *et al.,* 1995).

5.6 Conclusions

Oil coating can maintain the Haugh unit (albumin quality), pH of albumin and yolk and visual sensory attributes of broken out raw eggs. Foaming capacity of albumin was negatively affected by oil coating. Bakeries and processing industries where foaming capacity of eggs are important should not use oil coated eggs in their products or compensate in other ways for the lack of foaming ability. Coating with mineral oil reduces the loss of water and CO_2 in shell eggs, which in turn retard a rapid increase of the albumin pH.

This study demonstrates that mineral oil is effective in preserving the interior quality of eggs. Mineral oil coating preserves Haugh unit, visual sensory characteristics, and

albumin pH of WSE if stored at 16ºC; 58% RH. Oil coated eggs stored at 16ºC; 58% RH can possibly be graded as AA according to the US grading system even after 6 weeks of storage. The quality of coated WSE stored at 16ºC; 58% RH was maintained compared to uncoated eggs stored under the same conditions and the shelf life of such eggs was extended by three weeks based on Haugh unit. In comparison, uncoated eggs would be graded lower (US Grade A) from week 1 to week 3 and thereafter even lower (US Grade B). Mineral oil coating was also effective in maintaining the quality of eggs stored at 25ºC during the day and 15ºC at night (55% RH) for 5 weeks based on Haugh unit. Coating shell eggs with mineral oil may assist industries in reducing economic losses during storage. Storage conditions of 32ºC; 32% RH is not suitable for keeping the quality of both coated and uncoated WSE for longer than four weeks.

CHAPTER 6

GENERAL DISCUSSION

6.1 Discussion of methods used

The study was divided into two sections, the first to determine the effect of microwave pasteurisation at different power levels (250W and 300W) on the Haugh unit, pH, foaming properties of egg albumin as well as sensory properties of broken out eggs. Whole shell eggs were positioned on, either left or right hand side of the oven cavity. Oven position was identified as an additional experimental variable. The second section involved the determination of the effect of oil coating shell eggs on the functional properties and shelf life of WSE stored at 16°C (58% RH), 25°C & 15°C (55% RH) and 32°C (32% RH) for a period of six weeks.

The weight of a whole shell eggs was recorded by weighing balance. The albumin height is the height of the inner thick egg albumin surrounding the yolk (Lepule, 2003). To determine the height of albumin, the egg was broken and the height of the albumin was measured approximately 5mm away from the egg yolk at least three places for each egg with a tripod micrometer (Monira *et al*., 2003). The micrometer was mounted on a specially designed stand (Lepule, 2003). The poultry industry uses Haugh unit as the standard measure of egg quality. It could also be a measure of visual appearance, because it shows the appearance of the egg when it is broken onto a flat surface (Williams, according to Lepule, 2003). The Haugh unit values were calculated for individual eggs. The Haugh unit was calculated by a formula involving the weight of the egg and the height of the thick albumin (Monira *et al*., 2003) immediately surrounding the yolk. Fresh eggs exhibit high Haugh unit values and their albumin remained firm (Berardinelli *et al*., 2003). Haugh unit is thus a parameter used to evaluate the thinning of the thick albumin during storage due to change in the gelatinous structure (Robinson & Monsey, 1972) and probably influenced by the increase in albumin pH due to the loss of CO_2 through the shell (Burley & Vadehra, 1989). In the present study, the Haugh unit values of stored eggs especially uncoated eggs were very low which showed that thinning of thick albumin was observed.

Pasteurised eggs had higher Haugh units compared to unpasteurised eggs because some of the pasteurised albumin was slightly to partially coagulated, which appear then as higher albumin height giving a false perception of freshness. A Haugh unit was a useful measure to assess the quality of fresh, stored and slightly coagulated albumin but not for very coagulated albumin. For some of the stored eggs, particularly uncoated ones, it was difficult to distinguish the thick albumin from the whole albumin because it was runny and watery fluid.

For pH determination, WSE were individually broken with a knife. Egg albumin was separated from the yolk using an egg separator. Prior to pH determination, yolk pools and liquid egg albumin were transferred into two separate stomacher bags and then homogenised for 30s in a stomacher (Hou *et al*., 1996). pH and temperature of the homogenised albumin and yolk respectively were measured. The pH is one of the most useful indicators for shell eggs during storage due to the pH increase of albumin over storage time (Silversides *et al.,* according to Chang & Chen, 2000). The albumin pH is an important indicator of quality retention, processing and performance. When measuring pH, albumin temperature varied between 18ºC and 24ºC. The temperature could influence the measurement. It would have been better to standardise the temperature, by placing the eggs in an incubated storage area for a while prior to pH measurement.

Foaming capacity is one of the most important properties of albumin proteins in the manufacture and culinary preparation of food products. Several methods have been reported for incorporation of air into a protein solution, including whipping, injection, sparging and shaking (Waniska & Kinsella, according to Baniel, Fains & Popineau, 1997). Whipping is often used to measure foaming properties of protein solutions. Foaming ability of albumin is dependent on the quality of albumin proteins, and slight contamination with yolk can change protein functionality and reduce foaming properties of the albumin (Kirunda & McKee, 2000). Some proteins are denatured by the beating action and then aggregate to improve stability of the developing foam (Fennema, 1985). Stability of foam is determined by measuring drainage of liquid from a given quantity of foam over a specific period of time. The method used to determine foaming capacity was useful but it took long to measure foam stability of albumin.

Descriptive sensory evaluation is the identification, description, and quantification of the sensory attributes of a food product using human senses that have been trained specifically for this purpose (Einstein, 1991). It also include all sensory parameters (i.e. appearance, aroma, taste, and textural characteristics) to describe the complete profile of a specific product. In this study, there were 16 individuals selected as panel members. These people had the ability to consistently identify and quantify sensory characteristics in a given set of eggs using selected descriptors. A panel was trained properly and managed to develop egg sensory descriptors for broken out eggs that were used during evaluation. It was a good idea to use descriptive sensory evaluation because the panel was familiar with chicken eggs and understood the way fresh eggs looks like when broken out of their shells. So it was not too difficult to describe the sensory visual appearance of unpasteurised and pasteurised raw broken out eggs.

The triangle test, as a difference test method, was used to determine whether a significant difference existed between scrambled eggs prepared from control and microwave pasteurised (300W). There were 52 consumers that participated in this test. The Triangle test showed that there was a significant difference between control and pasteurised (300W) eggs. The control and pasteurised (300W) eggs varied in degrees or levels of denaturation of albumin in such a way that consumers did not find it very difficult to identify the different egg. Therefore, triangle test was a useful method.

In the present study, a consumer home use test was used to determine how much the visual appearance and overall acceptability of microwave pasteurised WSE eggs were liked or disliked by consumers. Panellists were asked to look at broken out eggs and indicate how much they like or dislike the visual appearance as well as overall acceptability of the eggs. Consumers seemed to like the visual appearance and overall acceptability of control and pasteurised (250W) eggs. Although products may differ slightly, these small differences do not necessarily affect consumer acceptance. This can only be assessed by the target consumer market. Hence, consumer home use test method was useful and had positive effect on this project.

6.2 Discussion of results

In this study, there were two microwave power levels (250W & 300W) used to pasteurise whole shell eggs. The effects of microwave heating of shell eggs on functional properties of albumin was assessed and compared to that of unpasteurised eggs. It was demonstrated that egg albumin, microwave heated at 300W, produced lower foams compared to albumin microwave heated at 250W. Heat pasteurisation of egg albumin leads to the formation of dense, irregular clumps of aggregates that affect functional properties such as foaming (Wong & Kitts, 2003).

Power level 250W managed to give at least a log 2 reduction in counts of *S*. Enteritidis for whole shell eggs. This power level (250W) was not sufficient to completely eradicate all *S*. Enteritidis from shell eggs as reported by CSIR microbiologists (Erasmus, Pinches, Rech, Rossouw, De Kock, Wiid, 2006). This explains that 250W did not produce enough heat to kill this pathogen but albumin functionality of these eggs was only slightly affected by the heat. The visual sensory properties of microwave pasteurised (250W) eggs were minimally affected compared to those processed at 300W. Pasteurising whole shell eggs at 300W resulted in overheating of the albumin and partially cooked eggs which led to low foaming capacity of the albumin. This power level (300W) produced enough energy to completely destroy *S*. Enteritidis from shell eggs (Erasmus *et al*., 2006) but with severe damage of the visual sensory properties of eggs and foaming capacity of albumin. At least a log 5 reduction in counts of *S*. Enteritidis was achieved by 300W power level. Thus, 300W was more effective in reducing counts of *S*. Enteritidis than 250W.

Uneven distribution of energy caused variations in the internal temperature of eggs. Some reach higher temperatures e.g. 57ºC while others were only heated to 48ºC. Some eggs reached protein denaturation temperatures and the egg albumin structure was affected. Some eggs were potentially under-heated and the functional properties were not affected but the heat treatment may not be sufficient to destroy salmonella.
There are several factors that affect heating in the microwave oven such as egg shell quality, egg mass, egg shape, time of exposure, and egg temperature before processing. Low egg shell quality causes breakages inside the oven cavity and hence it is not good for microwave heating**.** Combining different sizes and shapes of shell

eggs may influence internal temperature inside the eggs during microwave heating. If whole shell eggs had the same internal temperature at the beginning of the process, uniform heat can be distributed in the microwave oven and the process will be shortened. Though heating uniformity can be an issue in microwave heating, it can be overcome with the proper positioning of the eggs and by the precise design of the containers taking the eggs into the microwave cavity. Improving process control may reduce process variability that causes variations in egg quality (Erasmus *et al*., 2006). The results of this study subsequently led to new improved designs of the industrial microwave cavities with significantly improved heat distribution. Careful sorting of eggs within very tight weight classes also significantly alleviated the problem of individual variations in heat distribution. Additionally, the commercial units also incorporated rolling systems for the eggs, eggs are therefore exposed to the microwaves while moving which enhances heat distribution.

Microwave pasteurisation could contribute in producing safe shell eggs to the world. Furthermore, pasteurised eggs will have a longer shelf life and could improve the health of consumers worldwide by reducing known harmful microbes associated with whole shell eggs. The microwave system will help to increase product variety of egg packers locally and internationally as well as lead to job creation. From this project, scientific knowledge about microwave technology applications has been gained (Erasmus *et al*., 2006).

The second part of the study involved the determination of the effects of oil coating on the functional properties and shelf life of WSE stored under different conditions.

Application of coatings on shell eggs reduces weight loss and maintains their internal quality measured by several indices such as air cell height, Haugh units, yolk index, and albumin pH (Imai, 1981). In this study whole shell eggs were coated with mineral oil in order to preserve the albumin quality of eggs and prolong shelf life. Even though coating maintained the interior quality and the visual sensory properties of eggs, foaming capacity of albumin was affected. For this reason, food industries utilising albumin's foaming properties such as bakeries will not benefit, they will be affected. Food industries that do not use albumin's foaming ability will benefit from using oil coated eggs. Mineral oil was effective in reducing the loss of water through

the pores of shell eggs stored under different conditions. This is important because oil seals the pores on egg shells. This technology also slows down the increase of albumin pH. As pH of the albumin and yolk increases, the albumin becomes less firm and runny and the round shape of the yolk begins to deform and flatten. Eventually, these changes contribute to the loss of albumin functionality and sensory properties. The shelf life of the coated eggs was significantly longer, which will be of great benefit especially in the rural areas where optimal storage facilities for eggs at the pack stations and shops are not available. Therefore, mineral oil is an effective coating agent that can be useful if applied to chicken eggs by small scale as well as commercial producers.

CHAPTER 7

CONCLUSIONS AND RECOMMENDATIONS

Microwave pasteurisation induced denaturation of albumin proteins which lead to changes in the functional properties of eggs. Microwave pasteurisation leading to internal egg temperature exceeding 57°C destroys *S.* Enteritidis and reduces the functionality of albumin particularly foaming capacity. The quality of eggs pasteurised at 300W was lower than the quality of eggs pasteurised at 250W. The albumin foaming capacity of eggs pasteurised at 250W was slightly affected by microwave heating. Microwave heating process also affects the visual appearance and sensory properties of raw egg contents, particularly albumin firmness, albumin coagulation, prominence of chalazae and albumin clearness. Variation in microwave energy distribution causes variations in the internal egg quality. The visual sensory properties of eggs that were positioned on the left side in the microwave oven were more adversely affected and some were partially cooked or very coagulated. Damage was due to uneven distribution of microwaves in the oven.

Shell eggs pasteurised at the right hand side of the prototype microwave oven had acceptable functional properties. Eggs pasteurised on the left hand side of the microwave oven appeared to have partially cooked albumin. These eggs pasteurised on the left side of microwave oven should not be used to prepare either sponge cake or meringue, because they will not produce high quality foams.

The quality of eggs microwave pasteurised at 250W was closer to that of unpasteurised eggs. It is better to use 250W to microwave pasteurise eggs using the first prototype pilot-plant microwave oven. Variations in the distribution of microwaves to the eggs need to be fixed because it causes variation in quality. This will make a difference to ensure uniform quality of eggs. Microwave pasteurisation of shell eggs could become a significant break through for the poultry industry. Successful commercial applications will produce new and safer products as pasteurised shell eggs were not sold in South Africa before this development. Infants, pregnant women, sick person, weakened immune systems individuals and older people stand to benefit the most from the introduction of pasteurised WSE.

The value, reliability and validity of the Haugh unit measurement to determine the quality of pasteurised eggs are not clear and may require further investigation. Visual appearance and sensory properties of eggs pasteurised at 250W were acceptable to consumers. Foaming capacity of albumin was negatively affected by oil coating. Bakeries and processing industries where foaming capacity of eggs are important should not use oil coated eggs in their products or compensate in other ways for the lack of foaming ability.

This study demonstrated that mineral oil is effective in preserving the interior quality of eggs such as Haugh units, albumin and yolk pH, and visual sensory properties. Coated whole shell eggs also had a long shelf life. Coating shell eggs with mineral oil delays the deterioration of egg quality during storage. This technology of coating shell eggs with mineral oil may assist industries in reducing economic losses during storage. Coating eggs with mineral oil is economically important in rural areas where cooling facilities are not available and not affordable.

CHAPTER 8

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LIST OF APPENDICES

APPENDIX A

EGGCITING PROJECT

WANTED

We need 12-15 people that would be interested in a sensory evaluation research project over the next two months for the evaluation of eggs. Full training will be provided and no prior experience is necessary. You will have to be available three times a week for a 1-hour evaluation session.

YOU WILL BE PAID FOR YOUR PARTICIPATION.

If you are interested please contact MARISE urgently at the Dept of Food Science, UP before Monday, 30 August 2004.
Tel: 420 3238

E-mail: marise.kinnear@up.ac.za
Room: 2- 36 Old Agriculture Building

APPENDIX B

University of Pretoria

Department of Food Science

Thank you for your interest in our Egg project. Please complete the following questionnaire. Note that all information provided will be treated in a confidential manner.

Surname _____ Full Names _____

Age: _____

Postal address _____ Physical address _____

_____ _____

_____ _____

Tel. No (home) _____ Tel no (cell) _____

Degree _____

Department _____

Faculty _____

Student no _____

Personnel no _____

E-mail address _____

Do you smoke?	**YES**	**NO**
Are you pregnant?	**YES**	**NO**
Do you use any prescription medicine?	**YES**	**NO**

If yes, please specify:

Chronic conditions that may affect sensory perceptions? (e.g. poor eyesight, hay fever, dentures, other specify)	**YES**	**NO**
Do you suffer from any food allergies?	**YES**	**NO**

If yes, please specify:

Screening sessions

The screening tests will take place on 31 August 2004

What time will suit you best?

31 August	10:30	12:30
1 September	10:30	12:30

Other suggestions:

Training sessions

The training sessions will be on 2 & 6 September 2004:

What time will suit you best?

9:30	10:30

Other suggestions:

Laboratory evaluation times

Tuesday	Wednesday
7 Sept	8 Sept
14 Sept	15 Sept
21 Sept	22 Sept

9:30	10:30

Please make an X to indicate the time that suits you best

Other suggestions:

APPENDIX C

Set no: 1
Name: _____ Date: _____

You have received five egg samples. One sample is coded as the control sample, and the others have three-digit codes.

Please look at sample 456 and compare the visual appearance of this sample with that of the control sample. Please tick the relevant box.

Same as Control		If not the same, please describe the differences
Slightly different to Control		
Moderately different to Control		
Very different to Control		
Extremely different to Control		

Please look at sample 790 and compare the visual appearance of this sample with that of the control sample. Please tick the relevant box.

Same as Control		If not the same, please describe the differences
Slightly different to Control		
Moderately different to Control		
Very different to Control		
Extremely different to Control		

Please look at sample 651 and compare the visual appearance of this sample with that of the control sample. Please tick the relevant box.

Same as Control		If not the same, please describe the differences
Slightly different to Control		
Moderately different to Control		
Very different to Control		
Extremely different to Control		

Please look at sample 345 and compare the visual appearance of this sample with that of the control sample. Please tick the relevant box.

Same as Control		If not the same, please describe the differences
Slightly different to Control		
Moderately different to Control		
Very different to Control		
Extremely different to Control		

APPENDIX D

Triangle Test

Name: _____ **Date:** _____

Set: 1

You have to evaluate a total of 10 sets of eggs today. For each set, there are three coded samples. **Two samples are the same and one is different.** Please look at the samples and circle to code of the sample that is different.

290	872	912

Please note any further observations /comments here:

Set: 2

You have to evaluate a total of 10 sets of eggs today. For each set, there are three coded samples. **Two samples are the same and one is different.** Please look at the samples and circle to code of the sample that is different.

874	623	325

Please note any further observations /comments here:

Set: 3

You have to evaluate a total of 10 sets of eggs today. For each set, there are three coded samples. **Two samples are the same and one is different.** Please look at the samples and circle to code of the sample that is different.

585	809	106

Please note any further observations /comments here:

APPENDIX E

Yolk size	1 2 3 4 5 6 7 8 9 **Small** **Large**
Height of yolk	1 2 3 4 5 6 7 8 9 **Low** **High**
Brightness of yolk Intensity of bright yellow colour	1 2 3 4 5 6 7 8 9 **Dull yellow** **Bright yellow**
Blemishes on yolk Presence of round dark spots in the yolk that appear as shadows	1 2 3 4 5 6 7 8 9 **Not blemished** **Very blemished**
Firmness of albumin How firm the thick albumin of the eggs is	1 2 3 4 5 6 7 8 9 **Not firm and compact** **Very firm and compact**
Albumin coagulation Presence of signs of coagulation of the albumin	1 2 3 4 5 6 7 8 9 **Not coagulated** **Very coagulated**
Prominence of chalazae How easy it is to see the chalazae	1 2 3 4 5 6 7 8 9 **Not prominent** **Very prominent**
Albumin clearness	1 2 3 4 5 6 7 8 9 **Not clear** **Very clear**

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APPENDIX F

HAVE YOU EVER FIND YOURSELF EATING EGGS ?

If so, come taste scrambled eggs and tell us what your taste buds have perceived.

Date: 25 October 2004

Venue: Old Agric Building - Department of Food Science, Sensory Evaluation Laboratory (Room 2-6)

Time: 9h30, 11h00 & 12h30

For more information, call us now!

Cell no: 072 521 6608

E-mail address: s24374203@tuks.co.za

YOU WILL BE REWARDED !!!

APPENDIX G

Panellist Code: _____
Panellist Name: _____

Instructions

Please take a sip of water before you start. Please evaluate the samples in the order presented on the tray, from left to right.

Please indicate your gender.

- o Male
- o Female

Please indicate your age

- o 15-20yrs
- o 21-30yrs
- o 31-40yrs
- o 41-50yrs
- o 50+ yrs

Triangle test

In front of you are three samples. Two of the three samples are the same, one of the samples is different. Taste the samples in the order indicated below and identify the DIFFERENT sample.

515 **335** **808**

Any comments?

_____
_____
_____

THANK YOU FOR YOUR PARTICIPATION

PLEASE COLLECT YOUR REWARD

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APPENDIX H

EGG CITING PROJECT **Set no**: 1

Department of Food Science

Faculty of Natural and Agricultural Sciences

University of Pretoria

Tel no: (012) 420 3238

Hello, I'm Sylvia from the Department of Food Science, University of Pretoria. We are conducting a research project on consumer acceptability of eggs. If you eat eggs you are very welcome to be part of this survey. Please fill in your details on the table below. NB. All the information that you give will remain strictly confidential and will only be used for statistical purposes. The results of this test may help us as Food Scientists to improve the quality of our food supply.

Date:	Time:			
Respondent name (Mr/Mrs/Miss)				
Residential Address				
Contact telephone no				
E-mail				
Gender	Male		Female	
Age	18-25	26 -35	36 - 45	45+years
Contact number: 012 420 3413 Room 2-4, Dept of Food Science Old Agriculture Building	Name of Researcher: SYLVIA MUDAU Signature			

Acknowledgement & Disclaimer

Risks to the Individual: Please note that the risk involved in eating the eggs that you will receive is no greater than that of eating eggs purchased in the retail consumer market.

Confidentiality: Responses to questions via the evaluation form are tracked using numbers only. These numbers are not in any way related to the participant' s name.

I, the undersigned_____ do hereby acknowledge and understand

that:

1. Participation in this survey is completely voluntary and at my own risk.
2. University of Pretoria shall under no circumstances be liable for any loss, damage or claim of whatsoever nature as a result of, or arising, directly or indirectly, from my participation in this survey (including whether such loss, damage or claim be personal or that of a third party).

Signature: _____

Date: _____

Signature of Researcher (Dr H.L. de Kock) _____

Set no:_____

Project placement

You have received two eggs to evaluate over the next **TWO** days (**one egg per day)**. It is very important that you eat both eggs personally. Please do not give the eggs to a neighbour, relative or family member, as we are interested in your personal opinion.

Please break one egg at a time, look at the appearance, cook and eat the egg. While you are preparing and eating each egg, I would like you to complete a questionnaire to find out your opinion on the quality of that egg.

It is important that you use the same cooking method such as boiling in water, frying, poaching or scrambling for both eggs.

Day	**Break and eat**
Day 1	
Day 2	

I will collect the forms on----------

Please contact me if you have any questions
Name: Sylvia
Contact number: 012 420 3238

Day One

Please take the egg numbered | 995 | and break it open on a small plate.

1. **Look at the broken out egg and tell us how much you like or dislike the visual appearance of this egg by circling the appropriate number.**

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**

Comments: ----------

---

2. **Cook the egg, as you would normally do. Please indicate which cooking method you used by marking with a X.**

Boiling
Frying
Poaching
Scrambling
Other, please specify

3. **While you are eating the egg, indicate how much you like or dislike this egg by circling the appropriate number?**

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**

Comments: ----------

Day Two

Please take the egg numbered | 508 | and break it open on a small plate.

1. **Look at the broken out egg and tell us how much you like or dislike the visual appearance of this egg by circling the appropriate number.**

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**

Comments: ----------

---

2. **Cook the egg, as you would normally do. Please indicate which cooking method you used by marking with a X.**

Boiling
Frying
Poaching
Scrambling
Other, please specify

3. **While you are eating the egg, indicate how much you like or dislike this egg by circling the appropriate number?**

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**

Comments: ----------

Consumers were also asked questions of the home use test as follows:

1. On average, how often do you eat eggs? Mark X the appropriate box

Less than once per week	Once per week	2 times per week	3 to 6 times per week	Daily

2. When do you normally eat eggs? Choose only ONE option

Breakfast time	Lunch time	Dinner / Supper time

3. Rank the following factors used when you select eggs to buy in order of importance. Put a 1 in the box for the most important factor, 2 for the second most important factor and so on.

Factors	Rank (1 to 5)
Brand name	
Freshness (sell by date or expiry date)	
Packaging	
Price	
Visual appearance of shell eggs (clean, not broken)	

4. When breaking open a raw egg, what do you use as the main criteria for judging if it is fresh? Choose only ONE option.

Colour	Appearance of albumin	Appearance of egg yolk	Smell	Other, please specify

5. When eating an egg, what do you use as the main criteria for judging if it is fresh? Choose only ONE option.

Smell	Texture	Taste	Other, please specify

6. Where do you normally store eggs after purchase?

In the refrigerator	In the Cupboard	Other, please specify

7. On average, for how long do you normally store eggs after purchase?

1 week	2 weeks	3 weeks	4 weeks	More than 4 weeks

8. How concerned are you with potential safety risks when eating eggs?

Not concerned at all	Very little concerned	Slightly concerned	Moderately concerned	Very Much concerned	Extremely concerned