Microwave Pasteurization of Shell Eggs

- A Comprehensive Study

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ABSTRACT

Due to their rich nutritive value, eggs are potential hosts and carriers of pathogenic microbes like *Salmonella enteritidis*. Heat pasteurization is the best solution for controlling these pathogens, but affects the egg's vital functional properties due to protein denaturation. Therefore, microwave heating was considered for in-shell egg pasteurization.

Based on a few laboratory trials, finite difference time domain (FDTD) and finite element models (FEM) were developed to simulate the electric field and power distribution in the egg components (egg white and yolk), taking into consideration the complex shape, dielectric properties and heterogeneous composition of the in-shell egg.

Using the simulation results, process optimization was carried out to determine the most effective procedure and design for the process. Laboratory-scale experimental trials were conducted to test the validity and effectiveness of the optimized parameters. Operations under the optimal parameters set forth were found to be very efficient in terms of heating time and uniformity.

Based on the optimal parameters obtained by simulations, a slotted waveguide applicator for heating shell eggs was designed and built. The applicator consisted of a standard waveguide with an array of S–Parabolic slots. The issue of non-uniformity in microwave heating was overcome by optimizing the power density used for the process and by rotating the egg during the heating process. A power density of 1.5 W g^{-1} and an angular velocity of $\frac{\pi}{6}$ rad s^{-1} were found to be optimal. The applicator enhanced both penetration and focus, as well as providing the necessary temperature gradient from the egg yolk to the shell for pasteurization.

The pasteurization process was validated by inoculating eggs with a microbial contaminant and pasteurizing them in the designed applicator. Heat-induced changes in the egg white's physical properties brought about by in-shell pasteurization by microwave or water bath heating of the egg white were assessed in comparison with the initial state of untreated raw egg white. Microwave-heated in-shell egg white showed minimal changes in all these properties.

A protocol for real-time inline pasteurization quality assessment of shell eggs by hyperspectral imaging (400-1700 nm) was developed by identifying 10 informative wavelengths and doing an unsupervised k-means classification of treated eggs.

RESUME

La grande valeur nutritive de l'œuf le rend potentiellement susceptible de servir comme hôte et porteur de microbes pathogéniques tel *Salmonella enteritidis*. La pasteurisation par la chaleur est la meilleure solution au contrôle de ces pathogènes, mais, suite à une dénaturation des protéines, elle a un effet néfaste sur d'importantes propriétés fonctionnelles de l'œuf. C'est pourquoi la possibilité d'une pasteurisation par réchauffement par micro-onde fut considérée pour les œufs en coquille.

Fondé sur quelques épreuves en laboratoire, des modélisations par domaine de différence finie en temps et par éléments finis furent mise au point pour simuler la distribution du champ électrique et de la puissance dans différents constituants de l'œuf (blanc et jaune d'œuf), en tenant compte de la forme complexe, des propriétés diélectriques, et de la composition hétérogène d'un œuf en coquille.

Se basant sur les résultats de simulation, un procédé d'optimisation fut exécuté afin de déterminer le procédé et la conception les plus efficaces pour ces fins. Des essais expérimentaux à l'échelle du laboratoire visèrent à évaluer la validité et l'efficacité des paramètres optimisés. Le procédé opérant sous les paramètres optimisés énoncés se montra plus efficace en termes de la durée de mise en température et de son uniformité, que le procédé de pasteurisation à l'eau chaude. Ces paramètres optimaux, ayant leur origine dans des simulations, dirigèrent le design d'un applicateur équipé d'un guide d'ondes à fentes, servant au réchauffement d'œufs en coquille. Cet applicateur consiste en un guide d'ondes conventionnel avec une série de fentes paraboliques en forme de S. Les difficultés liées au manque d'uniformité du réchauffement par micro-ondes furent surmontées en optimisant la densité de puissance en place durant le procédé, et en tournant l'œuf durant toute la période de chauffage. Une densité de puissance de 1.5 W g^{-1} et une vitesse angulaire de $\frac{\pi}{6}$ rad s^{-1}

s'avérèrent optimales. L'applicateur améliora la pénétration et la concentration des micro-ondes, en plus d'engendrer un gradient de température entre le jaune et la coquille qui est idéal pour la pasteurisation.

Le procédé de pasteurisation fut validé en inoculant des œufs avec un contaminant microbien et en les pasteurisant avec l'applicateur conçu durant cette étude. Les changements dans les propriétés physiques du blanc d'œuf causés par la chaleur advenant de la pasteurisation en coquille par micro-ondes ou par chauffage dans un bain d'eau chaude furent évalués par rapport au blanc d'œuf n'ayant reçu aucun traitement. Les œufs en coquille ayant été chauffés aux micro-ondes ne montrèrent que des changements très limités à leurs propriétés.

Un processus d'évaluation en direct de la qualité d'œufs pasteurisés en coquille par radiométrie spectrale imageante (400-1700 nm) fut conçu en identifiant 10 longueurs d'ondes portant des informations utiles et procédant à une classification non supervisée par K-moyennes des œufs ayant subi un traitement.

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DEDICATION

I dedicate this research to the promotion of global food safety and the protection of consumer health. Especially to people who hate to cook their eggs and consumers who love to eat them raw.

Good Luck!

But please get them pasteurized.

CONTRIBUTIONS OF THE AUTHORS

The first four manuscripts in this thesis are by S.R.S. Dev, Y. Gariépy, V.Orsat and G.S.V. Raghavan; the fifth manuscript is by S.R.S. Dev, D. Lyew, V.Orsat and G.S.V. Raghavan; and the sixth manuscript is by S.R.S. Dev, N. Abdel-Nour, M. Ngadi and G.S.V. Raghavan. All the authors are from the Department of Bioresource Engineering, Macdonald Campus, McGill University, Ste-Anne-de-Bellevue, QC. This study was performed by the candidate and supervised by Dr. G.S.V. Raghavan. The entire research work was done at the postharvest Technology laboratory of McGill University. Mr. Y. Gariépy was involved in the technical issues of instrumentation and control for all the experiments in this study, giving expert guidance in the usage of equipment and making major contributions reviewing and revising the manuscripts. Dr. V. Orsat was personally involved in giving valid suggestions for improvement in every stage of the study and made great contributions in reviewing and improving the writing of the manuscripts. Dr. D. Lyew gave expert advice and technical support for the microbiological studies included in this research. Mr. N. Abdel-Nour and Dr. M. Ngadi provided support and guidance in using the hyperspectral imaging equipment.

NOMENCLATURE

E	Total Electric field intensity (V m^{-1})
E_x	Electric field intensity x component (V m^{-1})
E_y	Electric field intensity y component (V m^{-1})
E_z	Electric field intensity z component (V m^{-1})
H	Total Magnetic Field Intensity (A m^{-1})
H_x	Magnetic field intensity x component (A m^{-1})
H_y	Magnetic field intensity y component (A m^{-1})
H_z	Magnetic field intensity z component (A m^{-1})
f	Frequency of microwaves (Hz)
ε'	Dielectric constant
ε''	Dielectric loss factor
ε_0	Permittivity of free space (F m^{-1})
μ_0	Permeability of free space (H m^{-1})
P_{av}	Time average power dissipated (W)
P_c	Poynting Vector – power dissipated over unit area (W m^{-2})
ρ	Density of the material (kg m^{-3})
C_p	Specific heat capacity of the material (kJ kg^{-1} °K^{-1})
T	Temperature (°K)
T_c	Temperature (°C)
K	Thermal conductivity (W m^{-2} °K^{-1})
Q	Power Source Term (W m^{-3})
V	Volume (m^3)
n	Unit vector normal to the surface
A	Cross sectional area of the waveguide
α & β	Arbitrary constants

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Chapter 1

GENERAL INTRODUCTION

Egg is a popular ingredient in many foods and widely used in the food industry. Eggs are among the major foods of animal origin generally marketed and frequently consumed raw. Due to their rich nutritive value of their contents, eggs are potential hosts and carriers for pathogenic microbes like *Salmonella enteritidis*. More than 90% of food-borne Salmonellosis, caused by *S. enteritidis,* occurs through shell eggs (Schroeder et al. 2005; Woodward, Khakhria, and Johnson 1997).

The egg market in Canada is regulated. Canadian egg farmers produce about 420 million dozen grade A eggs, accounting for an average of 750 million dollars *per annum* (EFC 2008). On average, the cost of production is $1.70 (CAD) per dozen grade A eggs, nearly twice that of their American counterparts. This occurs because the Canadian poultry industry operates on the principle of “Start Clean and Stay Clean” (CEMA 2002). While more expensive, this makes Canadian eggs amongst the safest in the world.

Presently most of the commercially available pasteurized eggs are pasteurized using conventional heating methods by separating the yolk and egg white before processing. But breaking and repacking them aseptically involves huge additional costs. Therefore in-shell egg pasteurization has gained a great commercial importance in recent years.

Current techniques for in-shell pasteurization of egg involve heating the eggs in a water bath at 60ºC for 20-25 minutes, depending on the size of the eggs. This leads to the overheating of the egg white proteins (i.e. the egg white gets heated up more than

the yolk, which is against the recommendations) resulting in denaturation and coagulation (Hou et al. 1996). This greatly affects the functional properties of the egg constituents. Therefore a process that can heat the shell eggs from inside would be the best alternative to solve this problem.

Microwaves have the ability to generate heat from within a substance that is exposed to it. Theoretical mathematical studies have shown that even though albumen exhibits better dielectric properties than yolk, the egg's curvature has a focusing effect which leads to a suitable power distribution (Datta et al. 2005). Hence the shell egg appears ideally suited for pasteurization in a microwave environment (Fleischman 2004; Rehkopf 2005; Dev et al. 2008).

1.1 Problem Statement

Due to their rich nutritive value, eggs are potential hosts and carriers for pathogenic microbes like *S. enteritidis*. Heat pasteurization is the best solution for controlling these pathogens. More than 90% of food-borne Salmonellosis occurs through shell eggs (Schroeder et al. 2005; Woodward, Khakhria, and Johnson 1997). Although there are several advanced methods used for microbial disinfestation, including rapid chilling and ultrasonic treatments to destroy *Salmonella*, they are not effective on the *Salmonella* present inside shell eggs (Hou et al. 1996). It is also clear from the thermal conductivity values of the albumen (0.552 $Wm^{-1}K^{-1}$) and yolk (0.397 $Wm^{-1}K^{-1}$) (Coimbra et al., 2006) that the amount of energy and temperature gradient required to setup convection currents inside the eggs is much higher than those used for the pasteurization process. Therefore the majority of heat transfer occurs only through conduction and is very slow.

Pasteurization is considered as the best solution to the *S. enteritidis* problem in eggs. The Food Safety and Inspection Service (FSIS) of United States Department of Agriculture (USDA) recommends heating the egg white and the egg yolk to 57.5°C and 61.1°C respectively for 2.5 minutes to ensure egg safety against *Salmonella* and other food-borne pathogens (FSIS-USDA 2006).

The current technology uses batch hot water immersion or moistened hot air or both combined, which requires a long treatment time, in the order of hours, to complete. This process is neither very energy efficient given the poor thermal properties of the shell and shell membrane — though they are not really the focus of the pasteurization — nor is it cost effective (Mermelstein 2001). Furthermore, these treatments affect the functional properties of the egg components, which is an extremely important consideration in the food industry.

Proteins are highly heat-sensitive components of the egg. The functional properties of whipability, foamability, foam stability, *etc*. which make the egg an inevitable ingredient of various food products are severely affected by high temperatures. Experimentally it has been shown that for pasteurization the egg yolk needs to be heated to a higher temperature than the albumen. This is possible by conventional heating only if the yolk and albumen are separated (i.e. only if the shell is broken), as the yolk is concentric within the albumen in a shell egg. The existing methods of shell egg pasteurization result in overheating of the albumen and partially cooked eggs along the shell membrane (Hank et al. 2001).

1.2 Hypothesis

Microwaves can be used to raise the temperature of in-shell eggs to the required pasteurization temperature in minutes. Microwave have been shown to enhance the thermal destruction of microbes (Tajchakavit 1997). Microwaves are not ionising

radiation, but the dielectric properties of the microorganism enhance heat generation within it, leading to its destruction in a microwave environment. The microwave power distribution inside shell eggs also seems to be well-suited for uniform pasteurization. Very little work has been done on making microwave pasteurization viable for industrial use and there is very limited literature available.

Microwave pasteurization of eggs can make the process faster and continuous, such that the complete operation can be done in a few minutes. The shell egg appears to be ideally suited for pasteurization in a microwave environment (Fleischman 2004; Rehkopf 2005). Though heating uniformity can be an issue in microwave heating, it can be overcome with the proper orientation of the egg and a specially-designed waveguide, which is an engineering issue (Fleischman 2004), as well as the precise design of the container (equipped with microwave egg susceptors) taking the eggs into the microwave chamber (Yakovlev 2001).

1.3 Objectives

1.3.1 General Objectives

1. To define the conditions under which in-shell eggs can be successfully pasteurized using microwave energy at 2450MHz, without compromising quality.

2. To design a combination of waveguide and egg holders (susceptors) to accomplish the required temperature profile for pasteurization inside the shell egg.

1.3.2 Specific objectives

1. To develop a finite element model to predict the energy and temperature distribution inside in-shell eggs during microwave processing in order to improve uniformity and efficiency.

2. To optimize the microwave energy distribution in in-shell eggs within a multimode cavity using simulations validated by experimental trials.

3. To design a highly specific waveguide to suit this purpose and a carrier for taking the egg into the pasteurizer.

4. To validate the microwave pasteurization process using in-shell eggs inoculated with *Salmonella enteriditis* or equivalent non-pathogenic bacterial strains.

5. To assess and characterize the effects of microwave heating on the quality and functionality of in-shell eggs and their constituents.

6. To standardize a method for the industrial pasteurization of in-shell eggs using microwaves without compromising quality.

Chapter 2

GENERAL REVIEW OF LITERATURE

2.1The Incredible Egg

The egg is one of nature's marvels, designed to provide self-sustainability and excellent defence mechanisms to bring a fertilized cell to life as a chick. It is exquisitely simple, yet enormously complex. The eggs has remained a focus of research and development of food products for centuries. It has enthused several scientists and researchers in terms of its incredible functionality and functional properties, both as an individual entity and as an ingredient in several foods.

2.2 Composition of Eggs

The composition of a typical hen’s egg is illustrated in Figure 2.1. The intelligent design of Nature gives the eggs the best protection against most biological hazards. The egg has many natural, built-in barriers to help prevent bacteria from entering and growing. These protect the egg on its way from the hen to its hatching as a chick or to it entering our diet (American egg board: www.aeb.org).

However, although it does help, the porous shell itself is not a foolproof bacterial barrier. For further safety, government regulations require that eggs be carefully washed with special detergents and sanitized with chlorinated water. Then, the hen’s original protective shell coating is generally replaced by a thin spray coating of a tasteless, odorless, harmless, natural mineral oil. A shiny shell indicates oiling, rather than an unsafe or old egg.

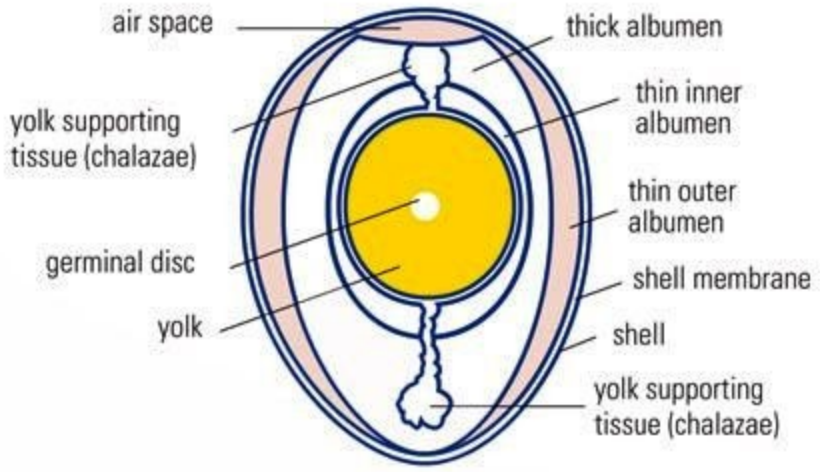

Figure 2.1 Composition of eggs

(Source: American egg board: www.aeb.org)

Other protective barriers include the shell and yolk membranes and layers of the egg white. These fight bacterial proliferation in several ways. The structure of the shell membranes helps prevent the passage of bacteria. The shell membranes also contain lysozyme, a substance that helps prevent bacterial infection. The yolk membrane separates the nutrient-rich yolk from the white.

In addition to containing antibacterial compounds such as lysozyme, layers of the white discourage bacterial growth because they are alkaline, bind nutrients that bacteria need and/or do not provide nutrients in a form that bacteria can use. The thick white discourages the movement of bacteria. The last layer of white is composed of thick ropey strands which have little of the water that bacteria need, but a high concentration of the white's protective materials. This layer holds the yolk centered in the egg where it receives the maximum protection provided by all the other layers.

2.2.1 Nutrient Value of Hen's Egg

Eggs provide significant amount of proteins as well as various other nutrients to one's diet. Chicken eggs are the most commonly eaten eggs, and are highly nutritious. Table 2.1 gives a comprehensive overview of the nutrient content of eggs.

Table 2.1 Nutritive value of a large chicken egg (weighing 65g)

Nutrient (unit)	Whole Egg	Egg White	Egg Yolk
Calories (kcal)	75	17	59
Protein (g)	6.25	3.52	2.78
Total lipid (g)	5.01	0	5.12
Total carbohydrate (g)	0.6	0.3	0 .3
Fatty acids (g)	4.33	0	4.33
Saturated fat (g)	1.55	0	1.55
Monounsaturated fat (g)	1.91	0	1.91
Polyunsaturated fat (g)	0.68	0	0.68
Cholesterol (mg)	213	0	213
Thiamin (mg)	0.031	0.002	0.028
Riboflavin (mg)	0.254	0.151	0.103
Niacin (mg)	0.036	0.031	0.005
Vitamin B6 (mg)	0.070	0.001	0.0069
Folate (μg)	23.5	1.0	22.5
Vitamin B12 (μg)	0.50	0.07	0.43
Vitamin A (IU)	317.5	0	317
Vitamin E (mg)	0.70	0	0.70
Vitamin D (IU)	24.5	0	24.5
Choline (mg)	215.1	0.42	214.6
Biotin (μg)	9.98	2.34	7.58
Calcium, Ca (mg)	25	2	23
Iron, Fe (mg)	0.72	0.01	0.59
Magnesium, Mg (mg)	5	4	1
Copper, Cu (mg)	0.007	0.002	0.004
Iodine, I (mg)	0.024	0.001	0.022
Zinc, Zn (mg)	0.55	0	0.52
Sodium, Na (mg)	63	55	7
Manganese, Mn (mg)	0.012	0.001	0.012

Source: (Li-Chan, Powrie, and Nakai 1995)

A typical hen's egg is made of 34% yolk and 63% white. Yolk is made up of 48% water, 31-35% lipids, 0.2-1.0% carbohydrates, 1-1.5% ash, and 15-16% protein. Comparatively egg white is made of 85% water, 0.02% lipids, 0.7% carbohydrate, 0.6% ash, and 13% protein. These proteins in the yolk and the white have very distinct functional properties and may undergo changes during processing (Lokhande et al., 1996).

Eggs supply a large amount of complete (containing all amino acids essential to humans), high-quality (readily absorbable) protein, and provide significant amounts of almost all vitamins (except vitamin C) and minerals (Li-Chan, Powrie, and Nakai 1995). Eggs are also one of the least expensive single-food sources of complete protein. One large chicken egg contains approximately 7 grams of protein. In fact, egg protein is of such high quality that it is used as the standard to which other proteins are compared.

Eggs have a biological value (efficacy with which protein is used for growth) of 93.7%, compared to values of 84.5% for milk, 76% for fish, and 74.3% for beef. Eggs really are the best protein money can buy, besides providing many other valuable vitamins and minerals. All of the egg's vitamins (A, D and E) are housed in the yolk. The egg is one of the few foods which naturally contain vitamin D. A large egg yolk contains approximately 60-75 calories, while the egg white contains about 15-17 calories.

A large yolk contains more than two-thirds of the recommended daily intake of cholesterol (300 mg) (Institute of Medicine, 2002), though human body does not absorb much cholesterol from eggs. Making up about one third of the liquid weight of the egg, the yolk contains all of the fat in the egg and slightly less than half of the protein and much of the nutrients. It also contains all of the choline. One yolk contains approximately half the recommended daily intake of choline. Choline is an important

nutrient for brain development, and is said to be important for pregnant and nursing women to ensure healthy foetal brain development.

2.2.2 Dietary Contribution and Affordability of Eggs.

Eggs are an important contributor to the nutritional quality of the Canadian diet. While eggs provide only 1.3% of the average caloric intake, they are so nutrient dense that they contribute a larger extent of the RDA (recommended daily allowance) for riboflavin (6%), folate (5%), vitamin E and vitamin A (4%), and protein (almost 4%) (CEMA 2004). Table 2.2 gives egg consumers' percentage nutrient intake from eggs. Eggs not only make a contribution to the nutrient value of the Canadian diet, they also make a major contribution to the affordability of the diet. At $2.20 (CAD) per dozen large eggs, the consumer pays only $1.35 (CAD) per pound for a nutrient-rich source with the highest quality protein available (CEMA 2004).

Table 2.2 Percentage nutrient intake from eggs by egg consumers

Nutrient	% RDA obtained from eggs
Energy (kcal)	9
Total fat (g)	17
Saturated fat (g)	16
Polyunsaturated Fat (g)	16
Cholesterol (mg)	61
Vitamin E (Tocopherol Equivalents)	21
Vitamin A (Retinol Equivalents)	25
Vitamin B6 (mg)	9
Folate (µg)	17
Vitamin B 12 (µg)	25

(Source: Song and Kerver, 2000)

2.3 Microbial Safety of Eggs

Eggs are highly rich in nutrients that form a suitable substrate for the growth and multiplication of microbes at room temperature. Hence, unless chilled to below 7℃ they are highly perishable and therefore require very careful handling to prevent food poisoning.The risk of getting a food-borne illness from eggs is very low. However, the nutrients that make eggs a high-quality food for humans are also a good growth medium for bacteria. In addition to food, bacteria also need moisture, a favorable temperature and time in order to multiply and increase the risk of pathogenicity.

The bacterium *Salmonella enteritidis* has been found inside a small number of eggs over the past couple of decades. Many of these were Grade A eggs, certified good for human consumption (St. Louis, Morse, and Potter 1988).

Since other types of microorganisms can also be deposited along with dirt on the outside of an egg, in Canada eggshells are washed and sanitized to remove possible hazards. Consumers are further protected by the discarding of eggs that are unclean, cracked, broken or leaking and by ensuring good hygienic practices in egg handling. Bacteria are most likely to be introduced into the white, but they will be unable to grow, mostly due to a lack of available nutrients and the white's antimicrobial activity. However, as the egg ages the white thins and the yolk membrane weakens, making it possible for bacteria to reach the nutrient-dense yolk where they can grow over time if the egg is kept at warm temperatures for extended periods of time (Fleischman et al. 2003). But, in a clean, uncracked, fresh shell egg, internal contamination occurs rarely.

If not properly handled, *Salmonella* bacteria can double every 20 minutes and a single bacterium can multiply into more than a million in 6 hours. To block *S. enteritidis* from multiplying in the egg, eggs must be held at cool temperatures (5ºC) following

packing and throughout transportation. Industry education programs encourage food preparers to use safe food-handling practices (FSIS-USDA 2006).

2.3.1 Sources of contamination

Microbial contamination of eggs can occur both within the hens and from the environment. The environment itself may harbor microbes in the poultry shed, soil, feed, water, rodents, bedding and contaminants brought in by people. The bacteria on the surface of a shell egg may come from the faecal contamination of the bird, as the egg exits through the same passage as faeces. Hence the surface of the eggs need to be washed and sanitized (USDA-FSIS, 2006). Microbes are also found inside a whole uncracked egg, where they are traced to the ovary or the oviduct of the bird.

From the outer shell they may enter the egg through the minute pores on the shell measuring 0.006-0.054 mm in diameter (Haines, 1939). The egg white is not favourable to the microorganisms as it contains antimicrobial principles including lysozyme, avidin, ovoflavonoids and ovotransferrin, which make the vital nutrients unavailable for microbial growth. The egg white also has an unfavorably high pH (9.6) that prevents the growth of microbes. The yolk is reported to be more easily infected than the white as it has the nutrients essential for the microorganisms' growth and the pH of the yolk is around 6.0, which is optimal for microbes. The vitelline membrane around the yolk harbors most of the microbes which bring about the weakening and rupture of the membrane (Haines, 1939).

2.3.2 Microbial profile of whole egg

Common contaminants found are micrococci, moulds, yeast and spore forming bacteria. The shell contains mostly Gram positive bacteria and some Gram-negative bacteria are found in rotten eggs (Board and Tranter, 1995). Most common

contaminants of the shell are *Micrococcus, Staphylococcus, Bacillus, Pseudomonas, Alcalgenes, Flavobacterium, Escherichia, Aerobacter, Acinetobacter* and *Cytophagia*. The common microorganisms in rotten eggs are *Pseudomonas, Alcalgenes, Escherichia, Serratia, Xanthomonas, Aeromonas, Citrobacter, Acinetobacter and Proteus*. Molds are of lesser importance, though under high humidity conditions they can grow on the shell and spread their hyphae into the inner surface of the membrane.

2.3.3 Salmonella- a criteria for egg safety

Of the pathogens in egg that bring about disease in man, *Salmonella* sp. are the most potent. This genus causes illness (salmonellosis) by invading the small intestines of the host and producing an enterotoxin that causes inflammation and diarrhea, which can, at times, be fatal. The most common *Salmonella* species in egg is *Salmonella enteritidis*. While these Gram-negative bacteria grow best at temperatures between 8-46°C, in a pH range of 3.8 to 9.5, and at water activities above 0.94 (Bell and Kyriakides 2002), they are capable of surviving in conditions of low water activity, and extreme pH and temperature conditions. They are destroyed at temperatures of 70°C and above, so they are susceptible to ordinary cooking temperatures if applied sufficiently long (Guthrie, 1992).

Raw eggs are used in some salad dressings and dessert preparations, where contaminated egg products can be hazardous if not stored at safe temperatures (4.4-21.1C) (Morrone, 2008; Zeilder, 2002). From 1993 to 1995, there were more than 20,000 laboratory-confirmed human cases of Salmonellosis reported in Canada (Woodward, Khakhria, and Johnson 1997). Food borne salmonellae are estimated to cause ≈1.3 million illnesses, 15,000 hospitalizations, and 500 deaths per year in the United States (Schroeder et al. 2005).

2.3.4 Status of Poultry Eggs in Canada

Today consumers' primary concern is the quality and safety of the food they consume. In-shell eggs and egg products are among the most commonly consumed animal food products. On an average Canadians eat around 15.6 dozen eggs *per capita, per annum*. In Canada, in-shell eggs are sold raw after being washed and surface-disinfected. The Agricultural Products Act describes the regulations for marketing and Health Canada is the regulatory body responsible to make sure that they are followed by the poultry industry. Amendments to the Act are presently being considered to define the conditions under which pasteurized in-shell eggs can be commercialized in the country (CEMA, 2004).

2.3.5 Pasteurization of Eggs

Pasteurization is defined as "a process of heating food for the purpose of killing harmful organisms such as bacteria, viruses, protozoa, moulds, and yeasts." (Lewis and Heppell 2000). The process was named after its inventor, French scientist Louis Pasteur.

Pasteurization does not completely kill or eliminate all the microorganisms present in the food. It is described as a mild process because the amount of chemical damage caused is small and the changes to the food's sensory characteristics are minimal. It aims to achieve a certain number of "log reductions" in the number of viable organisms, thus rendering the microorganisms ineffective.

Once pasteurized, it is also crucial to prevent the product from becoming recontaminated. Such recontamination is referred to in general terms as post-processing contamination, but more specifically in this instance as post-pasteurization contamination. To ensure this, care and attention should be paid to hygiene and general

aspects of cleanliness. After pasteurization, if the food is not refrigerated till consumed and/or not consumed within the recommended period, then the pasteurized food can no longer be considered safe for consumption.

Keeping quality is perhaps the most important commercial quality consideration. Since pasteurization only inactivates vegetative spores, the keeping quality will be influenced by a number of factors and may vary considerably. The important control factors are raw material quality, time/temperature conditions, reducing post pasteurization processing, and storage temperatures. Keeping quality can be extended by understanding and controlling the overall pasteurization process.

The U.S. Egg Products Inspection Act of 1970 introduced the regulation that egg products be rendered Salmonella-free through pasteurization (Stadelman and Cotterill, 1995). Heat treatment is also aimed at maintaining the functional properties of the egg like foaming, emulsification and gelling which can be altered by heat denaturation of proteins. Pasteurisation specifications vary with temperature, time and pH. The heat destruction of different serotypes of salmonellae at 60°C was reported to be greater in egg white (pH 9.0) than in yolk (pH 6.0) and whole egg (pH 7.6).

Hence the heat required for yolk pasteurization is greater than that required for the white. According to the Food Safety and Inspection Service (FSIS) of the United States Department of Agriculture (USDA) regulations whole egg is to be pasteurized at a minimum temperature of 60℃ for 3.5 min, while egg white and egg yolk must be brought to 57.5°C and 61.1°C, respectively, for at least 2.5 minutes to ensure egg safety against *Salmonella* and other food-borne pathogens (FSIS-USDA 2006; Zeilder, 2002). Pasteurisation systems include batch pasteurisation, high temperature short time systems and ultra-heat treatment (70°C for 1.5 sec).

2.3.6 Pasteurisation of shell eggs

Although there are several methods of preservation and sanitation of shell eggs such as washing, rapid chilling, UV - irradiation and ultrasonic treatment, they do not destroy *Salmonella* which may pose a serious health hazard to humans.

Pasteurisation techniques used for liquid eggs is reported to be unsuitable for in-shell eggs due to heat denaturation. Hou et al. (1996) developed a process of pasteurization of shell eggs using water bath and hot-air-oven heating systems. A combination of the two methods (water-bath heating at 57°C for 25 min followed by hot-air heating at 55°C for 60 min) produced 7 log reductions in *S. enteritidis* ATCC 13076 in shell eggs. Examination of lysozyme activity and other physical properties of egg white upon heating indicated that the overall functionality of pasteurized shell eggs is acceptable under the heating conditions defined in this study.

2.3.7 Sterilization of Eggs

Sterilization refers to any process that effectively kills or eliminates transmissible biological agents (such as fungi, bacteria, viruses, spore forms, etc.) from a surface, equipment, article of food or medication, or biological culture medium. Sterilization can be achieved through application of heat, chemicals, irradiation, high pressure or filtration (Pflug et al., 2001).

Thermal sterilization parameters for egg products include treatment with moist heat at 121°C for 15 to 20 minutes or at 115°C for 35 minutes or with dry heat at 160°C to 170°C for 1 to 2 hours (Berkowitz et al, 1984). But eventually this results in well cooked eggs. Therefore there is no effective thermal sterilization technique available for raw eggs.

2.3.8 Pasteurization *vs.* Sterilization

Pasteurization is a heat treatment that has been used extensively to inactivate food-borne pathogens. Unlike sterilization, pasteurization is not intended to kill all the micro-organisms in the food or liquid. Instead, pasteurization aims to destroy/inactivate all the viable pathogens, so they are unlikely to cause disease (assuming that the pasteurized product is refrigerated and consumed before its expiration date). Also commercial-scale sterilization of food is not common because it adversely affects the taste and quality of the product. It has been used successfully with liquid eggs but its utilization for in-shell eggs has been quite a challenge due to the geometry of the shell egg and the heat sensitivity of the egg proteins.

2.4 Effect of thermal treatments on eggs

Different temperatures have specific effects on different components of the eggs. Among these components, proteins which make up about 15% of the eggs are the most heat sensitive.

2.4.1 Proteins and peptides

Proteins are large biological macromolecules made of amino acids arranged in a linear chain and joined together by peptide bonds between the carboxyl and amino groups of adjacent amino acid residues. This sequence of amino acids in a chain is said to be the primary structure of the proteins (Walsh, 2002).

A peptide bond is a chemical bond formed between two molecules when the carboxyl group of one molecule reacts with the amino group of the other molecule, thereby releasing a molecule of water (H_2O). This is a dehydration synthesis reaction (also known as a condensation reaction), and usually occurs between amino acids. The resulting CO-NH bond is called a peptide bond, and the resulting molecule is an amide (Maton et al., 1993).

2.4.2 Protein composition of egg white and egg yolk

The egg white is approximately two-thirds of the egg's total weight outside its shell, with 90% of that weight coming from water. The remaining weight of the egg white comes from protein, trace minerals, fatty material, vitamins, and glucose. A U.S. large egg's white weighs 38 g and contains 4.7 g of protein, 0.3 g of carbohydrates and 62 mg of sodium (USDA, 2004). Egg white contains approximately 40 different proteins, of which the most abundant, on a weight basis, are ovalbumin 64%, ovotransferrin 12%, ovomucoid 11%, globulins 8%, lysozyme 3.5%, ovomucin 1.5% and avidin 0.06% (Lokhande et al., 1996).

Egg yolk proteins consist of glycoproteins, phosphoglycoproteins, lipoproteins and phosphoglycolipoproteins in almost equal quantities with some ovotransferrin and ovomucin (Lokhande et al., 1996; Kilara et al., 1986).

2.4.3 General structure of proteins

Proteins are an important class of biological macromolecules present in all biological organisms, made up of such elements as carbon, hydrogen, nitrogen, phosphorus, oxygen, and sulphur. They are polymers of amino acids. For biological functioning, proteins fold into one or more specific spatial conformations through hydrogen bonding, ionic interactions, Van der Waals forces and hydrophobic packing. It is necessary to study the three dimensional structure of proteins to understand their functions at molecular level.

Protein molecules have primary, secondary and tertiary structures. The primary structure consists of a sequence of amino acids held together by covalent peptide bonds. The secondary structure is made of alpha-helices and beta-pleated sheets and adopts a random coil configuration.

The tertiary structure is mainly due to the:

1. Covalent bonds between amino acid side chains (such as disulfide bridges between cysteine groups).
2. Non-covalent bonds between polar amino acid side chains (and the surrounding solvent).
3. Van der Waals interactions between non-polar amino acid side chains.

2.4.4 Heat sensitivity of egg proteins

Proteins are highly heat sensitive components of the egg. The functional properties like whipability, foamability, foam stability etc. which make the egg an important ingredient in various food products are severely affected by high temperatures. Also, experimentally it is found that the egg yolk needs to be heated to a higher temperature than the albumen to achieve pasteurisation. This is possible by conventional heating only if the yolk and albumen are separated (i.e. only if the shell is broken), as the yolk is concentric within the albumen in a shell egg.

The existing methods of pasteurizing shell eggs using hot water and/or hot air results in overheating of the albumen and produces partially cooked eggs along the shell membrane (Hank et al. 2001).

2.4.5 Protein denaturation

Protein denaturation is a physical change in which proteins lose their structure. Brought about by heat, UV rays, agitation, strong alkalis or acids, inorganic salts, organic solvents, it results in changes in solubility, loss of crystallizability, viscosity, coagulation and a host of other functional properties (Walsh, 2002).

2.4.5.1 Thermal denaturation of proteins

During thermal denaturation of proteins, the tertiary structure is first altered and then the secondary structure. The primary structure is not disrupted by thermal denaturation. Denaturation of the tertiary structure involves the disruption of the covalent and non-covalent bonds along with the Van der Waals interactions.

Comparatively, when secondary structure denaturation occurs, proteins lose all regular repeating patterns such as alpha-helices and beta-pleated sheets, and adopt a random coil configuration.

Heat can disrupt hydrogen bonds and non-polar hydrophobic interactions by increasing the kinetic energy that causes the molecules to vibrate, thus breaking the bonds. The proteins in eggs denature and coagulate during cooking, thus making it easier for enzymes to digest them in the human body(Virtual Chembook, 2003).

2.4.5.2 Role of water in thermal denaturation of proteins

Water is essential for the correct folding of proteins and the maintenance of its structure. The free energy change on folding or unfolding is due to the combined effects of both protein folding/unfolding and hydration changes. These compensate to such a large extent that the free energy of stability of a typical protein is only 40-90 kJ mol^{-1} (equivalent to a few hydrogen bonds), whereas the enthalpy change (temperature $\times$ entropy change) may exceed 500 kJ mol^{-1}. Both enthalpic and entropic contributions to this free energy, change with temperature and hence give rise to heat denaturation. Protein unfolding at low temperatures is accompanied by a decrease in entropy. Overall, protein stability depends on the balance between these enthalpic and entropic changes (Chaplin, 2008).

2.4.5 Mechanism of protein denaturation

Eggs have the best quality protein with the amino acid pattern almost matching the human requirement for essential amino acids (FAO protein value=100). A study by Van der Plancken (2006) showed that heating of egg white solutions in the temperature range of 50-85°C resulted in significant unfolding of the proteins, as evidenced by an exposure of hydrophobic groups and sulfhydryl (SH) groups previously buried in the protein core, resulting in a greater sensitivity to proteases. The decrease in denaturation enthalpy indicated a loss of the ordered three-dimensional protein structure. Depending on the pH during heat treatment, these changes can result in a drastic loss in protein solubility. This, in turn, leads to the formation of turbid protein suspensions after prolonged heating at elevated temperature, due to hydrophobic interactions and the formation of intermolecular disulfide (SS) bonds through SH/SS exchange reactions and SH oxidation.

Egg albumen is the egg protein most vulnerable to denaturation. Typically it is a translucent fluid that coagulates to a viscous thick white mass upon heating. This process is dependent on temperature, moisture of the albumen, presence of salts and the pH of the medium. Normal egg albumen coagulates at 80℃ and the heat sensitive ovotransferrin at 62-65℃. At the pH of 8.75 heating of albumen to 58℃ gave a better cake volume than unheated albumen, whereas between 77°C and 100℃ the egg white gels were firmer. This is due to the formation of sulfhydryl-disulphide interchange reactions (Li-Chan et al, 1995). It has been reported by Van der Plancken (2006) that egg white pasteurization at 60˚C for 3-5 min required the addition of additives like 0.2 M Tris–HCl to overcome small changes in functional properties.

2.5 Effect of heating on protein conformation and digestibility

Digestibility of egg protein is above 90%, that is, more than 90% of the egg protein is absorbed as amino acids for new protein synthesis and replacement of lost protein by human metabolic activity. Cooked egg protein is more digestible than raw egg protein (cooked egg protein digestibility is 90.9±0.8%, whereas the raw egg protein digestibility is only 51.3±9.8%). The biological value (a value that measures rate at which the protein in food supports growth) of egg protein is 94%. Eggs and milk have the highest biological value and provide more amino acids for growth and tissue maintenance than meat (ENC, 2004).

Evenepoel et al. (1998) showed that after ingestion of 25 g of raw egg protein, almost 50% remained unabsorbed in humans after 24 h. The higher digestibility of cooked egg protein could be due to structural changes in the protein molecule induced by heating, thereby enabling digestive enzymes to gain better access to the peptide bonds. It has been suggested that the reduced digestibility of raw egg white was partially related to the presence of trypsin inhibitors in raw egg white.

Browning in foods is a consequence of heating at very high temperatures like frying, baking and drying. During heating, certain amino acids in foods like lysine, by reaction of its epsilon amino group with reducing sugars like glucose, result in non-enzymatic browning termed as Maillard reaction. Severe heat treatments can bring about significant losses in the essential amino acid lysine, thus reducing the protein's nutritive value (Valle-Riestra et al., 1970). High temperature (140-160^{0}C) treatment of milk gave rise to denaturation of whey proteins, formation of large protein particles and gelation during storage. Phosphorus compounds were reported to split from proteins forming complexes with casein and lactoglobulins (Wilson, 1971).

The effect of a low-temperature, extended-time, in-shell pasteurization process on the protein quality of egg albumen was evaluated by Hank et al. 2001. Ten dozen fresh chicken eggs were pasteurized in a hot-air oven at 55°C for 180 min. They were refrigerated and evaluated after 0, 7, 14, 28, 42, and 56 days following pasteurization. There were no significant differences in total or soluble protein over the experimental period for the pasteurized or unpasteurized albumen. There were no significant differences over the experimental period in the digestibility of the samples. Free amino acids and discriminant-computed protein efficiency ratio (DC-PER) also did not differ between the pasteurized and unpasteurized albumens over the experimental period. The in-shell pasteurization process used had no significant effect on the protein quality of albumen (Hank et al., 2001).

2.6 Summary of effects of heat treatments on the conformation and digestibility of proteins

In light of all the discussion above, egg proteins, especially albumen, are vulnerable to heat in any form. At pasteurization temperatures of about 58-62℃, the changes that can bring about significant denaturation are likely to be very minimal, which would be further minimized by the shorter heating time when microwave heating is used. Microwave heating is favoured for its quick heating and good penetration capacity, such that it is suitable for High Temperature Short Time (HTST) treatments (Ohlsson, 2000). Non-thermal denaturation with MW could occur due to ionization, but the effect is very minimal and highly reversible (Bohr & Bohr, 2000).

Digestibility of proteins in pasteurized egg is expected to be more or less the same as raw egg due to a minimal change in the proteins, irrespective of the method used. Comparatively, during sterilization, which requires much more stringent heat treatments, the protein is denatured, bringing about structural disruption and change in

functional properties. At such high temperatures loss of specific amino acids may also occur through the Maillard reaction, a process which is less likely in MW heating. Non-thermal methods like irradiation are employed for sterilization where denaturation occurs purely due to ionization, which is partly reversible. Denaturation of proteins due to heat unfolds the protein structure making it more accessible to the proteolytic enzymes for digestion. Thus sterilization increases the digestibility of proteins.

2.7 Microwave interactions with food constituents in conjunction with eggs constituents

As the interaction of microwaves with the substance exposed to it is at the molecular level, the thermal and non-thermal effects produced are considerably depending on the molecular structure and intermolecular organisation of the material. As food materials contain a high concentration of organic matter, the interaction of food constituents with microwaves can be categorized based on their chemical classification as carbohydrates, lipids and proteins. In the context of an egg, the total amount of carbohydrates is less than 1% by mass (Table 2.1) and therefore its microwave interactions are negligible compared to the lipids and proteins.

2.7.1 Lipids

Lipids may be broadly defined as a group of naturally-occurring hydrophobic or amphiphilic small molecules that originate entirely or in part from two distinct types of biochemical subunits or "building blocks": ketoacyl and isoprene groups. This includes fats, oils, waxes, cholesterol, sterols, fat-soluble vitamins (such as vitamins A, D, E and K), monoglycerides, diglycerides, phospholipids, and others. The major biological functions of lipids are energy storage, acting as structural components of cell membranes, providing thermal insulation to the body and participating as important signaling molecules (King, 1996).

2.7.1.1 Dielectric Properties of lipids and their microwave interaction

The hydrophobic portion of lipids does not interact significantly with microwaves however, the ionizable carboxyl groups of fatty acids show limited interaction. The extent of saturation of the fatty acids has no effect on the dielectric response of the lipid molecules (Mudgett and Westphal, 1989). Therefore, the dielectric properties of fats and oils are very low. The dielectric constant (ε') of most lipids is between 2.5 and 3 and their dielectric loss factor (ε") is in the range of 0.1- 0.2. The effect of fat on dielectric properties of food systems is mainly due to their dilution effect in the system. An increase in fat content reduces the free water content in the system, which reduces the dielectric properties on the whole (Datta et al. 2005).

Loss factors of lipids at ambient temperature are greater in more liquid forms such as corn oil and cottonseed oil as compared to lard and tallow fats which are solids at that temperature. The effect of changing temperature on the dielectric properties of lipids is not significant (Pace et al., 1968). In many cases in their naturally occurring form, lipids exist as an emulsion in water (or) at least the fat containing cells are surrounded by lots of free water. Under these conditions lipids apparently seem to heat up faster during microwave heating, but this relates to the fact that the specific heat capacity of lipids is much lower than that of water and hence they require less microwave energy to heat up (Dev et al. 2008) and also energy is absorbed from the surrounding water molecules, which efficiently convert electromagnetic energy into heat energy.

2.7.1.2 Refractive index and penetration depth of microwaves in lipids

Refractive Index and penetration depth are key parameters in the design of microwave thermal applicators, especially when heating uniformity is the major focus of the design.

Based on the above cited dielectric properties, the refractive index of most lipids at a microwave frequency of 2450 MHz is around 1.45 –1.75 with an angle of refraction ranging from 35 – 45° for normally incident microwaves. The penetration depth of microwaves for most lipids is in the range of 15 – 30 cm in any given direction of propagation (Duck, 1990).

2.7.2 Microwave interaction with Proteins

Being relatively microwave-inert, proteins do not interact significantly with microwaves. Some portions of proteins are water soluble and some largely insoluble (Datta et al 2005). Nonetheless, proteins mainly interact with microwaves on three levels conforming to their primary, secondary and tertiary structures. The details of these structures were discussed in section 2.4 of this chapter.

2.7.2.1 Dielectric properties of proteins and their microwave interaction

Proteins have ionizable surface regions that may bind water or salts to give rise to zeta potential and double-layer effects associated with free surface charges (Mudgett and Westphal, 1989). These have a small but significant effect on dielectric behaviour at microwave frequencies. The solvated or hydrated form of protein, protein hydrolysates, and polypeptides are much more microwave-reactive than native proteins (Datta et al 2005). The dielectric properties of proteins depend on their side chains, which can be non-polar (in decreasing order: alanine, glycine, leucine, isoleucine, methionine, phenylalanine and valine), or polar (in decreasing order: thyrosine, tryptophan, serine, threonine, proline, lysine, arginine, aspartic acid, aspergine, glutamic acid, glutamine, cysteine and histidine (Shukla and Anantheswaran, 2001).

Free amino acids are more dielectrically reactive (Pething, 1979). Free amino acids contribute to an increase in dielectric loss factor. Since protein dipole moments

are a function of their amino acids and the pH of the medium, the dielectric properties and microwave reactivity of cereal, legume, milk, meat, and fish proteins are expected to be different. The bound water on the proteins also affects their dielectric properties (Shukla and Anantheswaran, 2001).

The dielectric activity of proteins can be assigned to four major reasons, given below in decreasing order of significance: (Datta et al. 2005)

1. Charge effects of ionization of carboxyl, sulfhydryls, and amines.
2. Hydrogen and ion binding as affected by pH
3. Net charges on dissolved proteins
4. Relaxation and conductive effects

Such activities are important for hydrolyzed proteins and free amino acids. Since most proteins are consumed in a cooked form, it is important to determine dielectric properties during denaturation of proteins to understand the microwave heating of these foods (Shukla and Anantheswaran, 2001).

2.7.2.2 Protein denaturation and its interactions with microwaves

Microwave irradiation can affect the kinetics of the folding process of some globular proteins, particularly beta-lactoglobulin. At low temperatures the folding from the cold-denatured phase of the protein is enhanced by microwave energy, while at a higher temperature the denaturation of the protein from its folded state is enhanced. In the latter case, a negative temperature gradient is needed for the denaturation process, suggesting that the effects of the microwaves are non-thermal. This supports the view that coherent topological excitations can exist in proteins (Bohr & Bohr, 2000).

As the protein structure is disrupted, the asymmetry of the charge distribution will increase along with dipole moment, polarization thus affecting dielectric properties.

Water is either released or bound to the protein. During denaturation of egg white ovalbumin the dielectric constant decreased due to binding of water, while low water mobility caused the loss factor to remain unchanged. In egg yolk lipovitelin denaturation brought about decrease in water dipole mobility, that resulted in a decrease in the dielectric constant and loss factor (Bircan & Barringer, 2002). Microwave cooking/heating has been reported to reduce cooking time and help retain nutrients and inactivate antinutritional components like polyphenols and trypsin inhibitors in foods (Petres et al, 1990; Laurena et al, 1987; Alajaji et al, 2006).

Protein denaturation is defined as the physical change of the protein molecule due to heat, ultraviolet (UV), or agitation, which results in a reduction in protein solubility, a loss of crystallizability, and an increase in solution viscosity (McWilliams, 1989). During denaturation of proteins, since the protein structure is changed, the asymmetry of the charge distribution will increase and the free water in the system will change during denaturation. As a result, a large dipole moment and polarization will affect the dielectric properties of foods. Moisture is either bound by the protein molecule or released to the system during denaturation. Various studies show that the dielectric properties can be used to understand protein denaturation (Ahmed et al., 2007).

Usually, the dielectric loss factor of proteins changes by exhibiting a peak during denaturation, due to the binding of water and ions by proteins. Comparatively, the dielectric constant did not change during denaturation, but rather decreased with increase in temperature (Bircan et al., 2001, Bircan and Barringer, 2002a, b).

The loss factor of egg yolk protein increased and then decreased with temperature, exhibiting a peak during denaturation (Bircan and Barringer, 2002a). The reduction of the loss factor after denaturation was due to the binding of water and decrease in mobility of ions. Comparatively, the loss factor of the meat protein

actomyosin increased during protein denaturation due to the release of water during denaturation (Bircan and Barringer, 2002b).

The dielectric constant and loss factor of a heated gluten–starch mixture were found to be less than those of the unheated mixture (Umbach et al., 1992). As the amount of gluten protein in the system increased, the dielectric constant decreased, but the loss factor remained constant. The interaction of gluten with microwaves has been known to have an adverse effect on the texture of microwave-baked breads (Yin and Walker, 1995). Microwave-baked breads containing a small amount of gluten were softer than those containing a larger amount of gluten (Ozmutlu et al., 2001).

2.7.2.3 Non-thermal effects of microwaves on protein denaturation

Microwave radiation also has non-thermal effects that can enhance the kinetics of the folding and denaturation processes. Bohr and Bohr (2000) observed effects of microwaves on β-lactoglobulin and found that microwaves had the ability to change the kinetics of folding and denaturation. This supports the view that coherent topological excitations can exist in proteins.

The amino acid proline differs from the other amino acids in its basic structure as it contains an unusual ring linked to the N-end amine group, which forces the CO–NH amide moiety into a fixed conformation. This leads proline to shift from the L-proline to D-proline isomeric form in a microwave environment (Lebuc et al., 1989).

2.7.2.4 Refractive index and penetration depth of microwaves in proteins

The refractive index and penetration depth of microwaves at 2450 MHz for proteins is highly variable, and depends on the type of protein and its primary, secondary and tertiary structures, along with the above mentioned factors.

For 2450 MHz microwaves, the refractive index of ovalbumin, a major protein in egg white is around 1.5 (Barer & Tkaczyk, 1954) with an angle of refraction of roughly 40° for normally incident microwaves.

The penetration depth of microwaves for ovalbumin is around 9.5 cm in any direction. There is a potential for birefringence based on the type and isomeric form of the amino acids present in the protein and the anisotropic behaviour of these components.

2.7.3 Microwave interaction with a bi-layer of lipid and protein

In light of the interaction of microwaves with lipids and proteins, it is clear that both lipids and proteins in their pure form are relatively much less reactive to microwaves than water. This results in much lower losses when microwaves traverse through these materials, compared to water. Therefore a considerably thick bi-layer of lipid and protein (15 – 20 cm) can be heated up effectively by microwaves, but it takes relatively much longer period of time to reach a particular temperature for a given power density. The dielectric properties of the type of lipid and protein as well as the amount of free fatty acids and free amino acids present in the bi-layer would determine the extent of microwave interaction with both the layers. However, the extent of heating (i.e. heat generated) and heating rate in the second layer could be significantly lower than that of the first layer depending on the thickness of the first layer in the bi-layer.

In case of eggs, such bi-layers are found in the yolk, with enormous amount of moisture (55%) surrounding it. This results in a significant reduction in the penetration depth to just 2-3 cm due to the high dielectric loss characteristic of the water molecules.

2.7.3.1 Penetration depth calculation for a lipid – protein bi-layer

From Equation (2.1) one can obtain the penetration depth (D_p) of microwave in a given direction for a given material (Meda et al. 2005).

$$D_p = \frac{1}{2\pi f}\sqrt{\frac{2}{\mu_0 \varepsilon_0 \varepsilon'}\left(\sqrt{\left(1+\left(\frac{\varepsilon''}{\varepsilon'}\right)^2\right)}-1\right)} \approx \frac{\lambda_0\sqrt{\varepsilon'}}{2\pi\varepsilon''} \qquad (2.1)$$

Where f is the frequency (Hz), ε' is the dielectric constant, ε'' is the dielectric loss factor, ε_0 is the absolute permittivity of free space (Fm^{-1}), μ_0 is the permeability of free space (Wm^{-1}) and λ_0 is the wavelength in free space.

It is important to note that the penetration depth depends on $\sqrt{\varepsilon'}$ which represents the real part of the complex refractive index of the material and is close to the actual refractive index for a low loss material like glass. Therefore the calculation of penetration depth accounts for the refraction happening within the material in a given direction. Thus the calculation of penetration depth is uni-dimensional, whereas microwave heating is volumetric (i.e. three dimensional).

In the case of a lipid-protein bi-layer, the incident angle of the microwave radiation is altered by the refraction occurring in the lipid layer and this would happen in all the three dimensions, thereby making the calculation of penetration depth more complex.

A general approximation for the calculation of effective penetration depth (ED_p) for a bi-layer of low loss material in a given direction would be to do a linear weighted averaging as given by Equation (2.2): (Halbritter, 1992)

$$ED_p = \frac{\mu_L Dp_L + \mu_p Dp_p}{\mu_L + \mu_p} \qquad (2.2)$$

where, μ_L and μ_p are the thickness of the lipid and protein layers respectively, and Dp_L and Dp_p are the individually calculated penetration depths for the lipid and protein, respectively. However, for practical purposes such approximations are not commonly used.

The most commonly used method for solving the microwave power distribution in three dimensions is to solve the Maxwell equations (equations (2.3), (2.4) & (2.5)) by using finite difference or finite element approximation.

$$\frac{\partial E_x}{\partial t} = \frac{1}{\varepsilon_0 \varepsilon'}\left(\frac{\partial H_z}{\partial y} - \frac{\partial H_y}{\partial z}\right) - \frac{2\pi f \varepsilon''}{\varepsilon'} E_x \qquad (2.3)$$

$$\frac{\partial E_y}{\partial t} = \frac{1}{\varepsilon_0 \varepsilon'}\left(\frac{\partial H_x}{\partial z} - \frac{\partial H_z}{\partial x}\right) - \frac{2\pi f \varepsilon''}{\varepsilon'} E_y \qquad (2.4)$$

$$\frac{\partial E_z}{\partial t} = \frac{1}{\varepsilon_0 \varepsilon'}\left(\frac{\partial H_y}{\partial x} - \frac{\partial H_x}{\partial y}\right) - \frac{2\pi f \varepsilon''}{\varepsilon'} E_z \qquad (2.5)$$

Under both finite difference and finite element methods the effect of polarization and thereby refraction is effectively taken into account while solving for the distribution of the electric field in different coordinates, which corresponds to the heat generated depending on the loss factor of the material.

2.7.4 Microwave interaction and penetration depth calculation for a homogenous mixture of lipids and proteins.

Based on the description about the interaction of microwaves with lipids and proteins, it is clearly enunciated that both of these compounds do not interact strongly with microwaves, which is evident from their poor dielectric properties. Therefore a considerably thick bi-layer of lipid and protein can be heated up effectively by

microwaves. Depending on the dielectric properties of the type of lipid and protein and also the amount of free fatty acids and free amino acids present in the mixture would determine the extent of microwave interaction. Also based on the proportions of the lipids to proteins, the microwave interactions could vary significantly with regards to microwave interactions with the individual compounds.

A weighted average of the dielectric properties would give a good approximation of the values of the same for a homogenous mixture. If *x* is the mass fraction of lipids and *y* is the mass fraction of proteins in the homogenous mixture, then the dielectric properties can be given by Equations (2.6) and (2.7):

$$\varepsilon' = x\varepsilon'_L + y\varepsilon'_p \quad (2.6)$$

$$\varepsilon'' = x\varepsilon''_L + y\varepsilon''_p \quad (2.7)$$

Where the dielectric properties of lipid and protein are subscripted as L and p, respectively. Also $x + y$ must be equal to 1. This means all the components of the mixture must be taken into account. This can be used in the calculation of effective penetration depth of microwaves in a homogenous mixture.

2.7.5 Microwave interaction with egg yolk (a lipid rich emulsion and a homogenous mixture of proteins and lipids with water).

The egg yolk is an emulsion that makes up about 33% of the liquid weight of the egg. It contains 34% fat (by weight) and accounts for approximately 60 calories, compared to 20 calories for the egg white, which weighs twice as much as the yolk. The yolk also contains more or less the same amount of protein (around 15% by weight) as the egg white.

Egg yolk being rich in lipids has dielectric properties in the frequency range of 200 MHz – 10 GHz and in the temperature range of 0-60℃ is given by Equations (2.8) and (2.9) as reported by Dev et al (2008).

$$\varepsilon' = 50.085 - 0.13\,T - 1.72\,f \qquad (2.8)$$

$$\varepsilon'' = 13.55 - 0.11\,T + 0.65\,f \qquad (2.9)$$

where,

T is the temperature (°C), and

F is the frequency (GHz)

These values are much higher than those for pure lipids and proteins since egg yolk is an emulsion containing nearly 50% water (NRC, 1976; Lokhande et al. 1996) and much of this hike in dielectric properties is attributed to the presence of water.

At a microwave frequency of 2.45 GHz and a temperature of 20℃, from the Equations (2.8) and (2.9), the dielectric properties of the yolk would be:

$$\varepsilon' = 43.3 \text{ and } \varepsilon'' = 12.9$$

By doing a weighted average of the dielectric properties as per equations (2.6) and (2.7) for a yolk composition of 51% water, 15% proteins and 34% fat, we would get:

$$\varepsilon' = 45.3 \text{ and } \varepsilon'' = 11.7$$

The above values bear a less than 10% error in comparison to model-predicted values. Thus a weighted average approach gives a good approximation for the calculation of the effective dielectric properties and thereby the effective penetration depth of microwaves for a homogenous mixture.

2.8 Conventional heating Vs. Microwave application for heat treatment of eggs

Presently most commercially available pasteurized eggs are pasteurized using conventional heating methods and by separating the yolk and egg white before processing. But breaking and repacking them aseptically involves huge additional costs. Therefore in-shell pasteurization has gained a great commercial importance in recent years.

The current technique for in-shell pasteurization of egg involves heating the eggs in a water bath at 60 ºC for about 20-25 minutes, depending on the size of the eggs. This leads to the overheating of the egg white proteins (i.e the egg white gets heated up more than the yolk, which is against the recommendations) resulting in denaturation and coagulation (Hou et al. 1996). This greatly affects the functional properties of the eggs. Therefore a process that can heat the shell eggs from inside would be an excellent alternative to solve this problem.

Microwaves have the ability to generate heat from within the substance that is exposed to them. Theoretical mathematical studies have shown that even though albumen exhibits better dielectric properties than yolk, the egg's curvature has a focusing effect which leads to a suitable power distribution (Datta et al. 2005). Hence the shell egg appears ideally suited for pasteurization in a microwave environment (Fleischman 2004; Rehkopf 2005; Dev et al. 2008).

Eggs are never sterilized in shell as the moist heating involves high pressures in order to raise the boiling point of water and the shell cannot withstand this pressure. Sterilization of egg components results in a completely cooked product.

In a microwave environment, due to extensive steam build up and high risk of explosion at this temperature, in-shell eggs are never subjected to microwave treatment

for sterilization. Also applying microwaves in a high pressure setup involves a complex engineering design, whereas microwaves have very good potential for the pasteurization of in-shell eggs without resorting to high pressures.

2.9 Microwaves and Their Properties

Microwaves are very short waves of electromagnetic energy that travel at the speed of light. They have all the basic properties of any electromagnetic radiation. They have excellent penetrating power, which is inversely proportional to their frequency. Figure 2.2 shows the position of the microwaves in the electromagnetic spectrum. Microwave spectrum has wavelengths ranging from millimetres to centimetres. Hence, a portion of the microwaves spectrum is also termed as centimetre waves (Pozar 2005).

Microwaves are electromagnetic radiation with frequencies approximately in the range of 300 MHz and 30 GHz, located between the infrared and radio frequencies in the electromagnetic spectrum (Fig. 2.2).

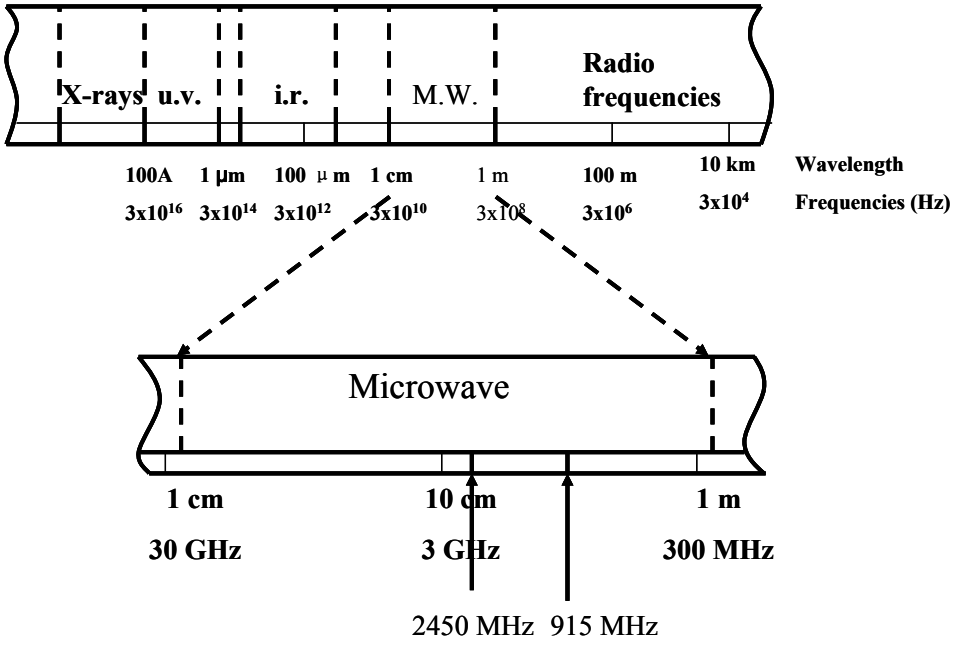

Figure 2.2. Locations of microwaves on the electromagnetic spectrum

The propagation of one complete cycle in the waveform of the microwaves or any electromagnetic radiation is shown in Figure 2.3. Thus microwaves, being an electromagnetic radiation, create an alternating electric field and an alternating magnetic field perpendicular to each other. This property is exploited in the thermal applications of microwaves.

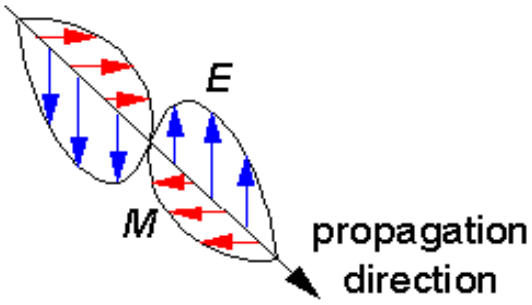

Figure 2.3 Microwave propagation

(E – Electric field, M- Magnetic field)

As microwaves are electromagnetic radiation similar to visible light, they follow all the basic laws of physics like reflection, refraction, interference, diffraction and polarization. The energy level of microwaves is usually measured in terms of power density ($W.g^{-1}$), corresponds to the dipolar rotational energy level of polar molecules. Therefore the interaction of microwave energy with matter is through the dielectric rotation of the molecules. The intermolecular friction between the fast rotating molecules causes a rapid volumetric heating. This is a unique characteristic that differentiates microwave heating from the conventional heating methods. Only those molecules that can couple with the microwave field can be heated with microwave energy.

Electrically, the complex relative permittivity (ε^*) is used to describe the interaction of microwaves and matter. The complex relative permittivity (ε^*) can be expressed as:

$$\varepsilon^* = \varepsilon' - j\varepsilon'' \quad (2.10)$$

where ε' is the dielectric constant and ε'' the loss factor. The dielectric constant describes the capability of molecules to be polarized by the electric field and the loss factor measures the efficiency of molecules to convert microwave energy into heat (Mingos and Baghurst, 1991). The complex refractive index n^* of a given material for a given wavelength is given $\sqrt{\varepsilon^*}$.

The following equation (2.11) is used to calculate the energy absorption:

$$P_v = 2\,\pi\, f\, \varepsilon_0\, \varepsilon''|\, E\,|^2 \quad (2.11)$$

where, P_v is the power absorbed per unit volume ($W.m^{-3}$)

f is the frequency (Hz)

ε_o is the absolute permittivity of a vacuum ($F.m^{-1}$)

|E| is the absolute value of the electric field strength inside the load ($V.m^{-1}$).

As it can be seen from equation (2.11) the power dissipated in a certain volume is proportional to the loss factor of the matter. High moisture food materials (Moisture content > 80 %), like egg white, which constitutes nearly two third of the total mass of an egg, usually have both their dielectric constant and dielectric loss factor close to that of pure water, and therefore have similar heating characteristics.

In the hen's egg, the egg white is the chief reservoir of water and it is the most alkaline of all natural liquids. The yolk contains the main reserve of food substances, required for the development of embryo. (Lokhande, Arbad, Landge, & Mehrotra, 1996). Theoretical mathematical studies have shown that even though albumen exhibits better dielectric properties than yolk, the egg's curvature has a focusing effect which leads to a suitable power distribution (Datta, Sumnu, & Raghavan, 2005).

2.9.1 Penetration Depth

An important concept associated with microwave-matter interaction is the penetration depth, which is defined as the depth at which the intensity of the radiation inside the material falls to $\frac{1}{e}$ (about 36.79 %) of the original value at the surface of the material.

When electromagnetic (EM) radiation is incident on the surface of a material, part of it is reflected and part transmitted into the material. This EM wave interacts with the atoms and electrons inside the material. Depending on the nature of the material, the EM wave might travel very far into the material, or on the other hand it might die out very quickly. For a given material, penetration depth can vary for different wavelengths of EM wave, and usually, is not a fixed constant.

Microwave heating exploits the dielectric behaviour of the substance exposed to it to generate heat from within the substance. But this direct heat generation occurs only up to a certain depth of the product from the surface, since, depending on the dielectric properties of the substance, there is an exponential decay of microwave energy as the waves penetrate into the product from the surface (Meda, Orsat, & Raghavan, 2005).

The penetration depth (D_p) is a function of dielectric constant and loss factor given by equation (2.1). Penetration depth is one of the restricting factors in the scale-up of microwave equipment design for a specific process. For high moisture substances, like that of the eggs, the penetration depth of microwaves at 2450 MHz is usually less than a couple of centimeters (Dev, Raghavan and Gariepy. 2008).

Microwaves undergo exponential decay inside the material through which it they are travelling. According to the Beer-Lambert law, the intensity of an EM wave inside a

material falls off exponentially from the surface of the material as shown in Equation (2.12) (Swami, 1982).

$$P = P_0 e^{-2\alpha d} \tag{2.12}$$

where, P_0 is the power at the surface (W),

d is the maximum distance measured from the surface, (m) and

α is the attenuation factor (dimensionless), $\alpha = \dfrac{\pi\sqrt{\varepsilon_0 \varepsilon_R \mu_0 \mu_R}\,\tan\delta}{\lambda_0}$

where $\tan\delta$ if the loss tangent and μ is the permeability of the material

2.9.2 Generation of Microwaves

The principal elements for both multi-mode and focused microwave devices are the following four major components:

(a) the microwave generator, usually called the ‘‘magnetron,’’ which produces the microwave energy;

(b) the waveguide, which is used to propagate the microwaves from the magnetron to the microwave cavity;

(c) the applicator, where the sample is placed; and,

(d) the circulator, which allows microwaves to pass only in the forward direction.

Microwaves are generated in a microwave oven by a high voltage system. The heart of this high voltage system is the magnetron tube. The magnetron is a diode-type electron tube which is used to produce the required frequency of microwave energy. A magnetic field imposed on the space between the *anode* (plate) and the *cathode* serves as the grid. While the external configurations of different magnetrons will vary, the basic internal structures are the same (Gallawa 1989). These include the *anode,* the *filament/cathode,* the *antenna,* and the *magnets.* Figure 2.4 shows a longitudinal and cross sectional diagram of a magnetron.

The magnetron's operation is based on the motion of electrons under the combined influence of electric and magnetic fields, (i.e.) electrons must flow from the cathode to the anode. There are two fundamental laws that govern this

1. When force is exerted by an electric field on an electron, it tends to move from a point of negative potential toward one of positive potential. Figure 2.5 A shows the uniform and direct movement of electrons in an electric field with no magnetic field present, from the negative cathode to the positive anode.

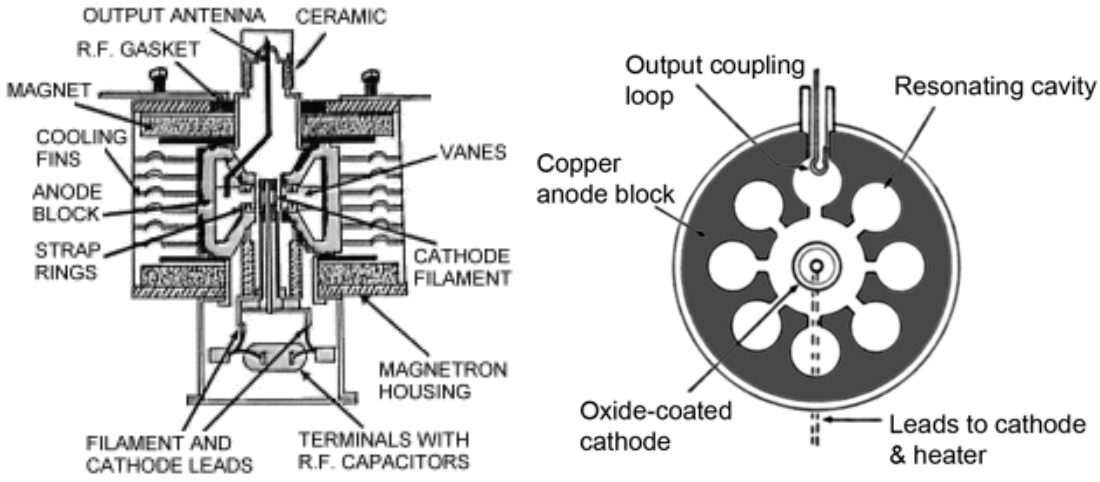

Figure 2.4 Longitudinal and cross sectional diagram of a resonant cavity magnetron (Modified from diagram in (Gallawa 1989) & (Morgan 1960))

2. When force is exerted on an electron by a magnetic field, which is at right angles to the electric field itself, and to the path of the electron, the direction of the force is such that the electron proceeds to the anode in a curve rather than a direct path (Figure 2.5 B and C).

Electrons, being negatively charged, are strongly repelled by other negative charges. So this floating cloud of electrons would be repelled away from a negatively charged cathode. The distance and velocity of their travel would increase with the

intensity of the applied negative charge. Momentum is thus provided by a high negative DC voltage, which is produced by means of the high-voltage transformer and the double action of the high-voltage diode and capacitor (Pozar 2005).

A high negative potential on the cathode puts a corresponding high positive potential on the anode. This makes the electrons blast off from the cathode. They accelerate towards the positive anode. This is when they encounter the powerful magnetic field of two permanent magnets. These are positioned so that their magnetic fields are applied parallel to the cathode. The effect of the magnetic fields tends to deflect the speeding electrons away from the anode. They curve to a path at almost right angles to their previous direction, resulting in an expanding circular orbit around the cathode, which eventually reaches the anode (Figure 2.5 D).

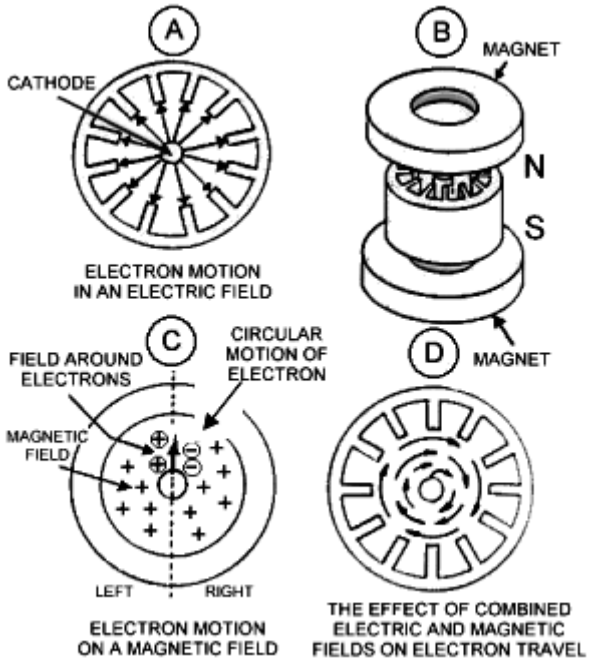

Figure 2.5 Working of a magnetron

(Modified from (Gallawa 1989))

2.9.3 Applications of Microwaves

Microwaves occupy a wide range in the electromagnetic spectrum. This frequency range is extensively used in RADAR transmission and telecommunications. Microwaves are good for transmitting information from one place to another because microwave energy can penetrate haze, light rain and snow, clouds, and smoke. Shorter microwaves are used in remote sensing. These microwaves are used for radar like the Doppler radar used in weather forecasts. Microwaves used for radar, are just a few centimeters long. Microwave applications are quite extensive and they are used in almost any form of wireless communication from military communication to personal communication and networking of computers and peripherals. Therefore regulations were made such that only limited frequencies could be used for industrial, scientific, and medicinal purpose (ISM frequencies) (Stuerga and Delmotte, 2002). The frequencies of 2450 MHz and 915 MHz are frequently employed in industrial uses. 2450 MHz is used for domestic microwave ovens and microwave-assisted extraction equipment.

Another important application of microwaves is its thermal application. Longer microwaves, those closer to 15 cm are the waves which heat the food in a microwave oven.

2.9.3.1 Major types of microwave applicators

Microwave energy - with a wavelength that is comparable with the dimensions of the installation - cannot simply be transported via standard conductors and discrete networks. Efficient power transmission is achieved with closed wave guides according to the principles of transmission lines. Wave guides are produced as metal pipes, mostly with a rectangular cross section.

The dimensions are dependent on the frequency. Wave guides can be both straight and curved. To keep the transmission losses to a minimum, metals that are

good conductors such as copper or aluminium are used. The inside surface must be smooth and clean. There are two basic designs in microwave installations:

- **Monomode applicators**: The product runs though a folded rectangular wave guide.
- **Multimode applicators**: a resonating space in which the product to be heated is placed

Figure 2.6 shows a schematic of both the monomode and multimode applicators

Single-Mode Cavity

In the single mode or monomode, or in other words a focused microwave cavity, the vessel is placed in the waveguide where focused microwaves are applied to the food material. Usually, focused systems of the open-vessel type cannot be pressurized.

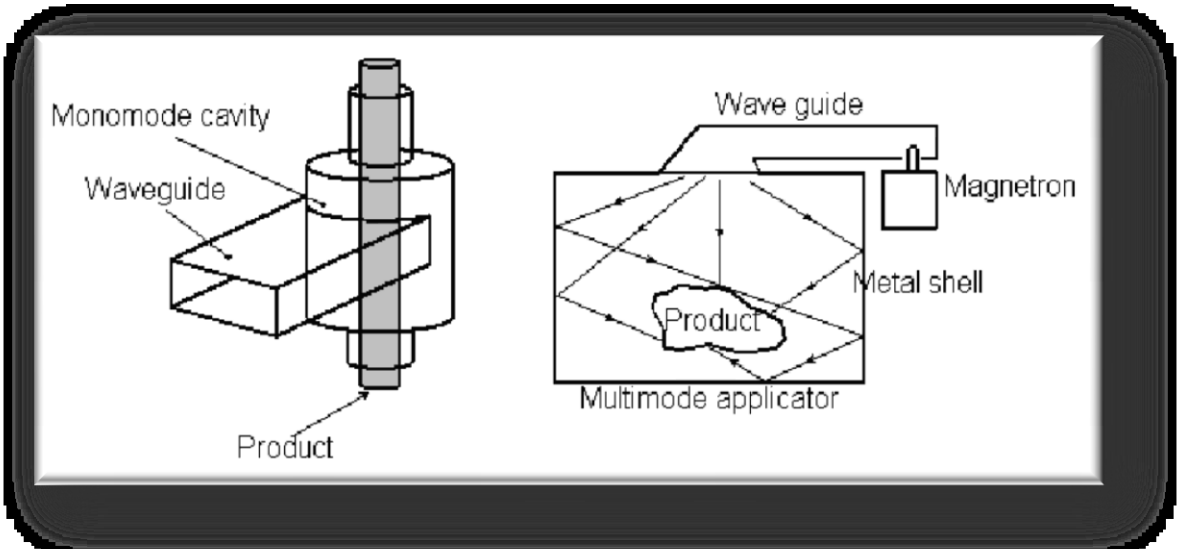

Figure 2.6 Monomode and Multimode microwave applicators.
(Callebaut, 2007)

With this method, a very high energy density can be obtained but the size of the sample is limited (Letellier et al. 1999). In focused systems, in which microwave radiation is focused on a restricted zone, the sample is subjected to a much stronger electrical field.

Focussed microwave systems have a single vessel placed directly in a microwave waveguide and that acts as the applicator. The bottom few inches of the vessel are directly exposed to the microwaves, whereas the upper region of the vessel remains cool. This results in an effective condensing mechanism inherent in the design. While vessels are open to the atmosphere, the refluxing action minimizes losses of solvent and some volatiles. Vessel openings have been designed to allow automated reagent addition and to restrict contamination from the atmosphere. (Luque-Garcia, 2003). Strictly adhering to the theory of resonant cavities is not required in designing monomode cavities, as we are not looking at resonating waves inside the cavity that is used to heat the material to be heated. Nevertheless, the waveguide itself must serve as a resonating cavity.

The latest advances in the use of microwaves in various fields of analytical chemistry include sample digestion for elemental analysis, solvent extraction, sample drying, moisture measurement, analyte adsorption and desorption, sample clean-up, chromogenic reactions, solid-phase retention, elution, distillation, microwave plasma atomic spectrometry and synthetic reactions. A vast majority of them employ focussed microwaves systems at a laboratory scale. A continuous liquid flow heating system can be designed with relatively little complexity using such a system. A focussed microwave system, properly tuned, can reach energy efficiencies as high as 90%.

As the dimensions of the waveguide are always limited by the frequency of the waves used, for microwave frequencies, the capacity of focussed microwave equipment is always limited and scaling up involves installing multiple units and heating in stages. Considering the large size it can reach, a multimode cavity type is preferred for the scaling-up of microwave equipment (Dai, 2006).

Multimode Cavity

A multimode cavity is the same in nature as that of a domestic microwave oven. Multi-mode systems are of the closed-vessel type and therefore can be pressurized. The large size in the cavity can provide more space for the extraction vessel and allows some new features, such as pressurized close-vessel extraction benefiting from the higher temperatures which can be achieved.

To the average consumer, the term "microwave" connotes a microwave oven, which is a multimode cavity. It is used in many households for heating food: industrial and medical applications also exist for microwave heating. As shown in Figure 2.6. a microwave oven is a relatively simple system consisting of a high-power source, a waveguide feed and the oven cavity. The source is generally a magnetron tube operating at 2.45 GHz, although 915 MHz is sometimes used when greater penetration is desired. Power output is usually between 500 and 1500 W. The oven cavity has metallic walls, and has large electrical conductivity. To reduce the effect of uneven heating caused by standing waves in the oven, a "mode stirrer," which is simply a metallic fan blade, is used to perturb the field distribution inside the oven. The food can also be rotated with a motorized platter.

In microwave heating, the inside of the material gets heated first. The process through which this occurs primarily involves the electrical conduction losses in materials with large loss tangents (Okress, 1968; Gardiol; 1984). An interesting fact is that the loss tangents of many foods decrease with increasing temperature so that microwave heating is to some extent self-regulating. The result is that microwave cooking generally gives faster and relatively more uniform heating of food than conventional cooking.

The efficiency of a microwave oven, when defined as the ratio of power converted to heat (in the food) to the power supplied to the oven, is generally around

50%. However, this is usually much greater than the cooking efficiency of a conventional oven. The most critical issue in the design of a microwave oven is safety. Since a very high power source is used, leakage levels must be very small to avoid exposing the user to harmful radiation. Thus the magnetron, feed waveguide and oven cavity must all be carefully shielded. The door of the oven requires particular attention; besides close mechanical tolerances, the joint around the door usually employs MW and RF absorbing material and a $\lambda/4$ choke flange to reduce leakage to an acceptable level. Almost all industrial microwave applications employ a multimode cavity as feasibility of scaling up the process is much higher while using a multimode cavity.

2.9.3.1 Thermal Application of Microwaves (Dielectric Heating)

The phenomenon of microwave heating of foods was discovered accidentally. In the late 1940s, a candy bar in the shirt pocket of an engineer softened considerably when the engineer stood in front of a microwave transmitter. It didn't take long for this phenomenon to be capitalized upon and in just a few years microwave ovens began to appear. Although microwave heating has been successfully applied at the industrial level in other fields, in food processing it has met with limited success.

A microwave oven uses microwave radiation, usually at a frequency of 2450 MHz (λ = 12.24 cm). These waves are passed through the food in order to heat it. Water, fat, and sugar molecules in the food absorb microwave energy in a process called dielectric heating.

Many molecules (such as those of water) are electric dipoles, meaning that they have a positive charge at one end and a negative charge at the other, and therefore rotate as they try to align themselves with the alternating electric field induced by the microwave beam. This molecular movement creates heat as the rotating molecules hit

other molecules and put them into motion. Figure 2.6 shows the dipolar nature of a water molecule with its polar energy field.

Microwave heating is most efficient on liquid water, but much less efficient on fats and sugars (which have less molecular dipole moment), and frozen water (where the molecules are not free to rotate). Microwave heating sometimes occurs due to rotational resonance of water molecules, which happens only at much higher frequencies, in the tens of Gigahertz.

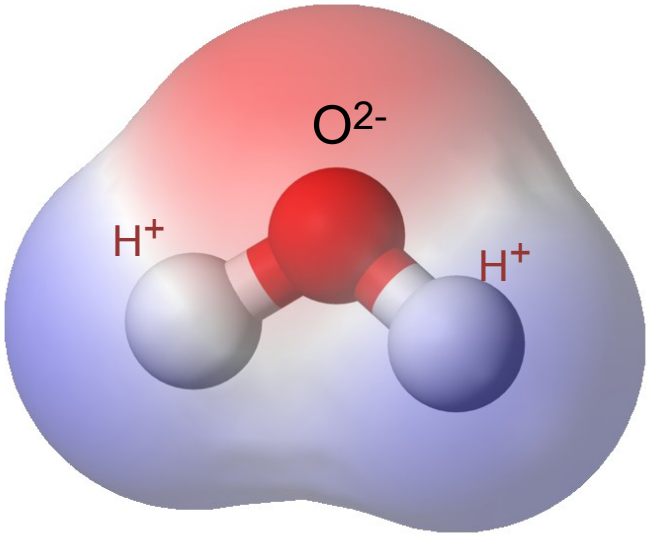

Figure 2.6. A dipolar water molecule with its polar energy field

In reality, microwaves are absorbed in the outer layers of food in a manner somewhat similar to heat from other methods. Microwaves penetrate dry substances at the surfaces of many common foods, and thus often deposit initial heat more deeply than other methods. Also the amount of heat lost to the surroundings is much higher on the surface of the food than in its interior. This gives an appearance that microwaves are heating the food from inside out, though they heat up almost every part of the food equally.

Depending on water content, the depth of initial heat deposition may be several centimeters or more with microwave ovens, in contrast to convection heating, which deposit heat shallowly at the food surface. Depth of penetration of microwaves is dependent on food composition and the frequency, with lower microwave frequencies being more penetrating.

At the consumer level, cheap ovens and fast heating (along with a tolerance for unevenly heated food) has led to the near saturation of microwave ovens. However, poor economics and complex heating patterns have led to its low industrial acceptance. Nevertheless, industrial application is possible if certain conditions are met. Special design of the microwave oven to address the complex heat distribution problem is possible if the food is fairly uniform in shape and composition. Furthermore, if the added quality is tangible to the point where the added expense of microwave processing can be passed along to the consumer, then an industrial process becomes more viable.

2.10 Microwave In-Shell Pasteurization of Eggs

Microwaves are energy rich electromagnetic radiations, whose energy is readily absorbed by substances containing dipolar molecules. The best example of a dipolar molecule is water. The frequently alternating polarity of the electromagnetic radiation (microwaves) causes a dipolar molecule to rotate and gets heated up due to molecular friction. Microwaves have the capability to penetrate substances that are opaque to visible light, thus making it suitable for heating up different food materials (Pozar 2005).

In order to minimize heat destruction of the food components and their functional and sensory qualities, novel methods have been investigated for pasteurization and sterilization (Rodriguez et al., 2003). Microwave processing offers the advantage of quick heating for pasteurization and sterilization. The current technique for in-shell

pasteurization of egg involves heating eggs in water bath at 60 ºC for about 20-25 minutes, depending on the size of the eggs. This leads to the overheating of the egg white proteins (i.e the egg white gets heated up more than the yolk, which is against recommendations) resulting in denaturation coagulation (Hou et al. 1996). This denaturation greatly affects the functional properties of the eggs. Therefore a process that can heat the shell eggs from inside will be the best alternative to solve this problem.

Between the egg white and the yolk, which are the two primary components of the egg, the albumen is the primary infection site as S*almonella* requires only indirect contact with the yolk for its growth and multiplication and hence the albumen is the primary target of microwave heating (Fleischman et al. 2003). When thinking of microwave heating of a shell egg, the first thing that comes to mind is the high risk of a great pressure build-up within the eggs. However this is not inevitable at pasteurization temperatures. With proper control of the process parameters, microwave heating can provide efficient and rapid heating for thermal pasteurization.

The issue of non-uniformity in microwave heating can be overcome with the proper orientation of the egg and a specially designed waveguide, which is an engineering issue (Fleischman 2004) and also by the precise design of the container (equipped with microwave susceptors) taking the eggs into the microwave chamber (Yakovlev 2001).

A complete understanding of the dielectric properties and egg curvature on power distribution will help design a system highly specific and efficient for this application. There are several ways of measuring the dielectric properties of different materials, such as the perturbation technique, transmission line technique, open-ended probe techniques, time domain reflectometry, free-space transmission techniques, microstrip transmission lines, etc. (Venkatesh and Raghavan 2005). Among these the open-ended coaxial probe technique was found to be more appropriate and precise for

measuring the dielectric properties of egg components. Dev et al. (2008) measured and modelled the dielectric properties of egg components and found eggs to be ideal for microwave heating.

Theoretical mathematical studies have shown that even though albumen exhibits better dielectric properties than yolk, the egg's curvature has a focussing effect which leads to a suitable power distribution. Laboratory trials on microwave heating of in-shell eggs indicated that, contrary to what one might expect, the heating rates of both albumen and yolk were similar. A combination of egg geometry, dielectric properties, and size were the main factors responsible for the enhanced interior heating. It was interesting to note that the yolk, which had the poorer dielectric properties, heated up a little faster than the egg white, when heated in the shell. Dev et al. (2008) suggested that the focusing effect of the egg-shell curvature, the spherical geometry and the central yolk position inside a shell egg resulted in a convergence of the microwave energy towards the center, hence increasing heat dissipation in the yolk (Datta et al., 2005). In addition, the radial penetration depth and loss/attenuation of the microwave energy could have contributed to the higher heating rate of the yolk. This addresses the issue of there being a greater potential for increased *Salmonella* concentrations in the yolk, than the egg white.

Also, though the cytoplasm of the microorganism (*Salmonella*) itself has its own dielectric properties and responds to microwaves (i.e. it is expected to be highly reactive to microwaves due to high moisture content of the cytoplasm), this does not guarantee the 5-log reduction required for pasteurization, which must be confirmed experimentally. The eggs that are subjected to microwave pasteurization are thoroughly washed with detergent and sanitized with chlorinated water to remove all surface contaminants. This process ensures complete removal of microbes on the surface of the egg if carried out as prescribed (Srikaeo and Hourigan, 2002).

Mermelstein (2001), citing Dr. Fleischman, senior research scientist, Food safety division of the US Food and Drug Administration, stated “if ever microwave processing needed a specific type of product it could do better than any other process it’s this. Microwaves are ideally suited for pasteurization of shell eggs.”

2.10.1 Reasons for choosing a multimode cavity for in-shell egg pasteurization

Focussed microwave systems nevertheless give better process control and higher efficiencies compared to multimode cavities; however, a multimode cavity was chosen for the in-shell pasteurization of eggs because of the following reasons.

1) In light of the penetration depth and skin depth ($2 \times Dp$) calculations for the in-shell egg, based on the dielectric properties of the egg white and egg yolk, microwaves can effectively penetrate only a little more than half the largest diameter of the egg along the smaller axis of the egg. Therefore a focussed microwave system will result in less uniform heating as far as the shell eggs are concerned unless the eggs are rotated while heating.
2) Even if rotated in a monomode cavity, theoretically the egg white would heat up much faster than the yolk, as the simultaneous heating from different directions as obtained in a multimode cavity cannot be obtained while using a focussed microwave setup.
3) Also, the slightly higher temperatures produced inside the yolk compared to the egg white, (Dev et al, 2008) due to the so called focussing effect happening inside the egg, (Datta et al, 2005) which is the requirement for pasteurization, cannot be taken advantage of while using a focussed microwave system.
4) Also as discussed earlier, scaling up the process gets much more difficult and limited, if one has to employ a monomode cavity for this purpose.

2.11 Speciality Eggs

There are several value-added eggs recently introduced into the market and sold at a premium for their value addition. These are collectively called as speciality eggs.

2.11.1 Organic eggs

These are eggs produced by hens that are fed a special feed having ingredients that were grown without pesticides, herbicides and commercial fertilizer so as to preserve the integrity of the soil. They have the same nutritional value as any other egg.

2.11.2 Vegetarian eggs

These are speciality produced by hens that are fed a special diet of feed containing ingredients of plant origin only (No animal by-products).

2.11.3 Omega 3 eggs

Recently, chicken eggs that are especially high in Omega 3 fatty acids have come on the market. These eggs are made by feeding laying hens a diet containing polyunsaturated fats using flax seeds and kelp meal. Nutrition information on the packaging is different for each of the brands.

2.11.4 Vitamin enhanced eggs

These eggs are from hens fed a nutritionally-enhanced diet having higher levels of certain nutrients (eg. vitamin E, folate, vitamin B-6, vitamin B-12). As a result, these eggs contain slightly higher amounts of these nutrients.

2.11.5 In- shell pasteurized eggs

These are eggs recently introduced into the market. As the name implies, the eggs are heat pasteurized in hot water or hot air or a combination of both. These eggs are the safest, but do not retain the exceptional functional properties of other raw eggs.

This study is aimed at improving the functional properties of in-shell pasteurized eggs and thereby, improving the functional quality of the pasteurized eggs by using microwaves for the task.

2.12 Feasibility of Industrial Application

In North America, the two microwave frequencies allowed for processing are at 915 and 2450 MHz. Microwave heating refers to the use of electromagnetic waves to generate heat directly into the food material (Datta et al., 2005). The temperature rise in the food depends on the duration of the exposure to the microwave, the frequency of the electromagnetic wave, the thermo-physical properties of the food, the extent of evaporation, and physical parameters of the food and the applicator. For solid and semi-solid food, one of the advantages of microwave heating over conventional heating is its ability to generate a much faster heating rate. High temperature and short time processing can be achieved whereby bacterial destruction is accomplished with minimal degradation of the desired components.

Industrial applications of microwave heating, especially in the food industry have been hampered by the lack of an appropriate model/simulator. This is due to the complexity of the equations to be solved to optimize the energy distribution for a given geometry and thermo-physical characteristics of the food materials (Knoerzer et al, 2005). As a result, product and process development was essentially done through trial-and-error. Nonetheless, commercial applications of microwave processing have been developed and documented in the literature (Decareau, 1985; Schlegel, 1992; Harlfinger, 1992; Orsat et al., 2005).

2.13 Existing Patents

A patent search revealed that three patents on the use of microwave energy for the pasteurization if in-shell eggs have been filed for protection at the International Bureau of the World Intellectual Property Organization (WO 2005/102064, 2005; WO 2004/037012, 2004; WO 2003/024249, 2003). These documents describe various approaches/contraptions to integrate microwave heating to egg packing line. However, none of them address the issues of egg quality and heating uniformity.

2.14 Recent Findings

Recent work published by Dev et al. (2008) confirmed that 2450 MHz microwaves can be successfully used to raise the temperature of in-shell eggs to the required pasteurization temperature in few minutes. It took about 65 sec. to reach the pasteurization temperatures at a power density of 3 W g^{-1} and 320 sec. at 0.75 W g^{-1}. Results also indicated that with adequate microwave power modulation the yolk reached a higher temperature of 61°C while simultaneously maintaining the albumen at 57°C. These are the exact temperatures required for proper pasteurization of the egg constituents. Lakins et al. (2008) had reported that applying directional microwave technology resulted in a 2-log reduction of *S. enteriditis.* Although the measurement of the dielectric properties indicated that the albumen absorbed microwave energy better than the yolk, this difference in heating was attributed to the combination of the egg curvature and the microwave frequency used for the treatment (Datta et al., 2005). Once again, it appears that in-shell eggs are perfectly suited for microwave pasteurization.

2.15 Economic Overheads due to Pasteurization

More than 85 % of Canadians are ready to pay a premium for safe and high quality food (CEMA 2002). It is further evident that the percentage of total eggs broken has increased from 5% in 1952 to more than 20% in 1998 (AAFC 2005). Though this has led to the growth of the processed egg industry, poultry farmers lose a major part of their profit, as they are paid only the minimum cost of production (COP) for breaking stock.

As well as assuring consumer safety, pasteurized eggs exhibit a better keeping quality and hence a longer shelf life. Though pasteurization will increase the COP by a few cents per dozen eggs, the returns that the farmers get out of this will be much more. This will also help to safeguard the interest of the farmers and to provide safe eggs to consumers. This may also increase the export market, thereby generating millions of dollars as revenue.

2.16 Preliminary studies

Preliminary studies have shown that in-shell eggs can be brought to the desired temperature and maintained there for pasteurization. In an in-shell egg exposed to microwaves, the yolk heated up faster than the albumen and this was a desirable effect as the pasteurization temperature requirement for the yolk is higher than that for the albumen. In a preliminary study, the dielectric properties of the constituents (egg shell, albumen and yolk) were measured at temperatures ranging from 0 to 65℃ and mathematical relationships were established. A computer controlled multi-mode microwave oven operating at 2450 MHz was used to study the heating characteristics of in-shell eggs (Dev at al., 2008).

Modifications were made to the existing unit to accommodate a rotating egg holder. Optic fiber temperature sensors were used to monitor the yolk and albumen temperature. The effects on egg quality of initial temperature, microwave power, location/orientation of the in-shell egg in relation to the microwave source, surrounding air temperature, heating rate, heating time, and temperature distribution were studied. Power levels of 0.5, 1.0, 2.5 and 5.0 W g^{-1} were investigated. Regression models were developed to relate the mass of the egg, initial temperature and microwave power to heating rates and time to reach the pasteurization temperature.

Assessment of the egg quality was limited to visual observations such as: shell integrity and discoloration, colour and turbidity (presence of micro-coagulates) of the albumen, colour of the yolk and incidence and extent of coagulation (Dev et al., 2010).

2.17 Modelling and Simulations

Though heating an egg is a straight forward thermal processing approach, measuring the temperature profile and the energy distribution within a shell egg is a challenge, due to the opaque and brittle nature of the shell. Nonetheless, one can insert probes into different locations inside a shell egg and measure the temperature. But the repeatability of measurements at the exact locations is impractical. Also there is a limit to the number of probes that can be inserted into a shell egg, as every hole drilled into the shell weakens the strength of the shell significantly and the shell will collapse beyond a certain number of holes. Due to this limitation, the temperature distribution at every point inside a shell egg is hard to measure and analyse. This task becomes even more challenging in a microwave environment. But the ability to measure and analyse the temperature distribution is mandatory to ensure egg pasteurization. Theoretical mathematical and numerical modelling and simulation helps understanding and

predicting the temperature distribution inside biological medium. Therefore modelling and simulation of microwave heating of eggs was carried out.

Simulations of electromagnetic energy distribution and heat generation involves solving sets of complex Partial Differential Equations (PDEs). Numerical approximation techniques like the Finite Difference Time Domain (FDTD) and Finite Element Method (FEM) are commonly used to solve for different variables in PDEs. The FEM technique competes very favourably with other numerical methods, as it is based on reducing the Maxwell's equations to a system of simultaneous algebraic linear equations (Delisle, Wu & Litva, 1991). FEMs can readily model heterogeneous and anisotropic materials as well as arbitrarily shaped geometries. It can also provide both time and frequency domain analyses, which are important in dealing with microwave heating problems like field distribution, scattering parameters and dissipated power distribution for various materials and geometries (Dai, 2006).

There is poor understanding of the mechanisms involved in the actual energy distribution inside the eggs when subjecting them to electromagnetic field. The electromagnetic field distribution inside the microwave oven can be traced out by solving the Maxwell's equations (Dev et al., 2008b). The FEM is commonly used for solving Maxwell's equations to get the energy distribution in a complex object or within a multimode cavity and it is capable of simulating power density distribution in a 3-D space. (Fu and Metaxas, 1994; Zhou, Puri, Anantheswaran et al.,1995).

A three-dimensional finite element model needs to be developed using proprietary software namely, MATLAB (Version 7.7) from Mathworks Inc, USA and COMSOL (Version 3.5a) from COMSOL Inc, Boston, USA. COMSOL is a finite element modeling software package, in which most of the modeling and simulation is done with the help of its graphical user interface, but this package lacks certain features like

simulation of a rotating/moving object in electromagnetic field. This gap is bridged by MATLAB coding specifically developed for a given object geometry. This model will be useful for determining microwave energy distribution and for the prediction of temperature profiles inside the in-shell eggs during microwave processing. This model will be developed taking into consideration the complex shape, dielectric properties and heterogeneous composition of the in-shell egg.

2.17.1 Finite Element Method

The finite element method (FEM) (sometimes referred to as finite element analysis) is a numerical technique for finding approximate solutions of partial differential equations (PDE) as well as of integral equations. The solution approach is based either on eliminating the differential equation completely (steady state problems), or rendering the PDE into an approximating system of ordinary differential equations, which are then numerically integrated using standard techniques such as Euler's method, Runge-Kutta, etc.

In solving partial differential equations, the primary challenge is to create an equation that approximates the equation to be studied, but is numerically stable, meaning that errors in the input data and intermediate calculations do not accumulate and cause the resulting output to be meaningless. There are many ways of doing this, all with advantages and disadvantages.

The Finite Element Method is a good choice for solving partial differential equations over complex domains (like cars and oil pipelines), especially when the domain changes (for instance situations such as during a solid state reaction with a moving boundary), when the desired precision varies over the entire domain, or when the solution lacks smoothness.

2.17.1.1 Mesh

The starting point for the finite element method is a mesh, a partition of the geometry into small units of a simple shape, *mesh elements*. The term “mesh element” means any of the mesh elements—mesh faces, mesh edges, or mesh vertices. When considering a particular *d*-dimensional domain in the geometry (that is, a subdomain, boundary, edge, or vertex), then its mesh elements mean the *d*-dimensional mesh elements contained in the domain.

The different types of mesh elements include:

- For a 1D geometry the mesh generation is simply partitioning the subdomains (intervals) into smaller intervals (or mesh elements). The endpoints of the mesh elements are called mesh vertices.
- For a 2D geometry the mesh generation involves partitioning the subdomains into *triangular* or *quadrilateral* mesh elements. If the boundary is curved, these elements represent only an approximation of the original geometry. The sides of the *triangles* and *quadrilaterals* are called mesh edges, and their corners are mesh vertices. A mesh edge must not contain mesh vertices in its interior. Similarly, the boundaries defined in the geometry are partitioned (approximately) into mesh edges, so-called boundary elements, which must conform to the mesh elements of the adjacent subdomains. If there are isolated points in the geometry, these also become mesh vertices.
- In 3D geometry the mesh generation involves partitioning the subdomains into *tetrahedral, hexahedral,* or *prism* mesh elements whose faces, edges, and corners are called mesh faces, mesh edges, and mesh vertices, respectively. The boundaries in this geometry are partitioned into triangular or quadrilateral boundary elements. The geometry edges are partitioned into edge elements. Isolated geometry vertices become mesh vertices.

2.17.1.2 Finite Elements Approximation Technique

Once the mesh is ready, approximations can be introduced to the dependent variables. For this discussion, let us concentrate on the case of a single variable, *u*. The idea is to approximate *u* with a function that one can describe with a finite number of parameters, the so-called *degrees of freedom* (DOF). Inserting this approximation into the weak form of the equation generates a system of equations for the degrees of freedom.

Starting with a simple example, linear elements in 1D and assuming that a mesh consists of just two mesh intervals: $0 < x < 1$ and $1 < x < 2$. Linear elements means that on each mesh interval the continuous function *u* is linear (affine). Thus, the only thing one needs to know in order to characterize *u* uniquely is its values at the *node points* $x_1 = 0$, $x_2 = 1$, and $x_3 = 2$. Let us denote these as $U_1 = u(0)$, $U_2 = u(1)$, $U_3 = u(2)$. These are the *degrees of freedom*.

Now this can be written as

$$u(x) = U_1\varphi_1(x) + U_2\varphi_2(x) + U_3\varphi_3(x) \tag{3.1}$$

where, $\varphi_i(x)$ are certain piecewise linear functions. Namely, $\varphi_i(x)$ is the function that is linear on each mesh interval, equals 1 at the i^{th} node point, and equals 0 at the other node points. For example,

$$\varphi_1(x) = \begin{cases} 1-x & \text{if } 0 \le x \le 1 \\ 0 & \text{if } 1 \le x \le 2 \end{cases} \tag{3.2}$$

The $\varphi_i(x)$ are called the *basis functions*. The set of functions $U(x)$ is a linear function space called the *finite element space*.

For better accuracy, another finite element space corresponding to quadratic elements can be considered. Functions *U* in this space are second-order

polynomials on each mesh interval. To characterize such a function, one must introduce new node points at the midpoint of each mesh interval: $x_4 = 0.5$ and $x_5 = 1.5$. One must also introduce the corresponding degrees of freedom $U_i = u(x_i)$. Then, on each mesh interval, the second-degree polynomial $u(x)$ is determined by the different degrees of freedom at the endpoints and the midpoint. Then

$$u(x) = U_1\varphi_1(x) + U_2\varphi_2(x) + U_3\varphi_3(x) + U_4\varphi_4(x) + U_5\varphi_5(x) \qquad (3.3)$$

where the basis functions $\varphi_i(x)$ now have a different meaning. Specifically, this is the function that is quadratic on each mesh interval, equals 1 at the i^{th} node point, and equals 0 at the other node points. For example,

$$\varphi_1(x) = \begin{cases} (1-x)(1-2x) & \text{if } 0 \le x \le 1 \\ 0 & \text{if } 1 \le x \le 2 \end{cases} \qquad (3.4)$$

In general, one can specify a finite element space by giving a set of basis functions. The description of the basis functions is simplified by the introduction of *local coordinates* (or *element coordinates*). Considering a mesh element of dimension d in an n- dimensional geometry (whose space coordinates are denoted $x_1,..., x_n$) and also the *standard d-dimensional simplex*

$$\xi_1 \ge 0, \xi_2 \ge 0, \cdots, \xi_d \ge 0 \text{ and } \xi_1 + \xi_2 + \cdots \xi_d \le 1 \qquad (3.5)$$

which resides in the local coordinate space parametrized by the local coordinates $\xi_1, \ldots, \xi_d$. If $d = 1$, then this simplex is the unit interval. If $d = 2$, it is a triangle with two 45 degree angles, and if $d = 3$ it is a tetrahedron. Now, one can consider the mesh element as a linear transformation of the standard simplex. Namely, by letting the global space coordinates x_i be suitable linear (affine) functions of the local coordinates, one can get the mesh element as the image of the standard simplex.

2.17.2 Finite element modelling in microwave pasteurization of shell eggs

The problem of electromagnetic analysis on a macroscopic level is the problem of solving *Maxwell's equations* subject to certain boundary conditions. Maxwell's equations are a set of equations, written in differential or integral form, stating the relationships between the fundamental electromagnetic quantities. These quantities are the *electric field intensity* **E**, the *electric displacement* or *electric flux density* **D**, the *magnetic field intensity* **H**, the *magnetic flux density* **B**, the *current density* **J** and the *electric charge density* ρ.

The equations can be formulated in differential or integral form. The differential form are presented here, because it leads to differential equations that the finite element method can handle. For general time-varying fields, Maxwell's equations can be written as

$$\begin{aligned} \nabla \times H &= J + \frac{\partial D}{\partial t} \\ \nabla \times E &= \frac{\partial B}{\partial t} \\ \nabla \cdot D &= \rho \\ \nabla \cdot B &= 0 \end{aligned} \tag{3.6}$$

The first two equations are also referred to as *Maxwell-Ampère's law* and *Faraday's law*, respectively. Equation three and four are two forms of *Gauss' law*, the electric and magnetic form, respectively.

Another fundamental equation is the *equation of continuity*, which can be written as:

$$\nabla \cdot J = -\frac{\partial \rho}{\partial t} \tag{3.7}$$

Out of the five equations mentioned, only three are independent. The first two combined with either the electric form of Gauss' law or the equation of continuity form such an independent system.

2.17.2.1 Constitutive Relations

To obtain a closed system, the constitutive relations describing the macroscopic properties of the medium, are included. They are given as:

$$\begin{aligned} D &= \varepsilon_0 E + P \\ B &= \mu_0 (H + M) \\ J &= \sigma E \end{aligned} \tag{3.8}$$

where, ε_0 is the permittivity of a vacuum, μ_0 is the permeability of a vacuum, and σ the electric conductivity. In the SI system, the permeability of vacuum is chosen to be $4\pi \cdot 10^{-7}$ H m^{-1}.

The velocity of an electromagnetic wave in vacuum is given as C_0 and the permittivity of vacuum is derived from the relation:

$$\varepsilon_0 = \frac{1}{c_0^2 \mu_0} = 8.854 \times 10^{-12} \text{ F m}^{-1} \approx \frac{1}{36\pi} \times 10^{-9} \text{ F m}^{-1} \tag{3.9}$$

The electric polarization vector P describes how the material is polarized when an electric field *E* is present. It can be interpreted as the volume density of electric dipole moments. *P* is generally a function of *E*. Some materials can have a nonzero *P* also when there is no electric field present.

The magnetization vector *M* similarly describes how the material is magnetized when a magnetic field *H* is present. It can be interpreted as the volume density of

magnetic dipole moments. *M* is generally a function of *H*. Permanent magnets, for instance, have a nonzero *M* also when there is no magnetic field present.

For linear materials, the polarization is directly proportional to the electric field, $P = \varepsilon_0 \chi_e E$, where χ_e is the electric susceptibility. Similarly in linear materials, the magnetization is directly proportional to the magnetic field, $M = \chi_m H$, where χ_m is the magnetic susceptibility. For such materials, the constitutive relations can be written:

$$\begin{aligned} D &= \varepsilon_0(1+\chi_c)E = \varepsilon_0\varepsilon_r E = \varepsilon E \\ B &= \mu_0(1+\chi_m)H = \mu_0\mu_r H = \mu H \end{aligned} \tag{3.10}$$

The parameter ε_r is the relative permittivity and μ_r is the relative permeability of the material. These are usually scalar properties but they can, for a general anisotropic material, be 3-by-3 tensors. The properties ε and μ (without subscripts) are the permittivity and permeability of the material.

2.17.2.2 Generalized Constitutive Relations

Generalized forms of the constitutive relations are well suited for modeling nonlinear materials. The relation used for the electric fields is:

$$D = \varepsilon_0\varepsilon_r E + D_r \tag{3.11}$$

The field D_r is the *remanent displacement*, which is the displacement when no electric field is present.

Similarly, a generalized form of the constitutive relation for the magnetic field is:

$$B = \mu_0\mu_r H + B_r \tag{3.12}$$

where $\mathbf{B_r}$ is the *remanent magnetic flux density*, which is the magnetic flux density when no magnetic field is present.

The relation defining the current density is generalized by introducing an externally generated current $\boldsymbol{J}^e$.

The resulting constitutive relation is:

$$J = \sigma E + J^e \tag{3.13}$$

2.17.2.3 Potentials

Under certain circumstances it can be helpful to formulate the problems in terms of the *electric scalar potential V* and the *magnetic vector potential* $\boldsymbol{A}$. They are given by the equalities:

$$\begin{aligned} B &= \nabla \times A \\ E &= -\nabla V - \frac{\partial A}{\partial t} \end{aligned} \tag{3.14}$$

The defining equation for the magnetic vector potential is a direct consequence of the *t* magnetic Gauss' law. The electric potential results from Faraday's law.

In the magnetostatic case where there are no currents present, Maxwell-Ampère's law reduces to

$$\nabla \times H = 0 \tag{3.15}$$

When this holds, it is also possible to define a *magnetic scalar potential* by the relation

$$H = -\nabla V_m \tag{3.16}$$

2.17.2.4 Electromagnetic Energy

The electric and magnetic energies are defined as

$$W_e = \int_V \left(\int_0^D E.dD\right) dV = \int_V \left(\int_0^T E.\frac{\partial D}{\partial t}\,\mathrm{dt}\right) dV$$

$$W_m = \int_V \left(\int_0^B H.dB\right) dV = \int_V \left(\int_0^T H.\frac{\partial B}{\partial t}\,dt\right) dV \qquad (3.17)$$

The time derivatives of these expressions are the electric and magnetic power

$$P_e = \int_V E.\frac{\partial D}{\partial t}\,dV$$

$$P_m = \int_V H.\frac{\partial B}{\partial t}\,dV \qquad (3.18)$$

These quantities are related to the resistive and radiative energy, or energy loss, through Poynting's theorem

$$-\int_V \left(E.\frac{\partial D}{\partial t} + H.\frac{\partial B}{\partial t}\right) dV = \int_V \mathrm{J.\,E\,dV} + \oint_s (\mathrm{E\,x\,H}).n\,ds \qquad (3.19)$$

where V is the computation domain and S is the closed boundary of V.

The first term on the right-hand side represents the resistive losses,

$$P_h = \int_V \mathrm{J}.E\,dV \qquad (3.20)$$

which result in heat dissipation in the material. The current density $\boldsymbol{J}$ in this expression is the one appearing in Maxwell-Ampère's law.

The second term on the right-hand side of Poynting's theorem represents the radiative losses,

$$P_r = \bigcirc \qquad (3.21)$$

The quantity $\boldsymbol{S = E \times H}$ is called the Poynting vector.

Under the assumption the material is linear and isotropic, it holds that

$$\mathrm{E}\,.\,\frac{\partial D}{\partial t} = \varepsilon E.\,\frac{\partial E}{\partial t} = \frac{\partial}{\partial t}\left(\frac{1}{2}\varepsilon\, E.\,E\right)$$

$$\mathrm{H}.\,\frac{\partial B}{\partial t} = \frac{1}{\mu}B.\,\frac{\partial B}{\partial t} = \frac{\partial}{\partial t}\left(\frac{1}{2\mu}\, B.\,B\right) \qquad (3.22)$$

By interchanging the order of differentiation and integration (justified by the fact that the volume is constant and the assumption that the fields are continuous in time), then we get

$$-\frac{\partial}{\partial t}\int_V \left(\frac{1}{2}\varepsilon \mathrm{E}.\,\mathrm{E} + \frac{1}{2\mu}\,\mathrm{B}.\,\mathrm{B}\right) dV = \int_V \mathrm{J}\,.\,\mathrm{E}\,\mathrm{dV} + \oint_S (\mathrm{E}\,\mathrm{x}\,\mathrm{H})\,.\,nds \qquad (3.23)$$

The integrand of the left-hand side is the total electromagnetic energy density

$$\omega = \omega_c + \omega_m = \frac{1}{2}\,\varepsilon\,\mathrm{E}.\,\mathrm{E} + \frac{1}{2\mu}\,\mathrm{B}.\,\mathrm{B} \qquad (3.24)$$

2.17.2.5 Material properties

These general constitutive relationships get a little more complicated for a non-homogeneous material like eggs, as it requires interfacial boundary conditions.

An non-homogeneous medium is one where the constitutive parameters vary with the space coordinates, so that different field properties prevail at different parts of the material structure.

2.17.2.6 Boundary and Interface Conditions

To get a full description of an electromagnetic problem, one must specify boundary conditions at material interfaces and physical boundaries. At interfaces between two media, the boundary conditions can be expressed mathematically as

$$\left.\begin{aligned} n_2 \times (E_1 - E_2) &= 0 \\ n_2 \times (D_1 - D_2) &= \rho_s \\ n_2 \times (H_1 - H_2) &= J_s \\ n_2 \times (B_1 - B_2) &= 0 \end{aligned}\right\} \quad (3.25)$$

where ρ_s and J_s denote *surface charge density* and *surface current density*, respectively, and n_2 is the outward normal from medium 2. Of these four conditions, only two are independent. One of the first and the fourth equations, together with one of the second and third equations, form a set of two independent conditions.

A consequence of the above is the interface condition for the current density,

$$n_2 \times (J_1 - J_2) = \frac{\partial \rho_s}{\partial t} \quad (3.26)$$

2.17.2.7 Interface between a Dielectric and a Perfect Conductor

A perfect conductor has infinite electric conductivity and thus no internal electric field. Otherwise, it would produce an infinite current density according to the third fundamental constitutive relation. At an interface between a dielectric and a perfect conductor, the boundary conditions for the E and D fields are simplified. If, say, subscript 1 corresponds to the perfect conductor, then $D_1 = 0$ and $E_1 = 0$ in the relations above. For the general time-varying case, it holds that $B_1 = 0$ and $H_1 = 0$ as well (as a consequence of Maxwell's equations). What remains is the following set of boundary conditions for time-varying fields in the dielectric medium.

$$\left.\begin{aligned} -n_2 \times E_2 &= 0 \\ -n_2 \times H_2 &= J_s \\ -n_2 \times D_2 &= \rho_s \\ -n_2 \times B_2 &= 0 \end{aligned}\right\} \quad (3.27)$$

2.17.2.8 Phasors

Whenever a problem is time-harmonic the fields can be written in the form

$$E(r,t) = \hat{E}\,(r)\cos\,(\omega t + \varphi) \tag{3.28}$$

Instead of using a cosine function for the time dependence, it is more convenient to use an exponential function, by writing the field as

$$E(r,t) = \hat{E}\,(r)\cos(\omega t + \varphi) = R_e\left(\hat{E}\,(r)e^{j\varphi}e^{j\omega t}\right) = R_e\left(\tilde{E}\,(r)e^{j\omega t}\right) \tag{3.29}$$

The field $\tilde{E}\,(r)$ is a *phasor*, which contains amplitude and phase information of the field but is independent of *t*. One thing that makes the use of phasors suitable is that a time derivative corresponds to a multiplication by $j\omega$,

$$\frac{\partial E}{\partial t} = R_e\left(\tilde{E}\,(r)e^{j\omega t}\right) \tag{3.30}$$

This means that an equation for the phasor can be derived from a time-dependent equation by replacing the time derivatives by a factor $j\omega$. All time-harmonic equations in the RF Module are expressed as equations for the phasors and the tilde is dropped from the variable denoting the phasor.

When postprocessing the solution of a time-harmonic equation, it is important to remember that the field that has been calculated is a phasor and not a physical field. For example, all plot functions visualize $R_e\left(\tilde{E}\,(r)\right)$ by default, which is ***E*** at time $t = 0$. To obtain the solution at a given time, one can specify a phase factor.

2.17.3 Finite Difference Method

Finite-difference methods approximate the solutions to differential equations by replacing derivative expressions with approximately equivalent difference quotients. That is, because the first derivative of a function f is, by definition,

$$f'(a) = \lim_{h \to 0} \frac{f(a+h) - f(a)}{h} \tag{3.31}$$

then a reasonable approximation for that derivative would be to take

$$f'(a) \approx \frac{f(a+h) - f(a)}{h} \tag{3.32}$$

for some small value of h. In fact, this is the forward difference equation for the first derivative. Using this and similar formulae to replace derivative expressions in differential equations, one can approximate their solutions without the need for calculus.

2.17.4 Comparison of finite element method to the finite difference method

The finite difference method (FDM) is an alternative way of approximating solutions of PDEs. The differences between FEM and FDM are:

- The FDM is an approximation to the differential equation; the FEM is an approximation to its solution.
- The most attractive feature of the FEM is its ability to handle complex geometries (and boundaries) with relative ease. While FDM in its basic form is restricted to handle rectangular shapes and simple alterations thereof, the handling of geometries in FEM is theoretically straightforward.
- The most attractive feature of finite differences is that it can be very easy to implement.
- There are several ways one could consider the FDM a special case of the FEM approach. One might choose basis functions as either piecewise constant functions or Dirac delta functions. In both approaches, the approximations are defined on the entire domain, but need not be continuous. Alternatively, one might define the function on a discrete domain, with the result that the continuous differential operator no longer makes sense, however this approach is not FEM.

- There are reasons to consider the mathematical foundation of the finite element approximation more sound, for instance, because the quality of the approximation between grid points is poor in FDM.
- The quality of a FEM approximation is often higher than in the corresponding FDM approach, but this is extremely problem-dependent and several examples to the contrary can be provided.

Generally, FEM is the method of choice in all types of analysis in structural mechanics (i.e. solving for deformation and stresses in solid bodies or dynamics of structures) while computational fluid dynamics (CFD) tends to use FDM or other methods like finite volume method (FVM). CFD problems usually require discretization of the problem into a large number of cells and gridpoints (millions and more), therefore cost of the solution favours simpler, lower order approximation within each cell. This is especially true for 'external flow' problems, like air flow around the car or airplane, or weather simulation in a large area

2.18 Summary

Eggs are highly nutritious but potentially dangerous when consumed raw. Egg proteins are heat sensitive and hence their functional quality and consumer acceptance is adversely affected by thermal treatments. Microwave heating provides an excellent alternative to this problem. The issue of non-uniformity and localized overheating in microwave environment needs to be researched and resolved. Numerical modelling and simulation techniques can help visualize the temperature distribution inside a shell eggs placed in a microwave environment and thereby help designing an appropriate microwave waveguide applicator and cavity for the in-shell pasteurization of eggs.

Connecting text

It is clear from chapter 2 that numerical modelling and simulation is inevitable to design and develop a microwave pasteurizer for shell eggs. One can take different numerical approaches for predicting the temperature profile inside a food material subjected to dielectric heating. FDTD modelling and simulation, though restricted to handle rectangular shapes and simple alterations thereof, is relatively a simple and less computationally intensive approach. It can also provide both time and frequency domain analyses which are important to microwave heating problems like field distribution, scattering parameters and dissipated power distribution for various materials and geometries. Therefore in order to obtain a preliminary conceptual visualization of the power distribution inside a shell egg while taking advantage of the ease of implementation, an FDTD simulation model for the microwave heating of shell eggs was developed.

Chapter 3

FDTD MODELING AND SIMULATION OF MICROWAVE HEATING OF IN-SHELL EGGS

3.1 Abstract

Considering microwaves as a viable alternative for the pasteurization of In-shell eggs, preliminary trials performed had confirmed that microwave at 2450 MHz can be successfully used to raise the temperature of in-shell eggs to the required pasteurization temperatures in a few minutes. Based on these trials a finite difference time domain (FDTD) model was developed using C language and MATLAB to simulate the *E* field and power distribution in lossy dielectric media like that of the egg components (egg white and yolk) taking into consideration the complex shape, dielectric properties and heterogeneous composition of the in-shell egg. This can be used to assist in the design and development of an industrial microwave in-shell eggs pasteurization unit.

Keywords. Microwave pasteurization, Simulation, In-shell heating

3.2 Introduction

Eggs have a rich nutritive value. Thus eggs are potential hosts and carriers for pathogenic microbes like *Salmonella enteritidis* and the most deadly strain (H5N1) of the avian flu virus. Heat pasteurization is a well-known process for enhancing food safety. However, egg proteins are extremely heat sensitive. Therefore heat pasteurization with minimal changes to the egg proteins needs consideration. Conventional methods of heat pasteurization using hot air and hot water bath severely affect the functional quality of the eggs. Microwaves provide a viable alternative for the pasteurization of In-shell eggs (Dev et al., 2008).

The following are a few among the several factors to be taken into account, while considering microwaves to do the job.

- Microwave heating is fairly non-uniform
- Heterogeneity of the egg
- Complexity in locating the points of overheating
- Remediation of cold spots through specific design alternatives

Finite element and Finite Difference Time Domain (FDTD) are two commonly used methods for solving Maxwell's equation to describe the energy distribution in a complex object or within a multimode cavity, and both methods are capable of simulating power density distribution in a 3-D space. (Fu and Metaxas, 1994; Harms, et al. 1996; Meredith, 1994; Zhou, et al. 1995; Ma, et al., 1995). The finite element method is suitable for arbitrarily shaped non-homogeneous objects and requires the solution of a sparse matrix which can prove very complicated. Comparatively, FDTD is a very straight forward method that can readily model non-homogeneous and anisotropic materials as well as arbitrarily shaped geometries; it can also provide both time and frequency domain analyses which are important to microwave heating problems like field distribution, scattering parameters and dissipated power distribution for various materials and geometries (Dai, 2006).

A normal egg would explode during microwave (MW) heating due to water vapour pressure build-up inside the shell. Besides this, when it comes to pasteurization, heating uniformity is a critical factor, but microwave heating is fairly non uniform. In this study, a FDTD method was used for the numerical simulation of microwave heating of a heterogeneous multilayered complex geometry like that of in-shell eggs and experimental validation of the simulation process was undertaken.

3.3 Materials & Methods

MATLAB version R2007a was used for the FDTD simulation of the Microwave heating of eggs. First the electromagnetic model for a microwave oven with a waveguide and the egg placed inside it was created and FDTD method was used to solve the model numerically and the electric field inside the cavity was traced. This field was then used to compute the power loss within the egg. This power loss was used as the source term for the heat equations which were, in turn, solved in order to calculate the temperature variation within the egg.

3.3.1 Electromagnetic Model for Field Distribution

At the macroscopic level, electromagnetic phenomena were defined using Maxwell Equations. The electromagnetic field distribution inside the microwave oven was traced out by solving the following Maxwell's Equations (Pozar, 1998).

$$\begin{aligned} \nabla \times E &= -j\omega\mu H \\ \nabla \times H &= j\omega\varepsilon_0 \varepsilon E \\ \nabla \cdot E &= 0 \\ \nabla \cdot H &= 0 \end{aligned} \tag{3.1}$$

where, E is electric field intensity in V m^{-1} and H is the magnetic field intensity in A m^{-1}

Also the Time Harmonic Function can be written as:

$$\begin{aligned} E(x,y,z,t) &= E_0(x,y,z)e^{j\omega t} \\ H(x,y,z,t) &= H_0(x,y,z)e^{j\omega t} \end{aligned} \tag{3.2}$$

The relative permittivity ε can be expressed in complex form as

$$\varepsilon = \varepsilon' - je'' \tag{3.3}$$

Where ε' is dielectric constant and ε" is the dielectric loss factor

For an isotropic lossy material, the loss characteristics of the material are given by the loss tangent, as defined by:

$$\tan\delta = \frac{\sigma}{2\pi f \varepsilon_0 \varepsilon_r} \tag{3.4}$$

where, f = frequency (Hz), σ = conductivity (S m^{-1}), ε_o = free-space permittivity (F m^{-1}) , and ε_r = relative permittivity (unitless)

For a material with complex permittivity as in (3), the loss tangent is defined by:

$$\tan\delta = \frac{-\varepsilon''}{\varepsilon'} \tag{3.5}$$

The following relationships exist between electric and magnetic field intensity and flux density,

$$\begin{aligned} \tilde{B} &= \mu_o \tilde{H} \\ \tilde{D} &= \epsilon_o \tilde{E} \end{aligned} \tag{3.6}$$

where, $\mu_0 = 4\pi \times 10^{-7}$ henry m^{-1}, is the free space permittivity, and $\varepsilon_0 = 8.85 \times 10^{-12}$ farad m^{-1}, is the free space permeability

The following scalar equations (Equations (7)-(12)) can be produced from the time dependent Maxwell's equations (Pozar, 1998)

$$\frac{\partial \tilde{D}_x}{\partial t} = \frac{1}{\sqrt{\varepsilon_o \mu_o}} \left(\frac{\partial H_z}{\partial y} - \frac{\partial H_y}{\partial z} \right) \tag{3.7}$$

$$\frac{\partial \tilde{D}_y}{\partial t} = \frac{1}{\sqrt{\varepsilon_o \mu_o}}\left(\frac{\partial H_x}{\partial z} - \frac{\partial H_z}{\partial x}\right) \tag{3.8}$$

$$\frac{\partial \tilde{D}_z}{\partial t} = \frac{1}{\sqrt{\varepsilon_o \mu_o}}\left(\frac{\partial H_y}{\partial x} - \frac{\partial H_x}{\partial y}\right) \tag{3.9}$$

$$\frac{\partial H_x}{\partial t} = \frac{1}{\sqrt{\varepsilon_o \mu_o}}\left(\frac{\partial \tilde{E}_y}{\partial z} - \frac{\partial \tilde{E}_z}{\partial y}\right) \tag{3.10}$$

$$\frac{\partial H_y}{\partial t} = \frac{1}{\sqrt{\varepsilon_o \mu_o}}\left(\frac{\partial \tilde{E}_z}{\partial x} - \frac{\partial \tilde{E}_x}{\partial z}\right) \tag{3.11}$$

$$\frac{\partial H_z}{\partial t} = \frac{1}{\sqrt{\varepsilon_o \mu_o}}\left(\frac{\partial \tilde{E}_x}{\partial y} - \frac{\partial \tilde{E}_y}{\partial x}\right) \tag{3.12}$$

Using the electric field obtained by electromagnetic analysis, the power absorbed from the electromagnetic microwaves per unit volume of egg was calculated by specifying different input power densities of 1, 2 and 3 W g^{-1} using the following equation:

$$P(x, y, z, t) = \frac{\omega \varepsilon_0 \varepsilon'' \left|E\right|^2}{2} \tag{3.13}$$

3.3.2 Heat Transfer Model for Heat Generation

As the source term temperatures inside the egg were obtained, the heat transfer equation (Eq. 3.14) was solved with the calculated power loss added.

$$\rho C_p \frac{\partial T}{\partial t} = k\nabla^2 T + P(x, y, z, t) \tag{4.14}$$

Where ρ =1031 and 1126 $kg.m^{-3}$

C_p= 3.58 and 2.77 $J.kg^{-1}.K^{-1}$

k = 0.550 and 0.389 $W.m^{-1}.K^{-1}$ for egg white and egg yolk respectively

(Coimbra et al, 2006)

3.3.3 Computer Simulation of Microwave Heating of an Egg

3.3.3.1 Material Models & Boundary Conditions

First the material properties were specified. The model consisted of the oven cavity, the egg yolk and the egg albumen. The egg shell is almost transparent to the microwaves. However the dielectric properties of the shell were also taken into account for the simulation. Hence the model had three layers.

3.3.3.2 Permittivity

- For the cavity and waveguide the permittivity is taken as being the same as that of a vacuum.
- For egg white (Dev et al., 2008):
  - $\varepsilon' = 72.38 - 0.17T - 1.75f$ and $\varepsilon'' = 17.22 - 0.19T + 1.58f$
- For egg yolk (Dev et al., 2008):
  - $\varepsilon' = 50.08 - 0.13T - 1.72f$ and $\varepsilon'' = 13.55 - 0.11T + 0.65f$

where, T is the temperature in °C, and f is the frequency in GHz. The initial (room) temperature was taken as 22°C and the frequency of operation was taken as 2.450 GHz. Figure 3.1 shows the geometry and dimensions of the cavity and waveguide with the position of egg in it and Figure 3.2 along with Table 3.1 detail the dimensions of the typical egg used for simulation studies.

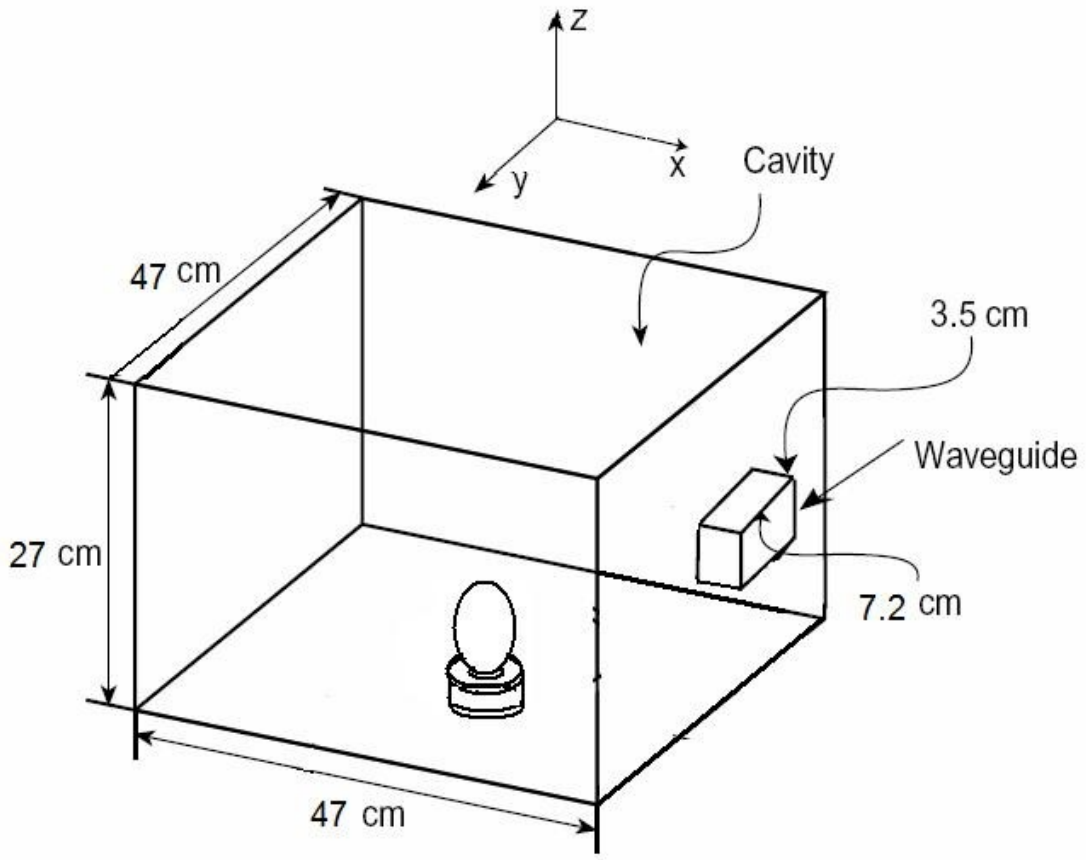

Figure 3.1: Egg in the Microwave Cavity

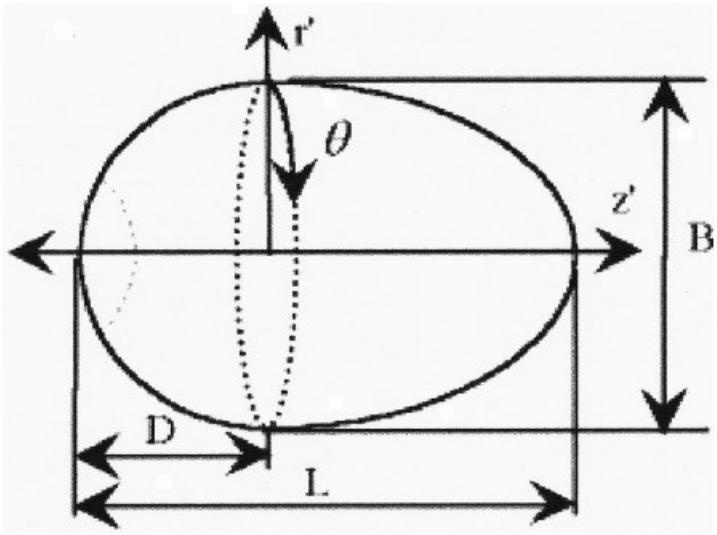

Figure 3.2: Egg geometry

Table 3.1: Dimensions of the egg

Description	Symbol	Value
Thickness of shell	h_s	1 mm
Length of shell	L	5.7 cm
Largest breadth of shell	B	4.2 cm
Distance from largest breadth to blunt end	D	2.5 cm
Radius of spherical shell	a	2.3 cm
Radius of yolk/embryo	a_e	1.5 cm
Area of contact	A_R	0.15 cm^2

3.3.3.3 Boundary Conditions and Excitations (Loads)

The walls of our cavity were specified as perfect electrical conductors (Figure 3.3). An exterior waveguide port (Figure 3.4) was used for the excitation of the port.

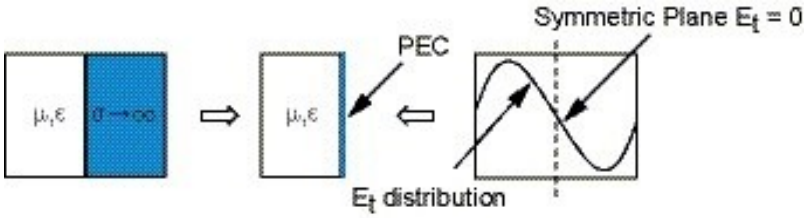

Figure 3.3: Perfect Electrical Conductor

Figure 3.4: Exterior waveguide port excitation

The excitation port option was specified in two steps. First the solid model area was selected to define the port location and assign a port number. The port number assigned must be between 1 and 50. For an exterior port, after assigning the port number, the port type was specified as a rectangular waveguide of TE_{10} (Transverse electric) mode.

The computational geometry of the egg was staggered with rectangular elements. A simplified self-explanatory logical flow diagram of the entire simulation process is given in Figure 3.5.

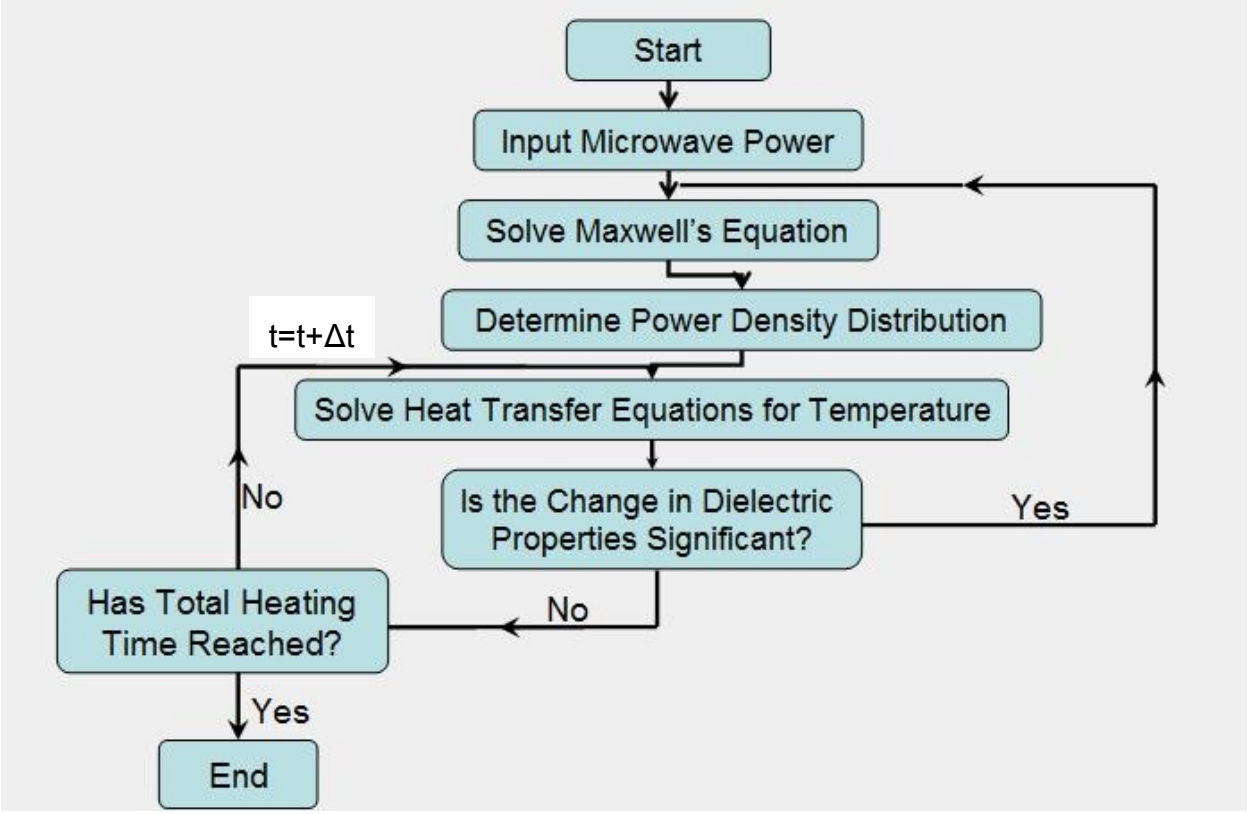

Figure 3.5: Flow diagram of the FDTD simulation process

3.3.4 Innovations in the simulation process

The following factors were taken into account for the first time in the simulation of microwave heating of a heterogeneous lossy dielectric medium.

- A coupled approach was used for the temperature and power distribution
- The dependency of dielectric properties on temperature of the material and frequency of the microwaves was taken into account.
- A microwave heat generation factor was introduced
- The conduction, convection and radiation modes of heat transfer (though negligible) were included.

3.3.5 Evaluation of the simulation

The simulations were qualitatively assessed for their validity by microwave heating of commercially available large sized grade A eggs and their quick freezing liquid nitrogen. A regular domestic microwave oven (Panasonic Model NNSN968B, Panasonic Inc, Canada) was used. Temperature was measured at two point (one in broad end and one in the narrow end of the egg). Quick freezing preserved the location of the coagulation and the egg retained its shape even after peeling off the shell. This also provided the required transparency to determine the position and size of the coagulation inside the egg white, which in turn indicated the region(s) of overheating.

3.4 Results & Discussion

3.4.1 Electric field Intensity

Figures 3.6, 3.7 and 3.8 depict the distribution of electric field at the central transverse section of an egg stratum in the cavity for the power densities of 1, 2 and 3 W g^{-1} respectively. These clearly indicate that the pattern of the electric field distribution was not affected by a change in power density, but that electric field intensity varied exponentially with a linear increase in the power density. These Figures (3.6, 3.7 and 3.8) also clearly indicate that the distribution of the electric field is strongly extensively affected by the presence of the egg inside the cavity, which can be seen from the drastically decreased electric field intensity immediately adjacent the egg in the cavity.

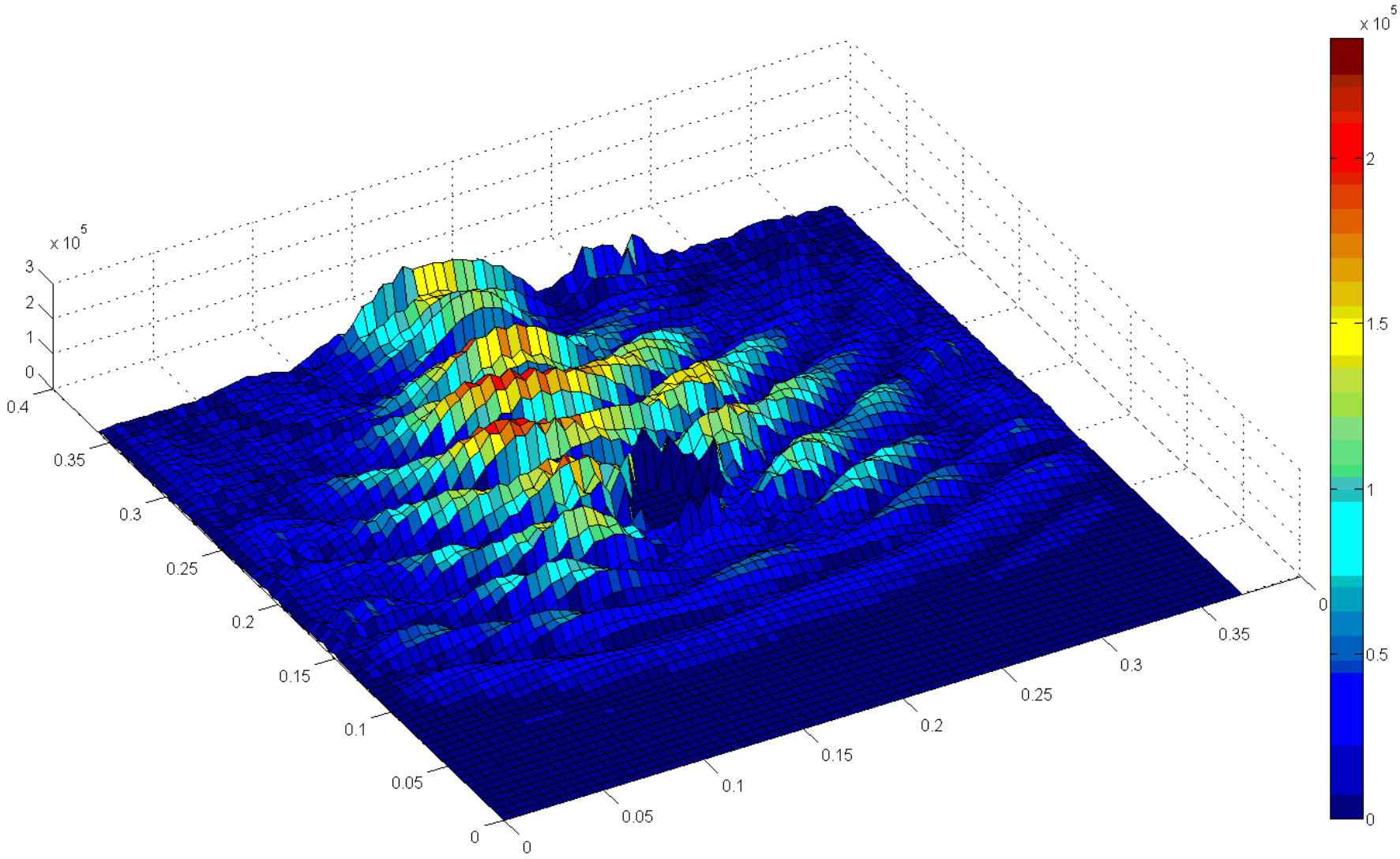

Figure 3.6: Distribution of electric field (V m^{-1}) at the central transverse section of an egg stratum in the cavity for 1 W g^{-1}

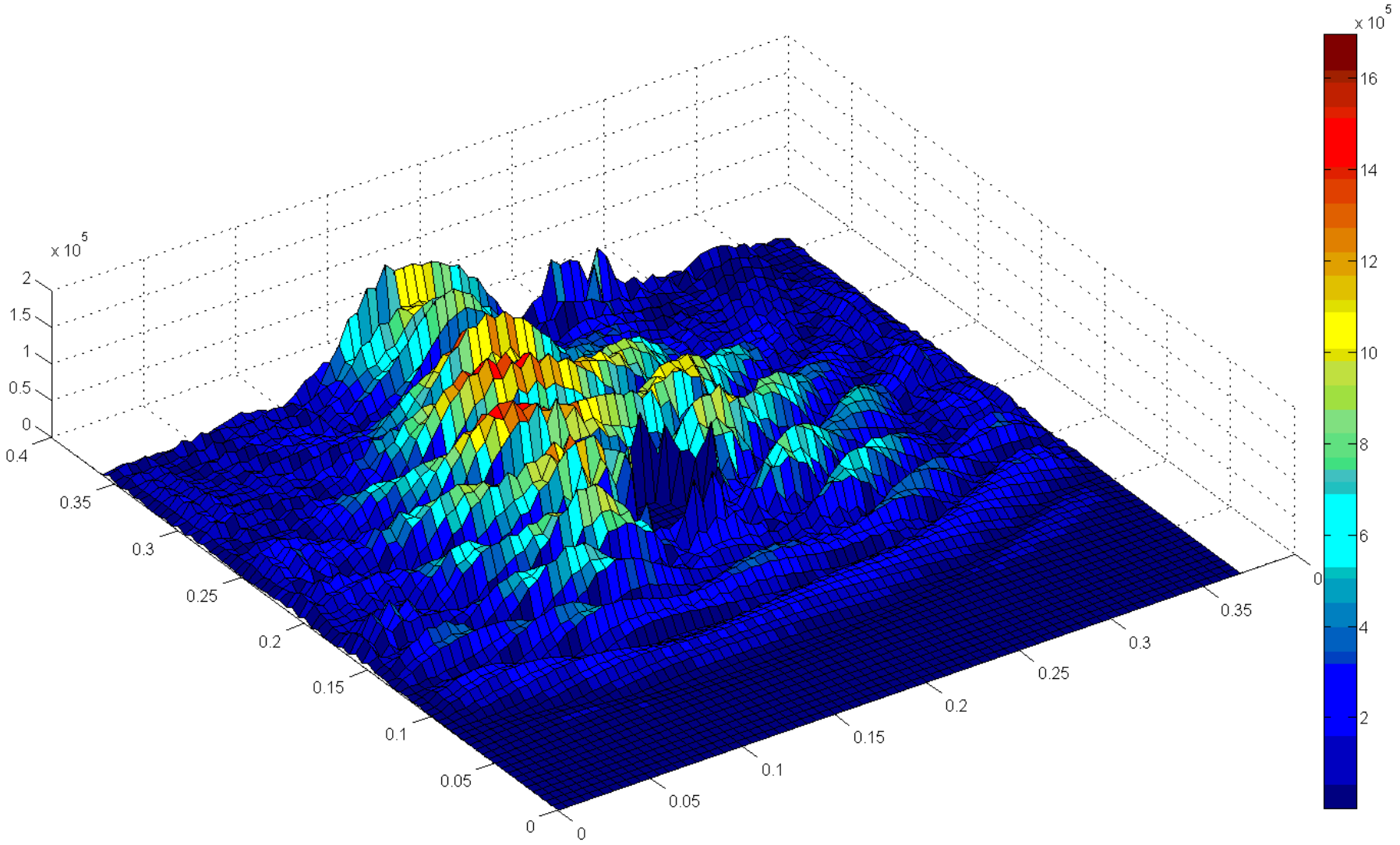

Figure 3.7: Distribution of electric field (V m^{-1}) at the central transverse section of an egg stratum in the cavity for 2 W g^{-1}

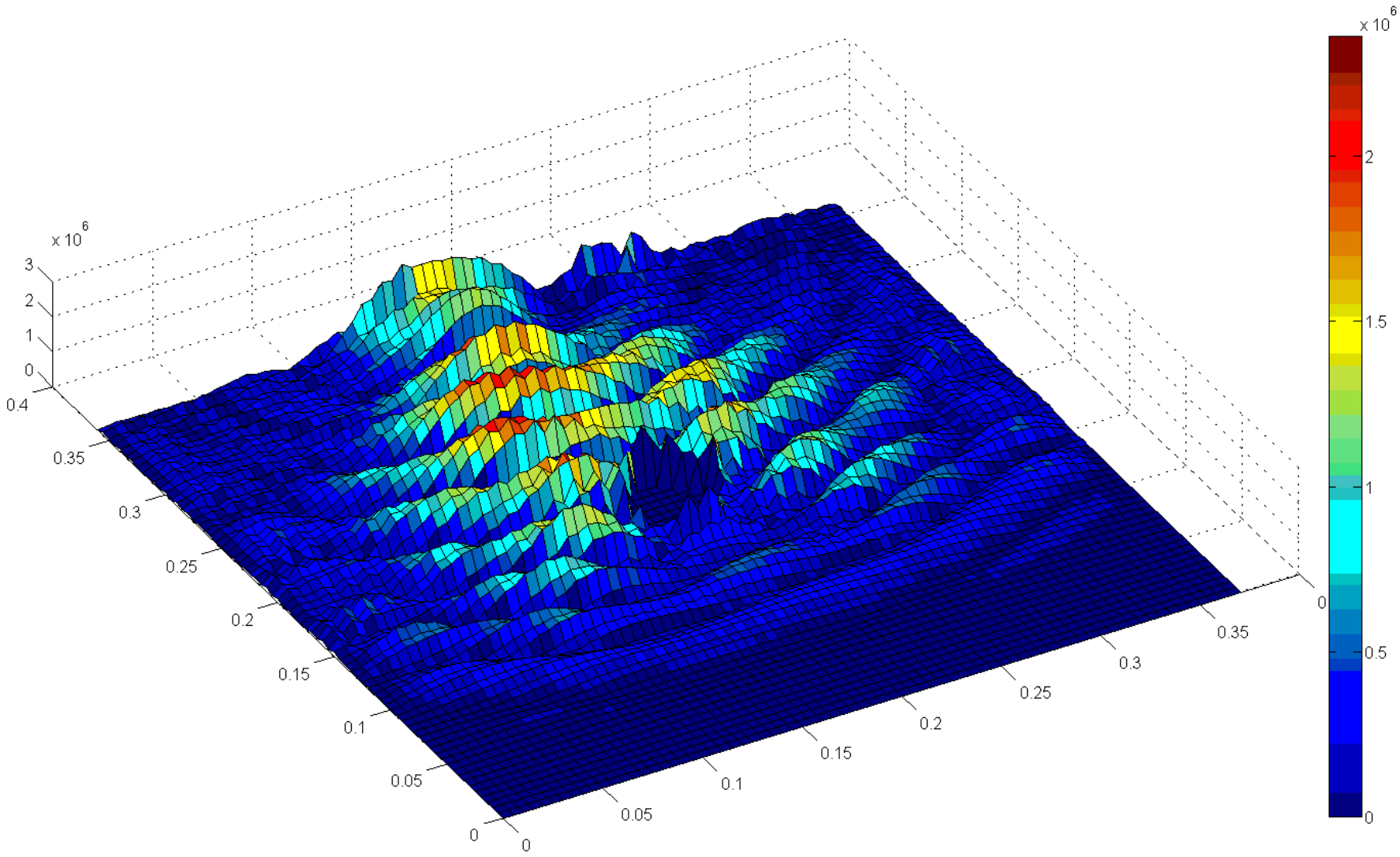

Figure 3.8: Distribution of electric field (V m^{-1}) at the central transverse section of an egg stratum in the cavity for 3 W g^{-1}

3.4.2 Power Loss inside the egg leading to heat generation

Power loss is the primary factor contributing to the microwave heating of the lossy dielectric media. Figures 3.9, 3.10 and 3.11 denote the power loss in watts inside the egg at the central transverse section for power densities of 1, 2 and 3 W g^{-1} respectively. (The images are rotated by 180 degrees to present a better view). The power loss increased linearly with the linear increase in power density, indicating that for a stationary (non-rotating) egg in a microwave cavity the power loss was at its maximum at the point of incidence and that the microwaves soon dissipated most of their energy.

Fu and Metexas (1994) and Dai (2006) followed similar approach to estimate the electric field intensity and power loss in a multimode cavity, whereas their models did not take into consideration a heterogeneous substance like the shell eggs and their thermodynamically changing properties.

3.4.3 Temperature distribution

The power distribution inside the egg was directly related to the extent of heating at a particular location inside the egg. Figures 3.12, 3.13 and 3.14 represent the simulated temperature values inside the egg at the central transverse section of the egg in °C for power densities of 1, 2 and 3 W g^{-1}, respectively.

The heating pattern of the eggs clearly had the same non-uniformity as that of the power loss, shedding some light on the extent and pattern of overheating and non-uniform temperatures inside the egg. The temperature gradient within the egg increased drastically with an increase in power level, as can be seen by comparing values in plots generated for increasing power levels.

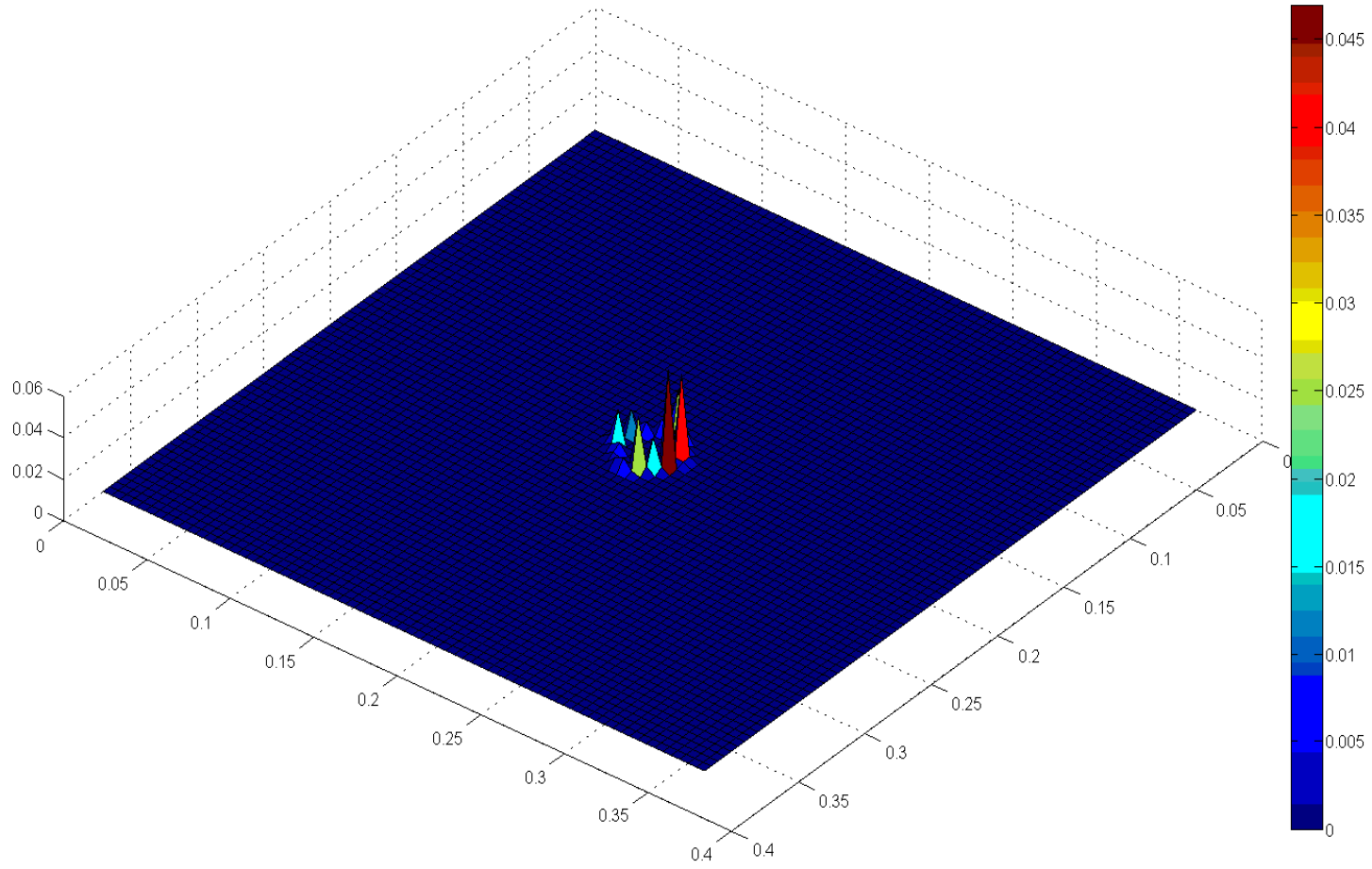

Figure 3.9: Power loss (W) inside the egg at the central transverse section for 1 W g^{-1}

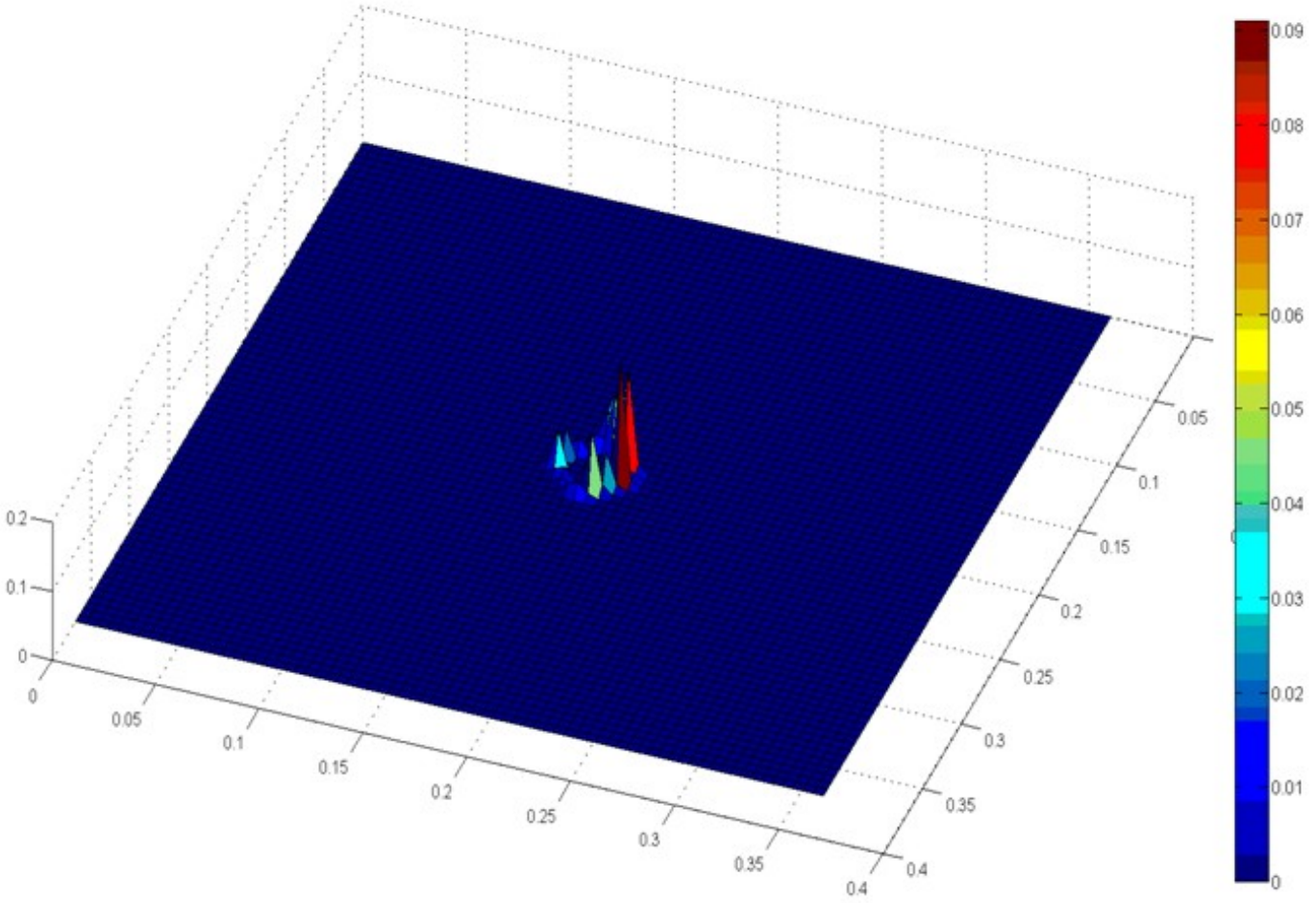

Figure 3.10: Power loss (W) inside the egg at the central transverse section for 2 W g^{-1}

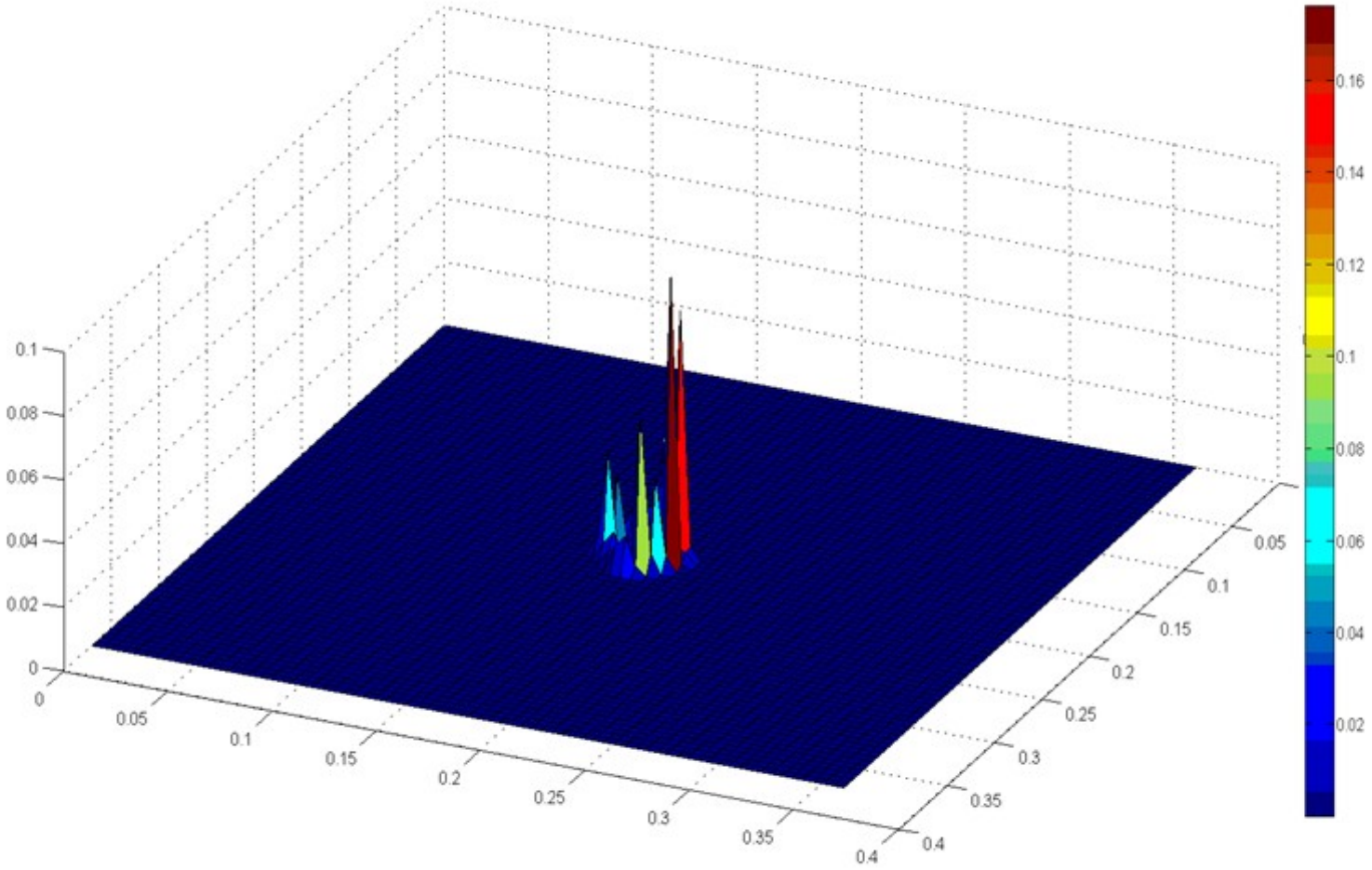

Figure 3.11: Power loss (W) inside the egg at the central transverse section for 3 W g^{-1}

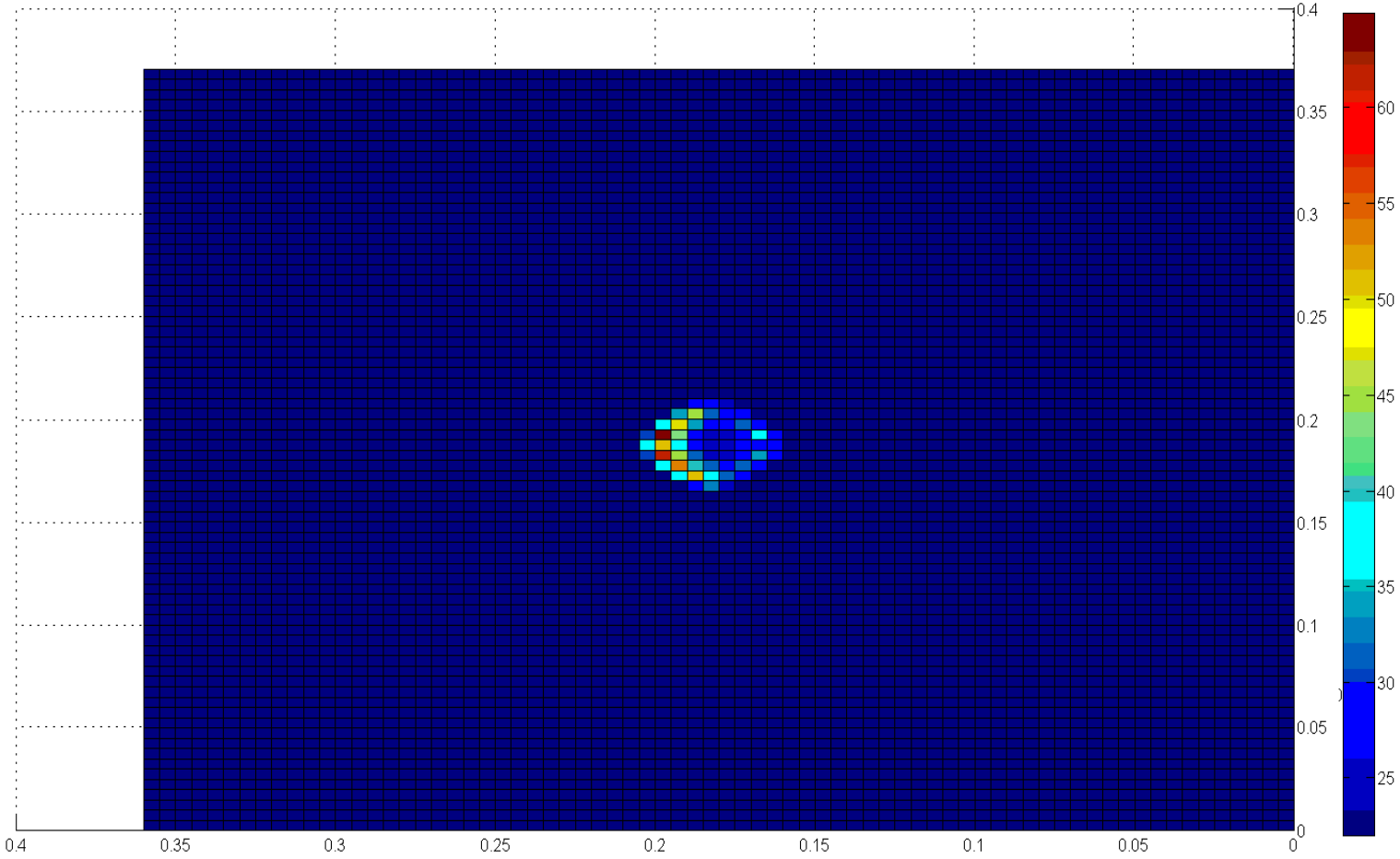

Figure 3.12: 2D -Temperature distribution (°C) inside the egg at the central transverse section for 1 W g^{-1}

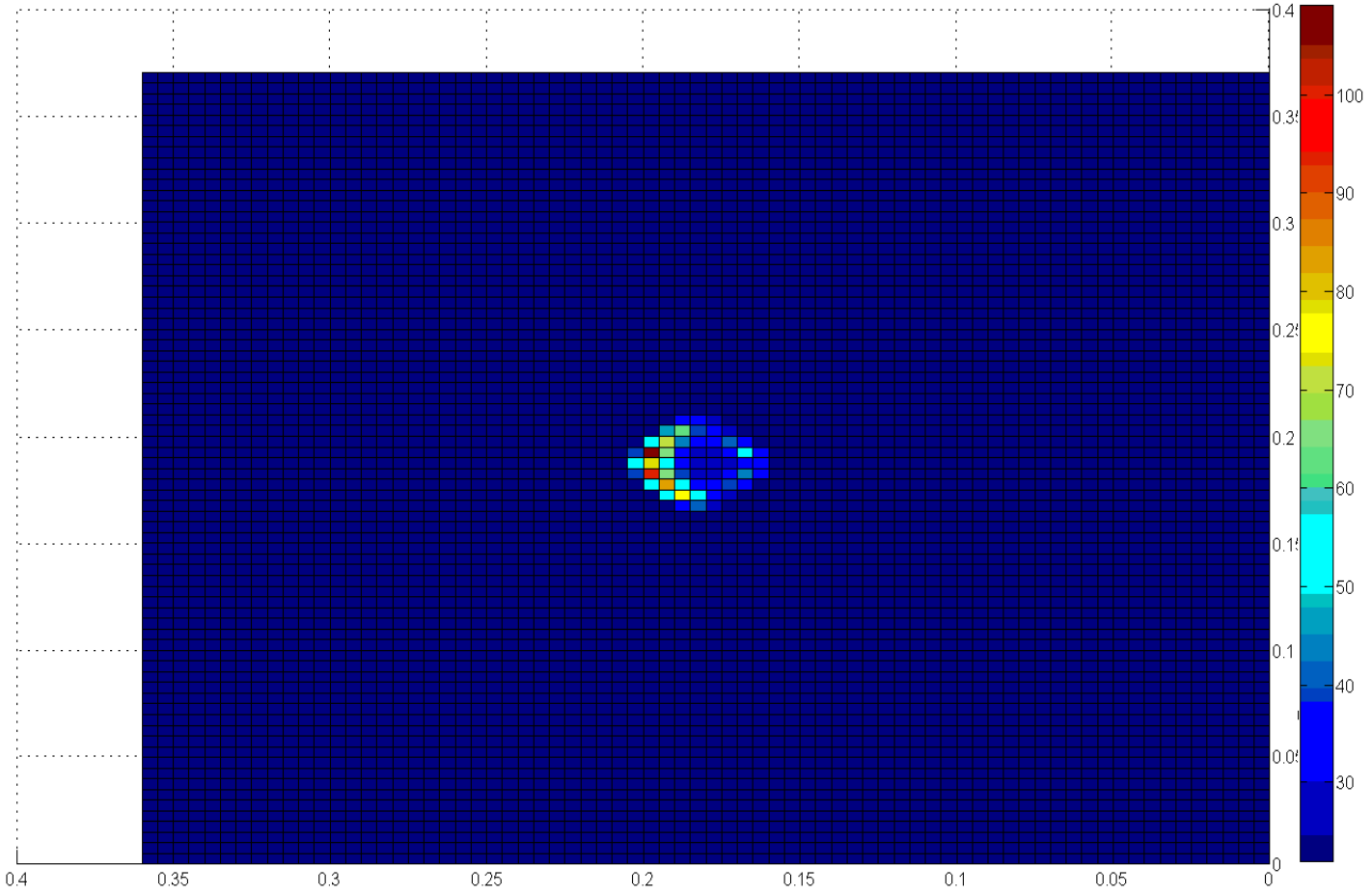

Figure 3.13: 2D -Temperature distribution (°C) inside the egg at the central transverse section for 2 W g^{-1}

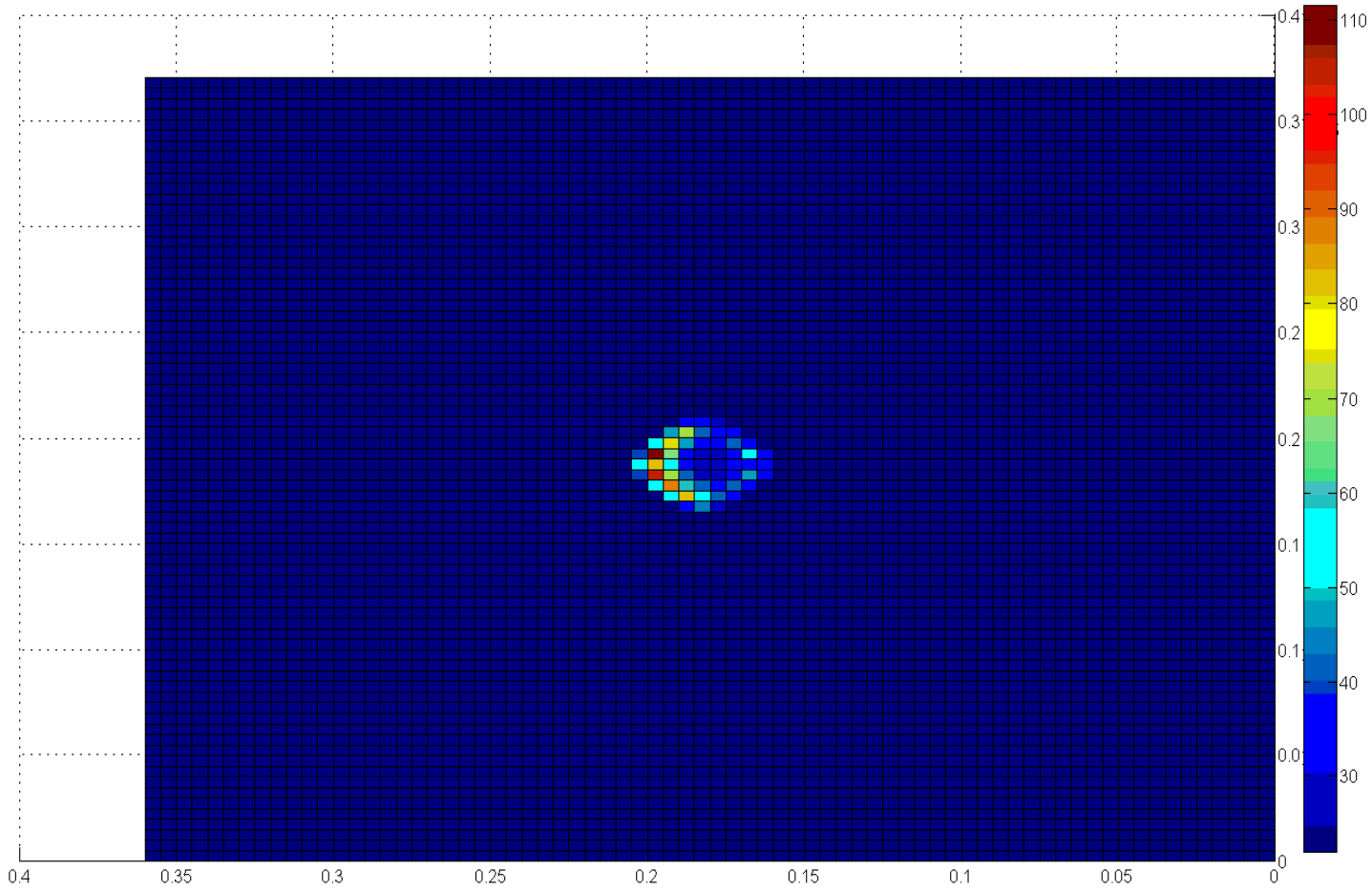

Figure 3.14: 2D -Temperature distribution (°C) inside the egg at the central transverse section for 3 W g^{-1}

The simulations showed that at some locations the temperature shot up to 110°C within a 100 seconds at a power density of 3 W g^{-1}, while other portions of the egg that remained below 30°C. Comparatively, at lower power densities the temperature gradient was much lesser due to the slower heating rates and greater quantities of heat transfer occurring through conduction. Harms et al. (1996) obtained results with good precision by the variability of the validation data was considerably high leading to 15 % error in estimation. As this model involves simultaneous solutions for all the three modes of heat transfer, the error was kept within 10% for all the measured values.

3.4.4 Experimental Validation:

Actual microwave-heated eggs showed coagulation of egg white exactly in the regions predicted by the simulation, indicating an excellent corroboration of the experimental results by those of simulated microwave heating (Figure 3.15).

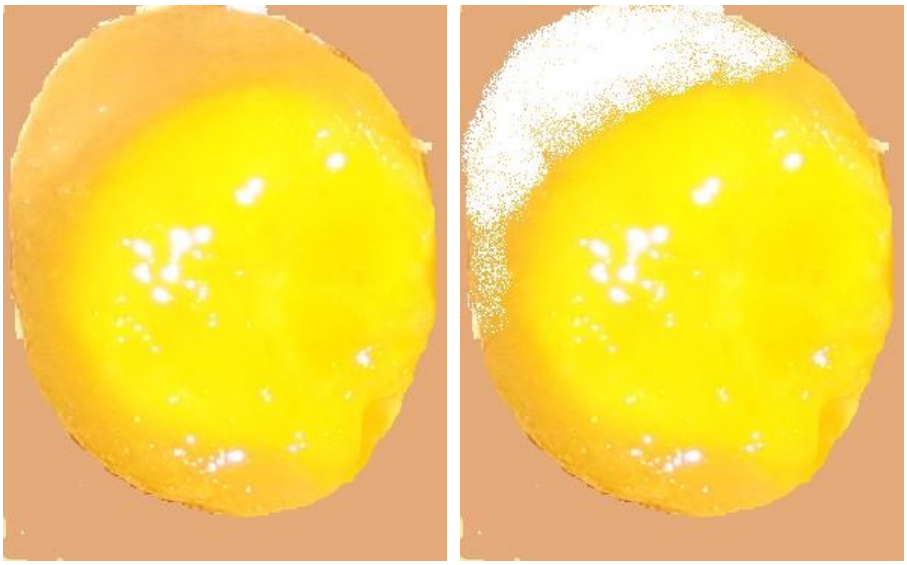

Figure 3.15: Quick frozen Microwave heated egg showing coagulation of egg white (right hand side) compared to the control (left hand side)

At the higher power levels the eggs exploded quickly in the microwave indicating that the temperature had risen above the boiling point in certain parts of the egg. Such an explosion can be avoided by the slower heating of the egg which occurs under lower power densities.

3.5 Conclusions

Results of the actual microwave heating and numerical simulations corroborate each other very well, thereby confirming the accuracy of this approach in simulating novel wave guide wave guide and/or microwave cavity designs. The non-uniformity of heating was more pronounced at higher power levels, thereby suggesting lower power levels to be better in producing a quality pasteurized product.

Thus, “microwave heating is a viable alternative for the pasteurization of in-shell eggs.”

3.6 Recommendations for Further research

FDTD Simulation of a rotating egg inside a microwave cavity can help better understanding of the heating behaviour of rotating objects. Also Finite Element Modeling and Simulation of the microwave heating of in-shell eggs would bring about better and more accurate simulation results. Hyperspectral imaging of the coagulation of egg protein due to microwave heating would reveal any non-uniform heating with greater discrimination.

3.7 Acknowledgements

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3.8 References

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Connecting text

The results obtained by the Finite Difference Time Domain (FDTD) method were satisfactory, but, due to the unique shape of hen's eggs this model resulted in staggered edges. In order to design a specific type of microwave applicator, we need adaptive meshes and variable size elements which will give precision and accuracy. Therefore modelling using finite element method (FEM) was considered.

The most attractive feature of FEM is its ability to handle complex geometries (and boundaries) with relative ease. While FDTD, in its basic form, is restricted to handle rectangular shapes and simple alterations thereof, the handling of geometries in FEM is theoretically straightforward. There are reasons to consider the mathematical foundation of the finite element approximation more sound, because, for instance, the quality of the approximation between grid points is poor in FDTD, and so, the quality of a FEM approximation is often higher than the corresponding FDTD approach. Also process optimization needs to be carried out using validated simulation approaches before fabrication.

Chapter 4

OPTIMIZATION OF MICROWAVE HEATING OF IN-SHELL EGGS THROUGH FINITE ELEMENT MODELING AND EXPERIMENTAL TRIALS

4.1 Abstract

Considering microwave heating as a viable alternative for in-shell pasteurization of eggs, after the simulation of the microwave-heating process using an FEM model, process optimization was carried out to determine the most effective procedure and design for the process. The varying parameters obtained by using different modelling techniques for microwave heating of in-shell eggs, were optimized using MATLAB. Laboratory-scale experimental trials were conducted to test the validity and effectiveness of the optimized parameters. The optimal parameters set forth were found to be more efficient in terms of heating time and uniformity. Microwave heating appeared to be a viable alternative for the pasteurization of in-shell eggs.

Keywords. Microwave Pasteurization, Finite Element Modelling, Optimization

4.2 Introduction

Eggs are potential hosts and carriers for pathogenic microbes like *Salmonella enteritidis* and the most deadly strain (H5N1) of the avian flu virus. Heat pasteurization is well known to enhance food safety (FSIS-USDA, 2006).

Egg is an exceptional nutritional supplement. It is a good source of vitamin A, B3 and folate. It also contains useful amounts of many other vitamins and

minerals (Li-Chan, Powrie, & Nakai, 1995). Egg is an essential ingredient in several foods, especially given their exceptional functional properties. But egg proteins are extremely heat sensitive. Therefore heat pasteurization with minimal changes to the egg proteins needs consideration.

Eggs are mostly marketed raw and frequently consumed raw especially in North America. More than 90% of food borne Salmonellosis, caused by *Salmonella enteritidis*, occurs through shell eggs (Schroeder et al., 2005). Most Salmonella outbreaks generally involved grade A eggs that were washed and disinfected and also met quality requirements regarding the state of their shell (St. Louis, Morse, & Potter 1988).

Conventional methods of heat pasteurization using hot air or hot water severely affect the functional quality of eggs. Protein denaturation is a complex process which primarily depends on the time-temperature combination in the heating process. Therefore denaturation can be reduced by rapid heating. Considering microwaves as a viable alternative for the pasteurization of in-shell eggs, preliminary trials confirmed that microwaves at 2450 MHz could be successfully used to raise the temperature of in-shell eggs to the required pasteurization temperatures (57.5°C for egg white and 61.1°C for yolk) (FSIS-USDA, 2006) within a few minutes (Dev et al, 2008).

Microwave heating exploits the dielectric behaviour of the substance exposed to it, to generate heat from within the substance. But this direct heat generation occurs only up to a certain depth from the surface of the product. Depending on the dielectric properties of the substance, there is an exponential decay of microwave energy as the waves penetrate into the product from the surface (Meda, Orsat, & Raghavan, 2005). Beyond a certain depth it is the

conductive and/or convective form of heat transfer (based on the state of the substance) that heats the rest of the material. Also the dielectric behaviour of the biological materials vary considerably with temperature and frequency. Dev *et al.* (2008) derived linear mathematical models that can be used to predict these changes for the egg components.

At the macroscopic level electromagnetic phenomena are defined using Maxwell's equations. The electromagnetic field distribution inside the microwave oven can be traced out by solving these Maxwell's equations. Finite Element Method (FEM) is commonly used for solving Maxwell's equations to get the energy distribution in a complex object or within a multimode cavity and it is capable of simulating power density distribution in 3-D space. (Fu and Metaxas, 1994; Harms, Chen, Mittra *et al.* 1996; Meredith, 1994; Zhou, Puri, Anantheswaran *et al.*,1995).

FEM is based on reducing a complex problem into a solution with a large number of simple problems. The FEM technique competes very favourably with other numerical methods as it is based on reducing the Maxwell's equations to a system of simultaneous algebraic linear equations (Delisle, Wu & Litva, 1991). FEM can readily model heterogeneous and anisotropic materials as well as arbitrarily shaped geometries. It can also provide both time and frequency domain analyses, which are important with respect to microwave heating issues like field distribution, scattering parameters and dissipated power distribution for various materials and geometries (Dai, 2006).

Lin et al. (1989) studied the sensitivity of microwave heating to variations in thermal diffusivity, dielectric properties and incident microwave power. Oliveira *et al.* (2002) simulated microwave heating, taking into account the transient heat

transfer during heating. However, neither of these studies took into account the effect of geometry and orientation. Furthermore, simulation studies available in the literature seldom consider the dynamic change in dielectric properties of the material exposed to microwaves with increases in temperature. Besides this, most of them are process- not product-specific and hence never account for the heterogeneity of the biological materials such as in-shell eggs.

A normal egg would explode during microwave (MW) heating due to water vapour pressure build-up inside the shell. Thus, when it comes to pasteurization, heating uniformity is a critical factor and microwave heating is fairly non uniform. Therefore the objective of this study was to develop a Finite Element Model and optimize the microwave heating process of in-shell eggs within the pasteurization temperature limits, through simulations and experimental trials, taking into account

- the continuously changing dielectric properties with temperature,
- the heterogeneous composition of the egg and
- the complex geometry of the egg.

Optimization is a mathematical programming method to minimize or maximize a real function by systematically choosing the values of real or integer variables from within an allowed set. In this study the parameters of power density (which affects the total heating time), waveguide position and waveguide orientation were optimized, with uniformity being the criterion for process effectiveness, given the highly uneven heating which can occur when using microwaves.

4.4 Materials and Methods

4.4.1 Simulation

A Finite Element Model was developed using COMSOL Multiphysics version 3.4 (COMSOL Inc., USA) and MATLAB R2008a software packages to simulate the MW heating process within the pasteurization temperature limits for three different power densities (1, 2 and 3 W g^{-1}), two different waveguide positions (one simulated and verified and the second one only simulated) and with and without rotation in both the custom built laboratory microwave setup and the regular domestic microwave oven. Figure 4.1 shows the flow diagram of the simulation technique.

A custom-built computer with Intel Core 2 Quad 2.4 GHz processor and 8 GB primary memory was used to run the simulations. The Bilateral symmetry of the cavity and waveguide was taken advantage of for the simulations, thereby greatly reducing the resources required for running these simulations.

4.4.2 Mathematical Model

4.4.2.1 Electromagnetics

The Maxwell's equations that govern the electromagnetic phenomena evolving in a given configuration resolved in 3D space are given by equations (1)-(6) (Dai, 2006).

$$\frac{\partial E_x}{\partial t} = \frac{1}{\varepsilon_0 \varepsilon'}\left(\frac{\partial H_z}{\partial y} - \frac{\partial H_y}{\partial z}\right) - \frac{2\pi f \varepsilon''}{\varepsilon'} E_x \tag{4.1}$$

$$\frac{\partial E_y}{\partial t} = \frac{1}{\varepsilon_0 \varepsilon'}\left(\frac{\partial H_x}{\partial z} - \frac{\partial H_z}{\partial x}\right) - \frac{2\pi f \varepsilon''}{\varepsilon'} E_y \tag{4.2}$$

$$\frac{\partial E_z}{\partial t} = \frac{1}{\varepsilon_0 \varepsilon'}\left(\frac{\partial H_y}{\partial x} - \frac{\partial H_x}{\partial y}\right) - \frac{2\pi f \varepsilon''}{\varepsilon'} E_z \tag{4.3}$$

$$\frac{\partial H_x}{\partial t} = \frac{1}{\mu_0}\left(\frac{\partial E_y}{\partial z} - \frac{\partial E_z}{\partial y}\right) \quad (4.4)$$

$$\frac{\partial H_y}{\partial t} = \frac{1}{\mu_0}\left(\frac{\partial E_z}{\partial x} - \frac{\partial E_x}{\partial z}\right) \quad (4.5)$$

$$\frac{\partial H_z}{\partial t} = \frac{1}{\mu_0}\left(\frac{\partial E_x}{\partial y} - \frac{\partial E_y}{\partial x}\right) \quad (4.6)$$

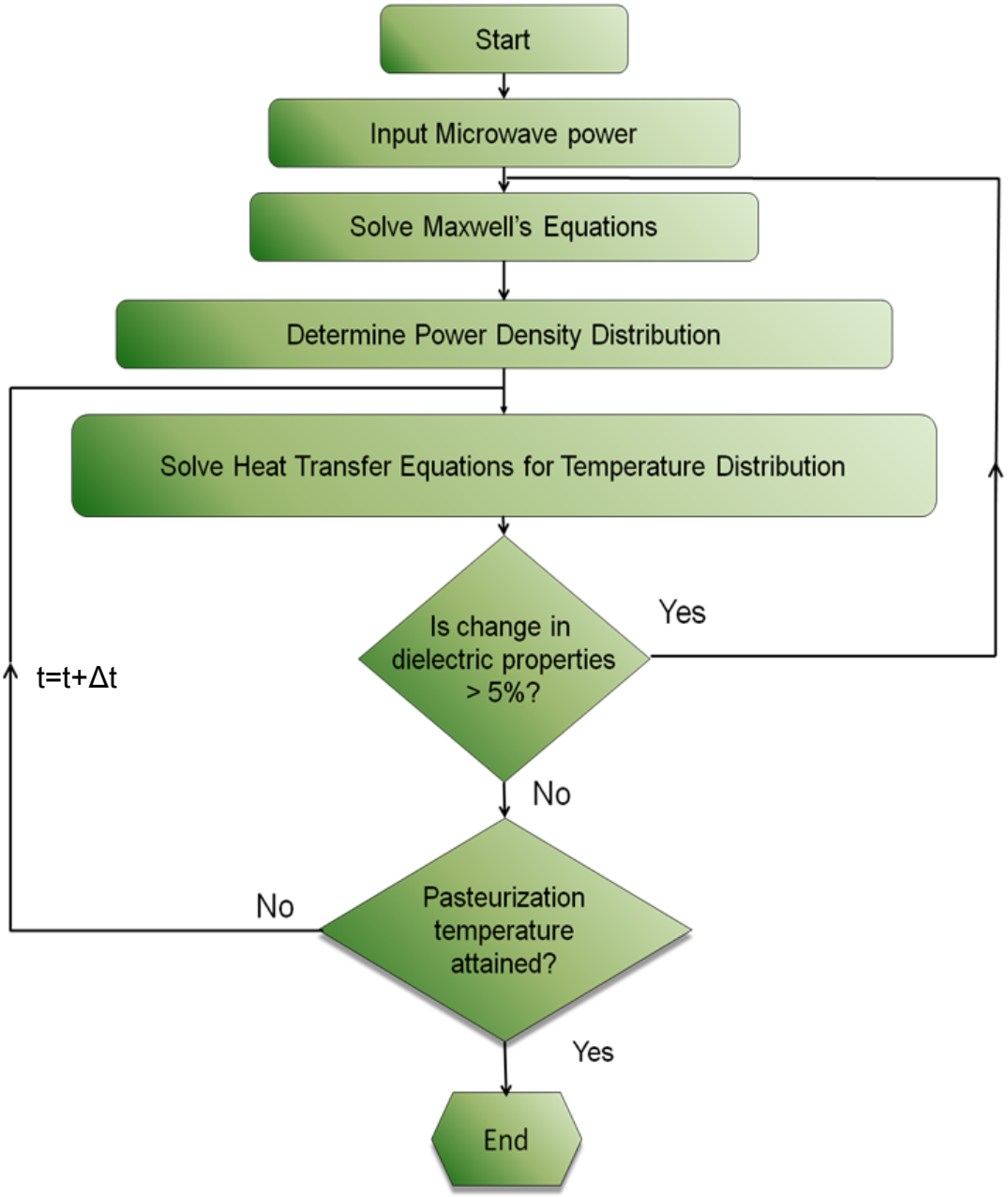


Figure 4.1 Flow Diagram of FEM Simulation Technique

The dynamically changing dielectric constant and loss factor were calculated using equations 4.7 and 4.10, (Dev *et al.* 2008) modified to SI units.

- For egg white:
  - $\varepsilon' = 72.38 - 0.17\,(T_c + 273) - 1.75(f \times 10^9)$ (4.7)
  - $\varepsilon'' = 17.22 - 0.19\,(T_c + 273) + 1.58\,(f \times 10^9)$ (4.8)

- for egg yolk:
  - $\varepsilon' = 50.085 - 0.13\,(T_c + 273) - 1.72\,(f \times 10^9)$ (4.9)
  - $\varepsilon'' = 13.55 - 0.11\,(T_c + 273) + 0.65\,(f \times 10^9)$ (4.10)

Constant values of 3.5 and 0.5 were taken for the ε' and ε" values of both the shell and shell membrane (Dev *et al.,* 2008).

The time average power dissipated in each element in a dielectric material was obtained by integrating the Poynting vector over the closed surface S for each tetrahedral element: (Jia and Jolly, 1992).

$$P_{av} = -\frac{1}{2}\int_S P_c.dS \tag{4.11}$$

where $P_c = E \times H$

Volumetric heat generation Q can be expressed in terms of power intensity in three orthogonal directions: (Lin *et al.*, 1989).

$$Q = \frac{\partial P_{av(x)}}{\partial V} + \frac{\partial P_{av(y)}}{\partial V} + \frac{\partial P_{av(z)}}{\partial V} \tag{4.12}$$

4.4.2.2 Boundary conditions

Perfect Electrical Conductor boundary conditions (n x E = 0) were used for the walls of the cavity and Perfect Magnetic Conductor boundary condition (n x H = 0) was used for the symmetry boundaries (Fu et al. 1994).

Boundary conditions at the port were as follows:

$$H_y = ACos(\frac{\pi x}{\alpha})\, Cos(\omega t + \beta y) \tag{4.13}$$

$$E_z = (\omega \mu_0 \frac{\alpha}{\pi}) ASin(\frac{\pi x}{\alpha})\, Sin(\omega t + \beta y) \tag{4.14}$$

$$H_x = (\frac{\beta \alpha}{\pi}) ASin(\frac{\pi x}{\alpha}) Sin(\omega t + \beta y) \tag{4.15}$$

4.4.2.3 Heat transfer

As the pasteurization temperatures are relatively low to build significant amount of pressure, constant pressure conditions were assumed. For an incompressible food material heated under constant pressure, the thermal energy equation is given by equation (4.16) (Zhou *et al.*,1995)

$$\rho C_p \frac{\partial T}{\partial t} = \nabla \cdot (K \nabla T) + Q \tag{4.16}$$

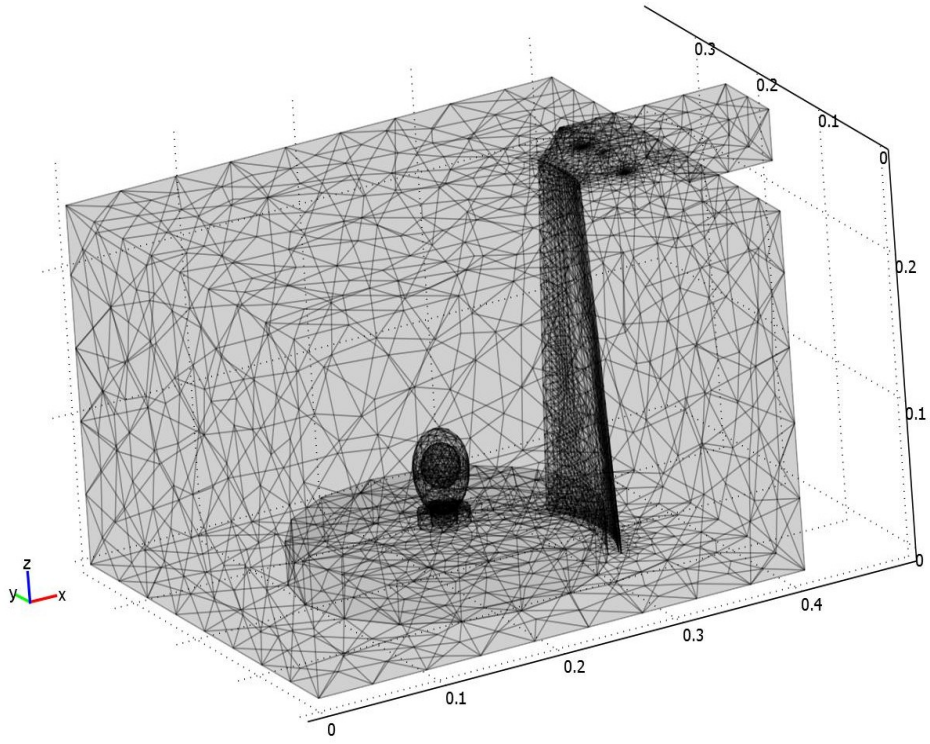
Different mesh element sizes were used for different sub-domains based on the dielectric properties of the sub-domain and the precision required in the sub-domain of interest. Also egg rotation was simulated by moving meshes with an angular velocity of $\frac{\pi}{18}$ rad s^{-1}, programmed using COMSOL Script version 1.2. Different configurations of microwave cavities, viz. a regular domestic microwave oven (Panasonic Model NNSN968B, Panasonic Inc, Canada) and a virtually

modified version of the same with a different wave guide position, the laboratory microwave setup fitted with a focussing shield and a virtually modified version of the same with a different wave guide position were simulated. Figures 4.2 and 4.3, give the FEM structure of the cavities with actual and virtually modified wave guide positions.

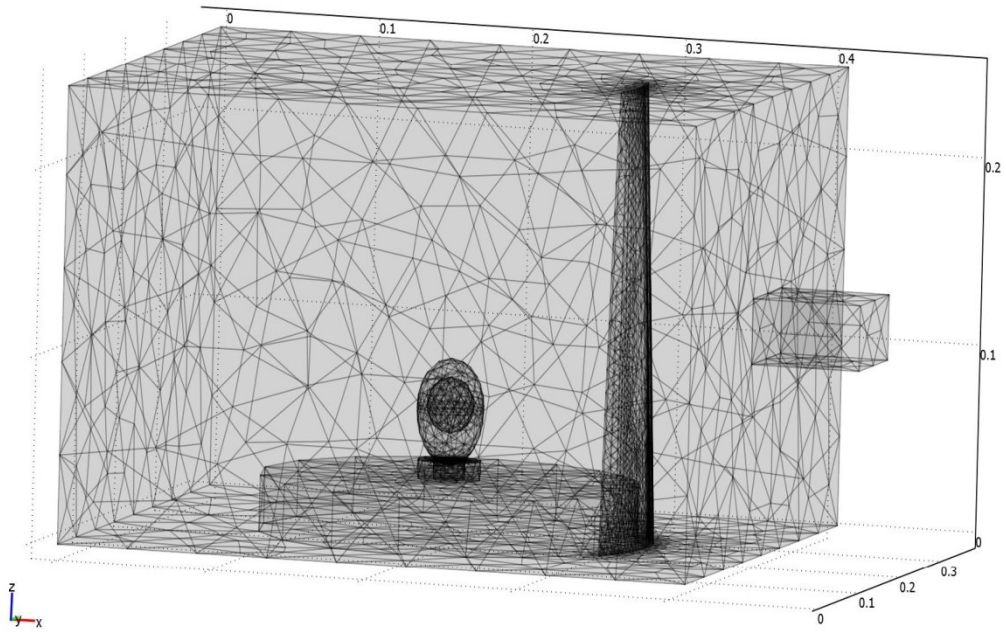
4.4.3 Experimental verification

Simulations were also computed for an all white egg in order to be able to verify the simulation approach with an artificial egg (a transparent glass egg made with real egg white). The simulation results were experimentally verified by heating the artificial egg in a custom built instrumented and computer-controlled laboratory-scale microwave (MW) oven (Figure 4.4) and in a regular domestic microwave oven (Panasonic Model NNSN968B, Panasonic Inc, Canada), with and without rotation, using three different power densities. The main components of the laboratory microwave oven were: a 2450 MHz microwave generator (Gold Star 2M214, South Korea) with power adjustable from 0 to 750 W, waveguides, a three-port circulator, a manual three stub tuner to match the load impedance, microwave couplers to measure forward and reflected power, a carbon load to absorb reflected power and a microwave cavity made of brass, (410 x 370 x 245 mm) in which the egg samples were processed. The wave guides were rectangular (70 x 35 mm) and TE10 mode of application was used.

The microwave generator (magnetron) produced microwaves with varying power densities based on the supplied power. The generated microwaves were guided using the waveguides into the microwave cavity via the above mentioned components in a sequence. A manual three-stub tuner was used to adjust the reflected power, thereby keeping it at the minimum possible value (<10% of the incident power).

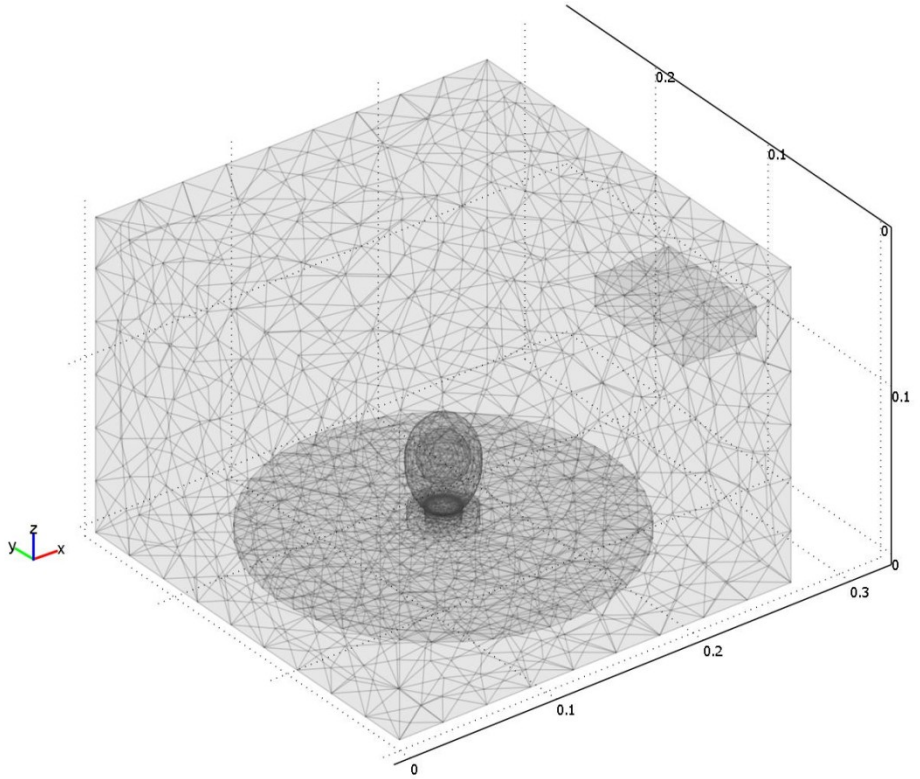


(a)

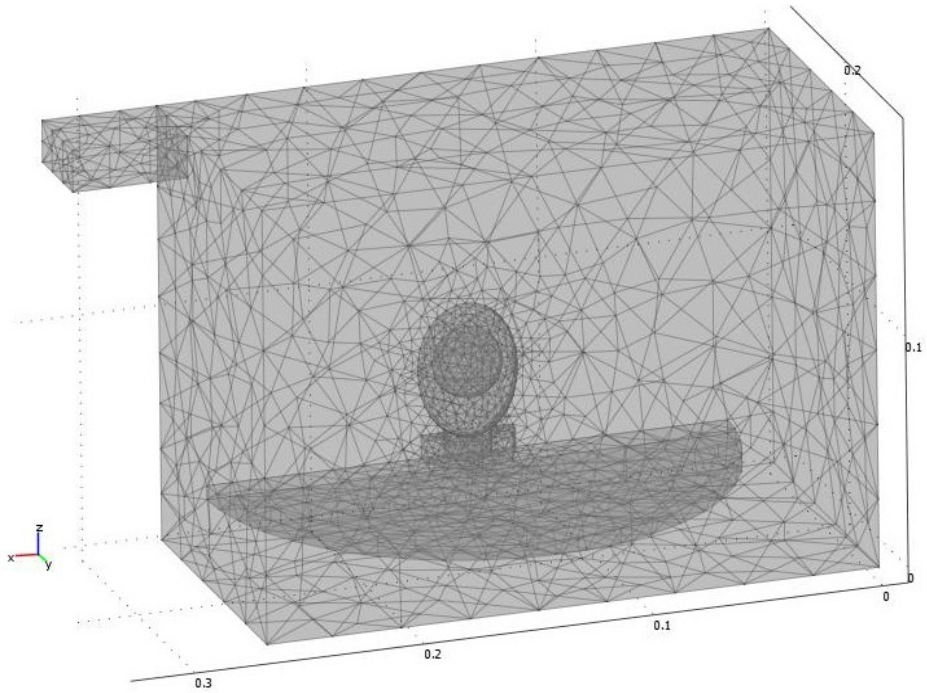


(b)

Figure 4.2 FEM structure of laboratory microwave cavity with turn table and focusing shield – (a) Actual laboratory configuration (b) Virtually modified configuration



(a)



(b)

Figure 4.3 FEM structure of regular domestic microwave oven with turn table – (a) Actual domestic microwave configuration (b) simulated configuration

Figure 4.4 Instrumented and computer controlled microwave (MW) oven

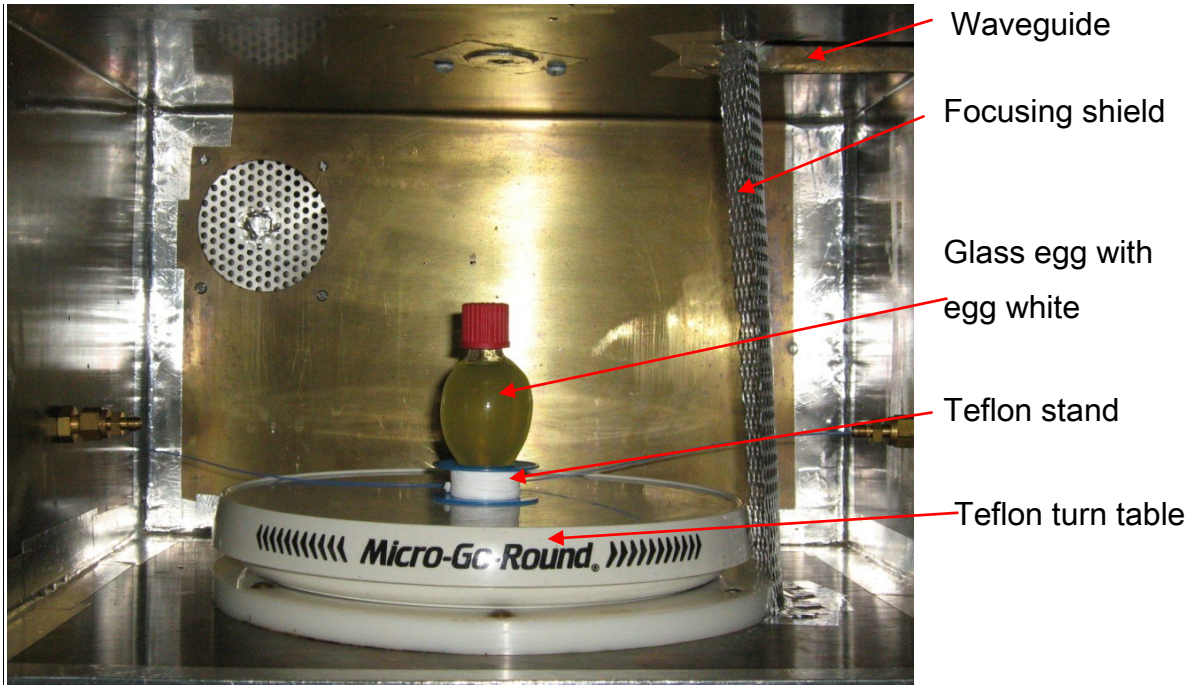


Figure. 4.5 Laboratory microwave cavity setup with artificial egg

The temperatures were measured using fibre optic probes (Nortech EMI-TS series, Quebec City, Canada). The probes were connected to a data acquisition unit (Agilent 34970A, Santa Clara, USA) which was itself connected to a computer. The entire setup was monitored and controlled using the HP VEE (Agilent, Santa Clara, USA) object oriented programming language.

In the laboratory microwave oven, a focusing shield was installed as shown in Figure 4.5. The focusing shield was nothing but a sector of a conical cylinder made of aluminum mesh. This reflects and diverges the microwaves near the port's entry point into the cavity, at the same time preventing the reflected waves from entering the waveguide and focusing the microwaves on the object of investigation. Figure 4.6 shows a schematic of the laboratory microwave setup.

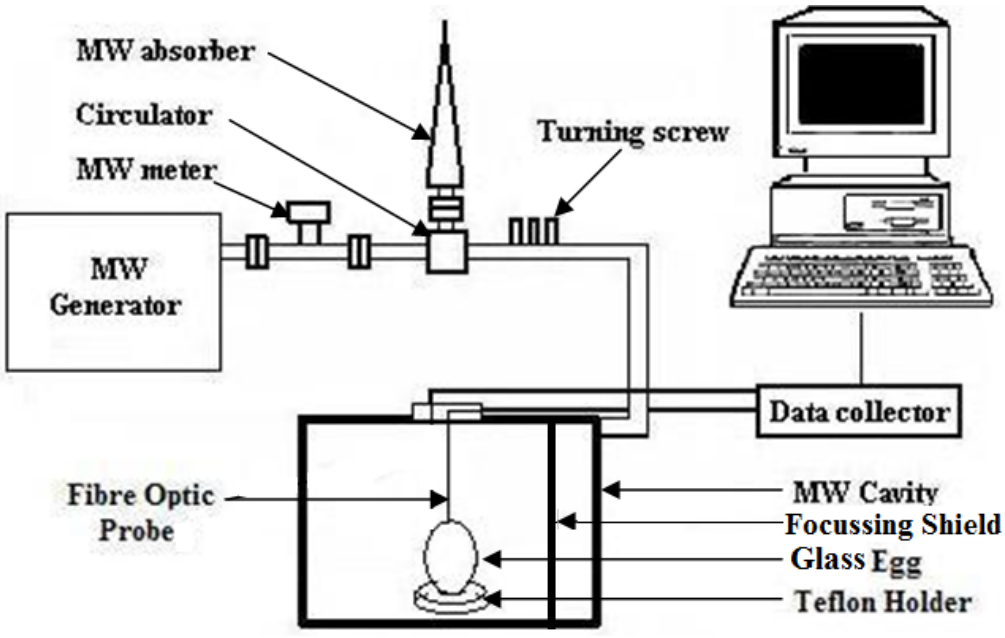


Figure 4.6 Schematic of the laboratory Microwave setup

Experiments were conducted in duplicates for each combination of parameter variations.

4.4.4 Optimization

Maximum uniformity was characterized by a minimum number of coagulated spots and smaller sized coagulations, if no treatment is free of cagulation; therefore, uniform heating implied no coagulated spots. The number and size (diameter) of the coagulated spots obtained were tabulated for each simulation trial and the combination of parameters for which there was no coagulation was considered to be the optimal set of parameters for microwave pasteurization.

4.5 Results and Discussions

4.5.1 Simulation

Figures 4.7 - 4.10 show some of the simulation results for the laboratory microwave cavity and the regular domestic microwave oven. These results indicate that MW heating is highly non-uniform, and that when the egg is rotated, the centre of the egg (yolk) heats up faster than outer contents (white). This difference, when fine tuned, will give the required pasteurization temperature gradient for the egg white and yolk.

Among the different configurations tested, the temperature profile of a shell egg heated in the laboratory oven with rotation at a power density of 2 W g^{-1} for 120 s (Figure 4.8) showed the greatest uniformity and appeared the most suitable for microwave pasteurization. Figures 4.9a and 4.9b show that the heating occurs mainly on one side if a stationary (non-rotating) egg is directly facing the waveguide port in a regular domestic microwave oven. Also the non-rotating egg in the laboratory microwave setup (Figure 4.7) and the rotating egg in a domestic microwave oven (Figures 4.10a and 4.10b) did not seem to undergo uniform heating. This was evident from their temperature profiles both in the simulation as well as in experimental validation trials.

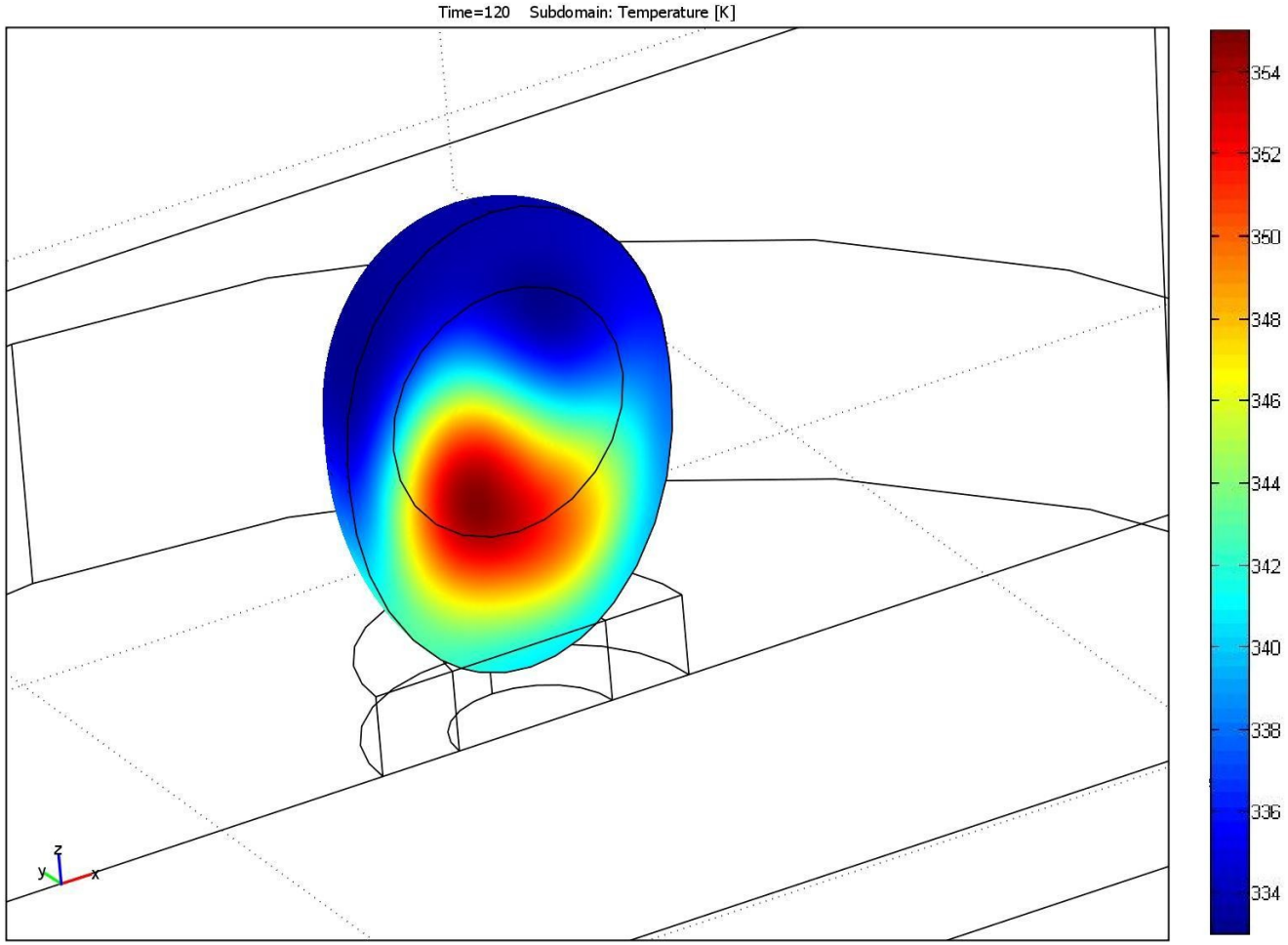


Figure 4.7 Temperature profile of shell egg heated in the laboratory oven without rotation, for power density 2 W g^{-1} after 120 s

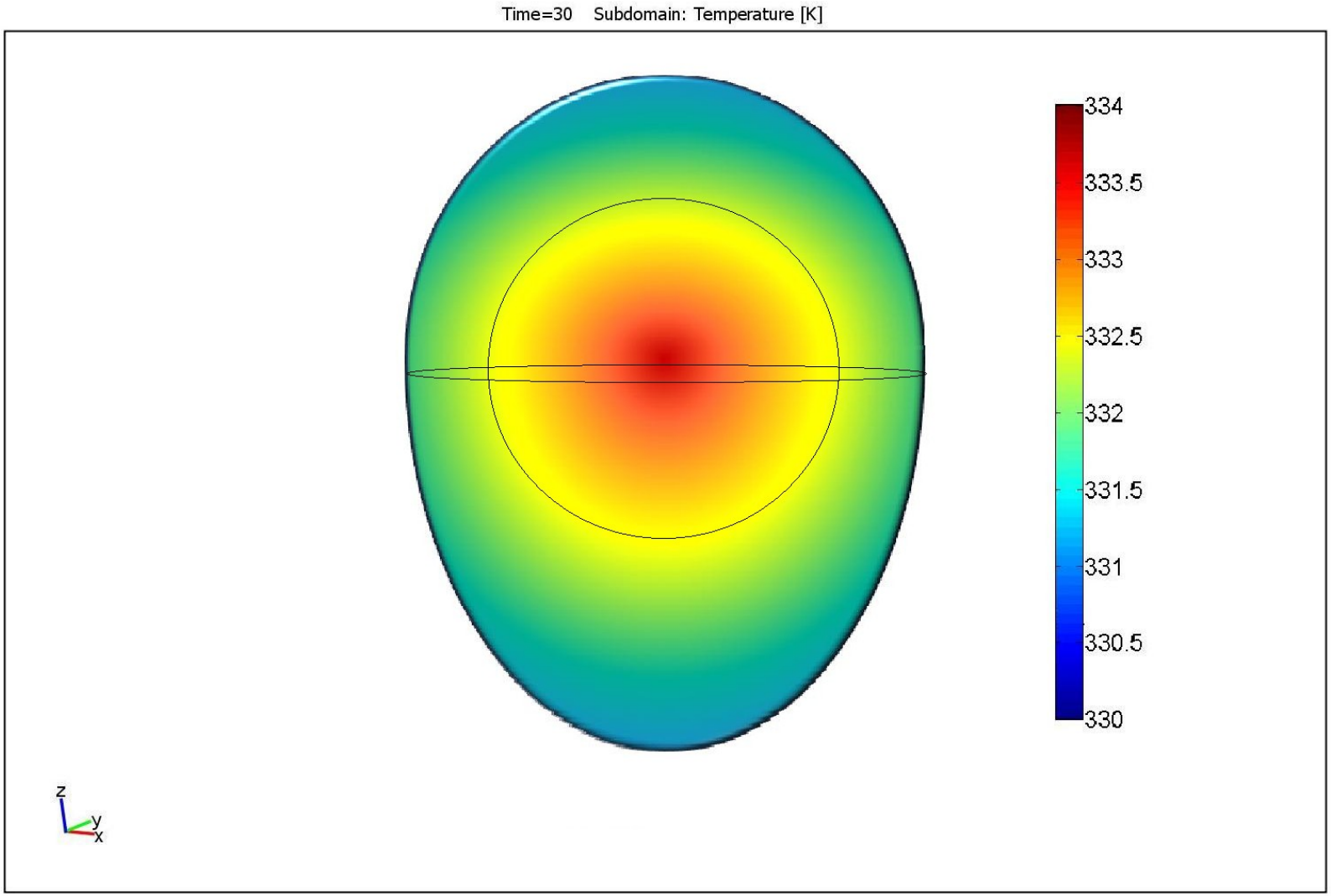


Figure 4.8 Temperature profile of shell egg heated in the laboratory oven with rotation - power density 2 W g^{-1} after 120 s

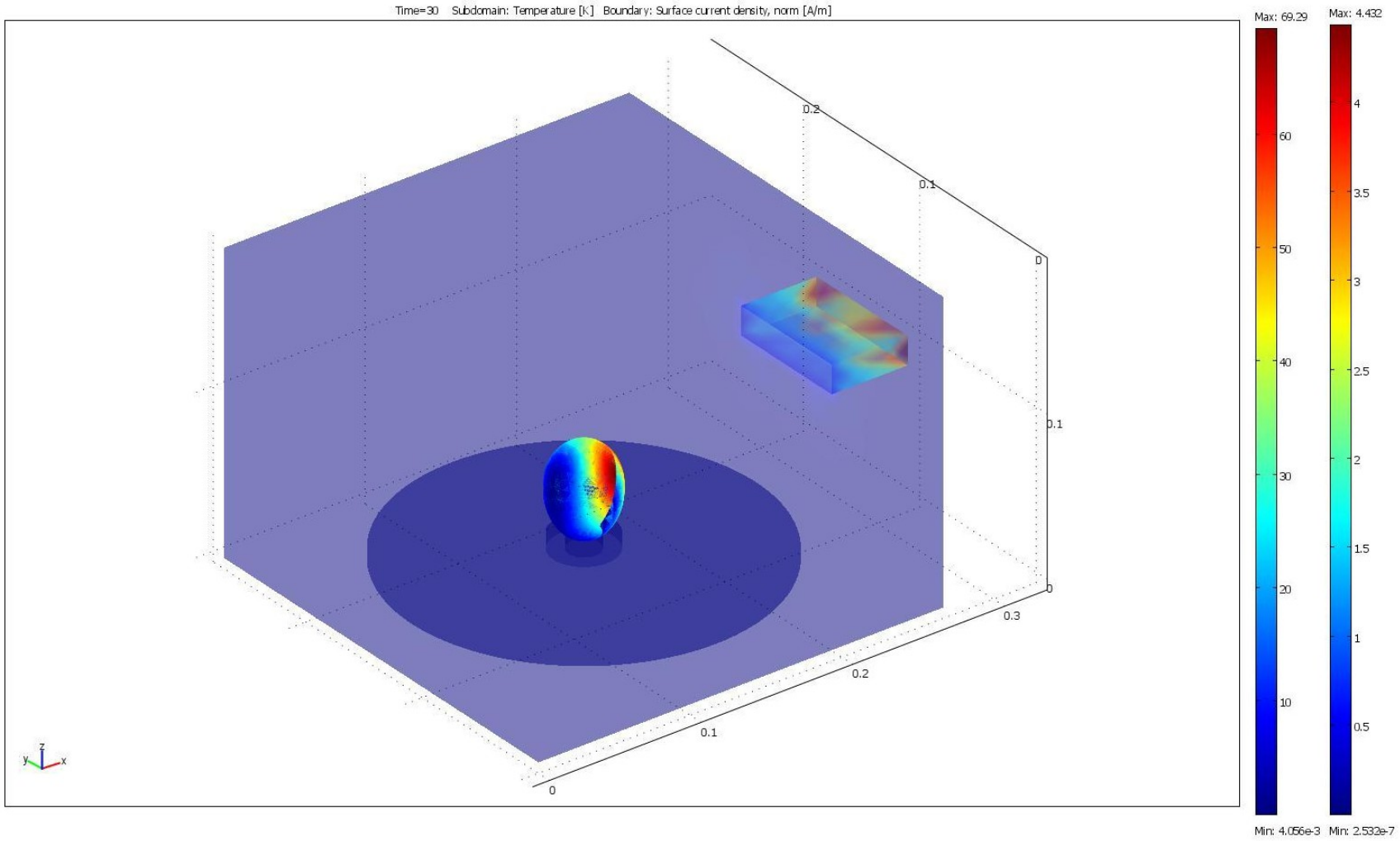


Figure 4.9 a Temperature profile and surface current density at the port for shell egg heated in a regular domestic oven without rotation, for power density 2 W g^{-1} after 120 s

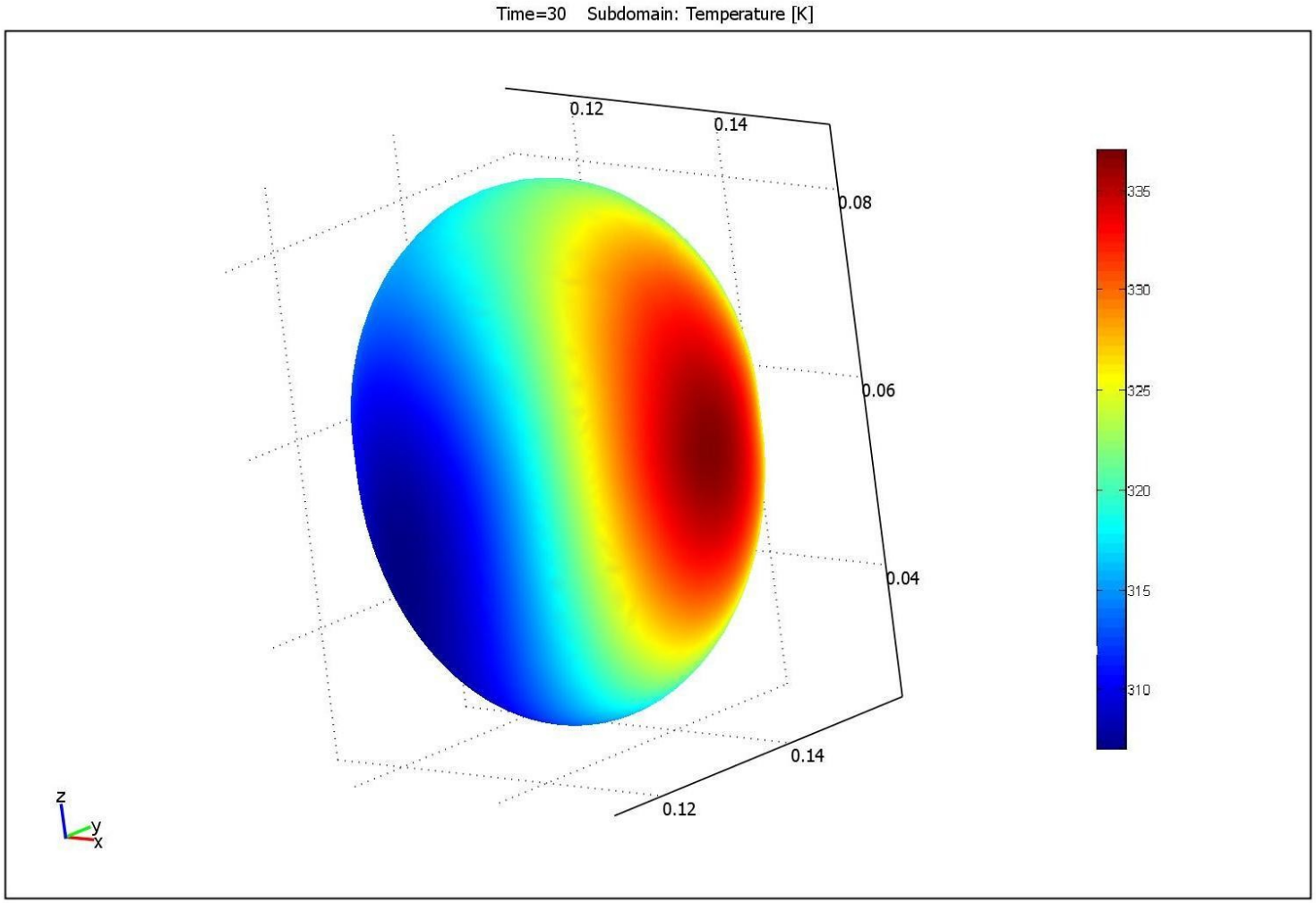


Figure 4.9 b Temperature profile of shell egg heated in a regular domestic oven without rotation, for power density 2 W g^{-1} after 120 s

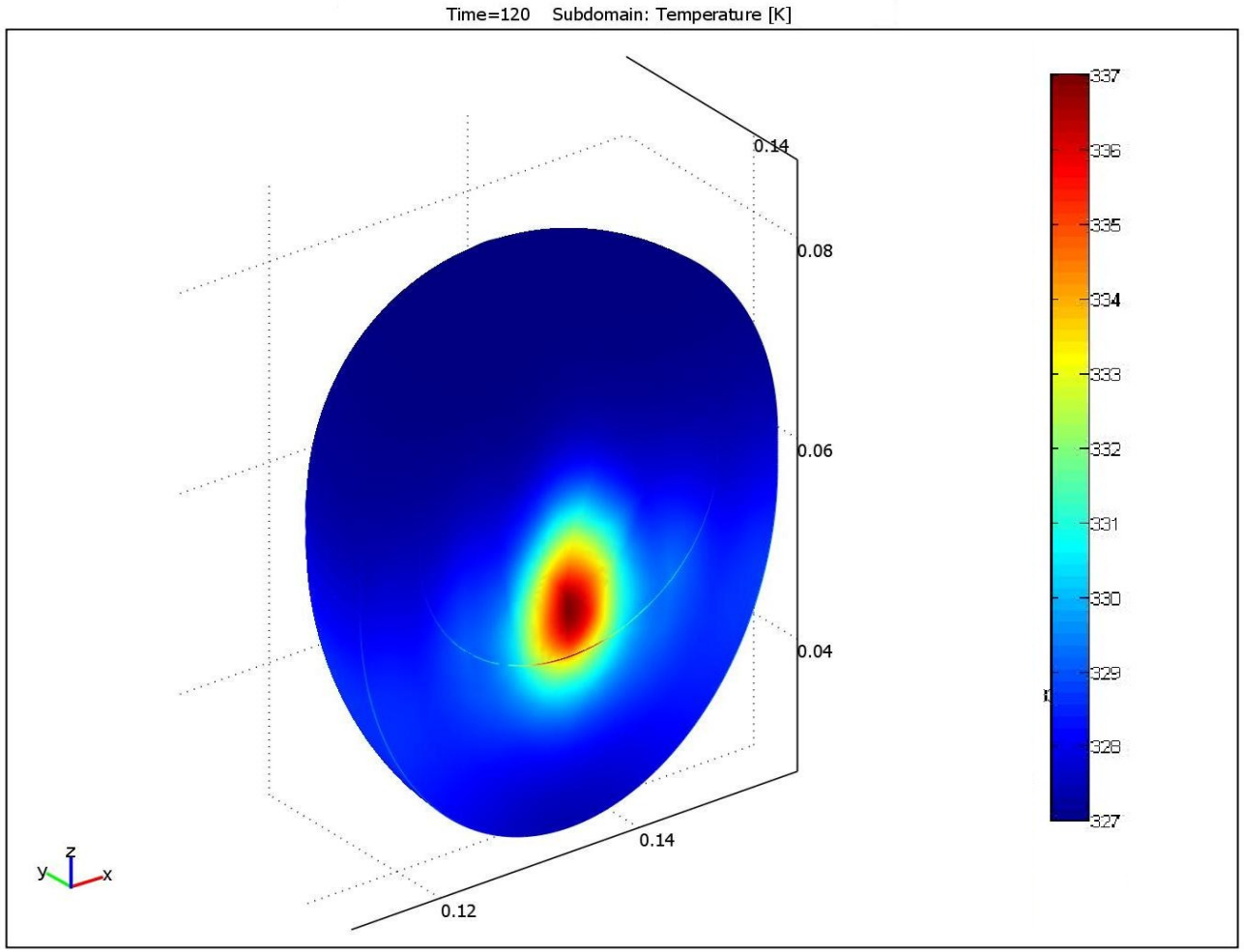


Figure 4.10a Temperature profile of shell egg heated in the regular domestic oven with rotation, for power density 2 W g^{-1} after 120 s

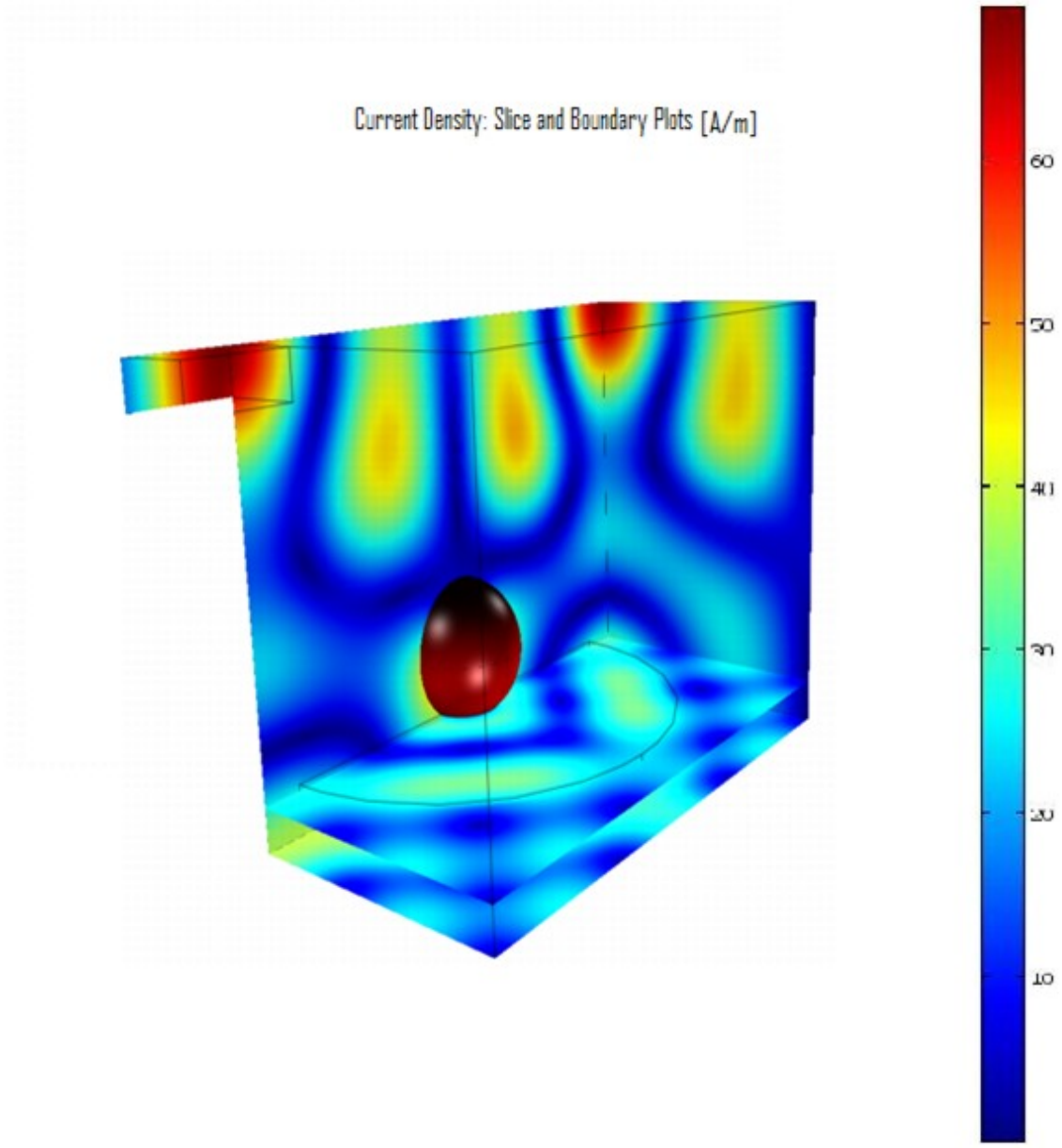


Figure 4.10b Current density profile of shell egg heated in the simulated domestic oven with rotation for power density 2 W g^{-1} after 120 s

4.5.2 Experimental Validation

Numerical simulated results corroborated well with the experimental data. The number of coagulated spots obtained was accurate and any difference in the size of the coagulated spots from the simulated ones was less than 5% in their largest diameter.

Figures 4.11 – 4.14, show the number of coagulated spots and their average size for both the verifiable existing waveguide configuration and simulated waveguide configurations. For simulation results, the regions that had reached temperatures greater than 75℃ were considered to be a coagulated spots and the size of coagulation was measured by the maximum distance of gradience to reach 70℃ from the hottest point. From the numbers and the size of the coagulated spots it is clear that the number and size of the coagulated spots increase with power density. Also rotation increases the uniformity as indicated by the lesser number and smaller size of the coagulated spots. The purely simulated results were also included in the optimization problem.

Although both power densities 1 W g^{-1} and 2 W g^{-1} for the actual laboratory microwave cavity setup with rotation are the optimal set of parameters for in-shell egg pasteurization, 2 W g^{-1} would be more preferable considering the time taken to accomplish the task. At a 2 W g^{-1} power density the pasteurization takes place in 2 minutes *vs.* 5 minutes (Dev *et al.* 2008) when using 1 W g^{-1}, which gives a 60% savings in processing time and a 20% savings in terms of energy consumption. Figures 4.12 and 4.14 do not include statistical information as they are only simulated and not experimental verification was done to determine the percentage error with respect to the actual measurements.

Jia and Jolly (1992) applied similar techniques for determination of electric field and power distribution but their models did not take into consideration a heterogeneous substance like the shell eggs and their thermodynamically changing properties.

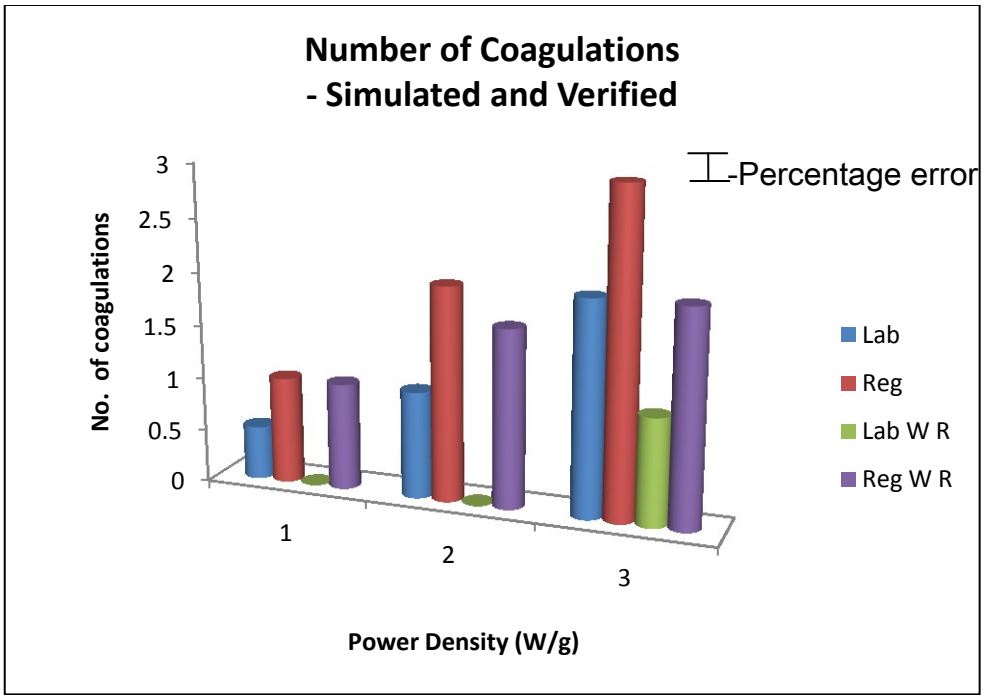


Figure 4.11 Number of coagulations – simulated and verified with the actual waveguide positions

Legend: Lab- Laboratory oven; Reg – Regular oven; W R – With Rotation

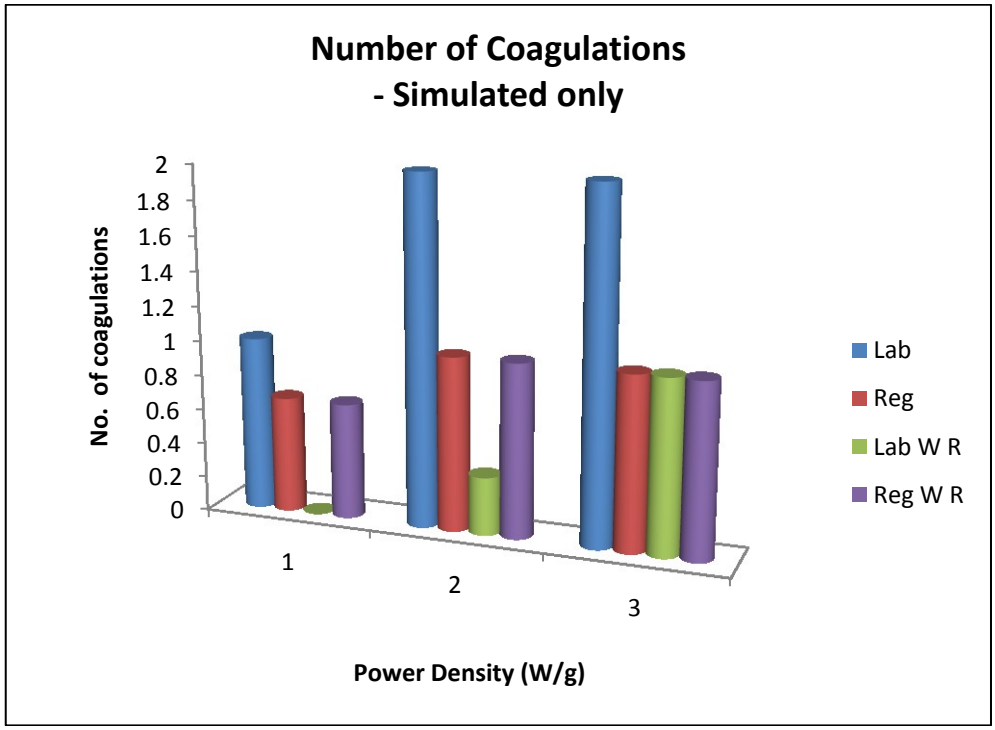


Figure 4.12 Number of coagulations – simulated with different waveguide positions

Legend: Lab- Laboratory oven; Reg – Regular oven; W R – With Rotation

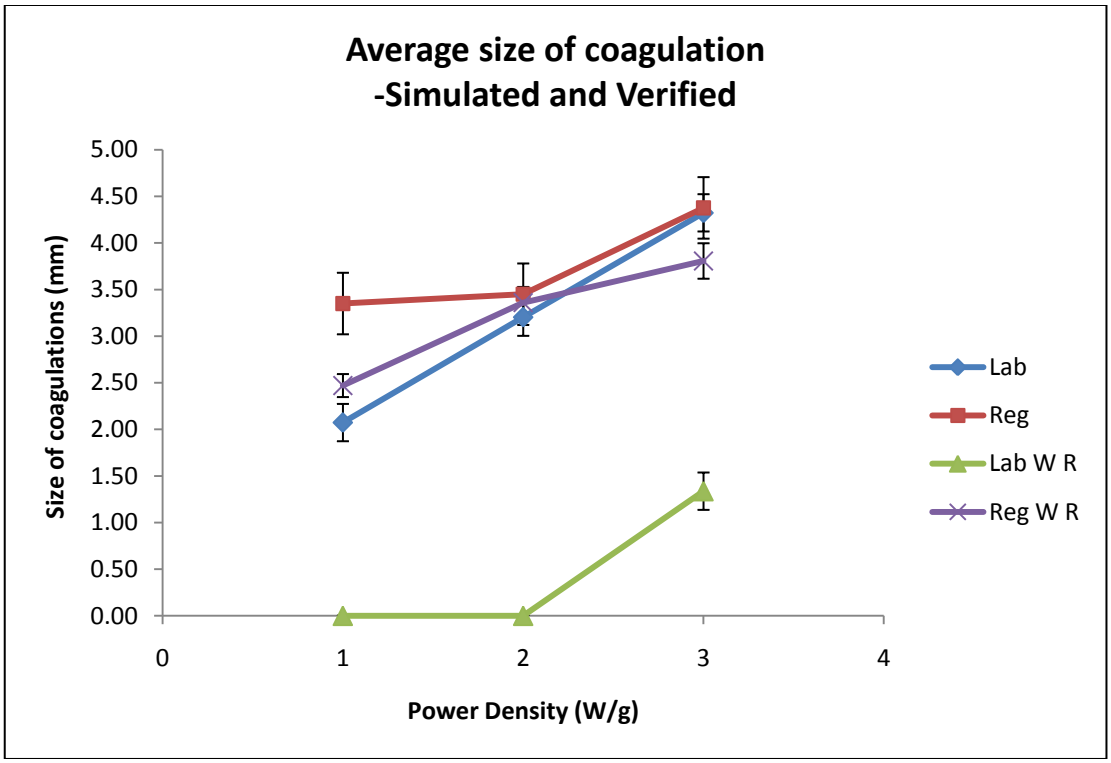


Figure 4.13 Average size of coagulation – Simulated and verified

Legend: Lab- Laboratory oven; Reg – Regular oven; W R – With Rotation, I Percetage error

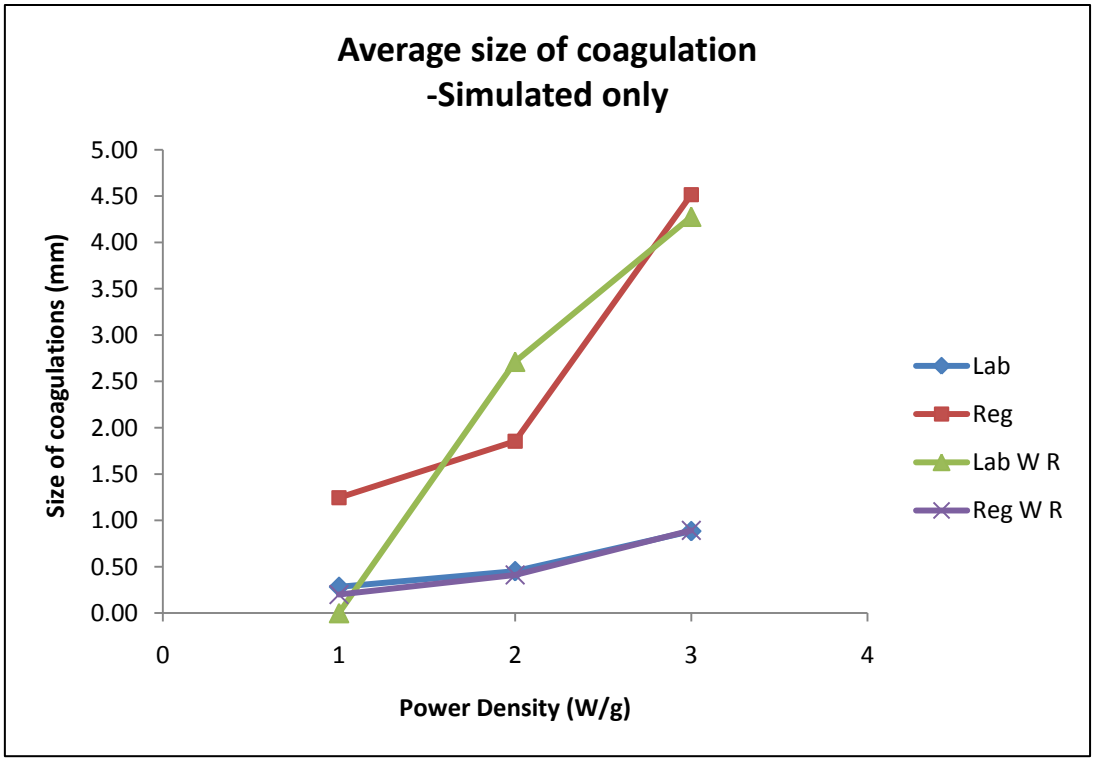


Figure 4.14 Average size of coagulation – Simulated only

Legend: Lab- Laboratory oven; Reg – Regular oven; W R – With Rotation

Zhou et al (1995) developed a model with a similar approach but their model solved for only one mode of heat transfer for a given time step. As this model involves simultaneous solutions for all the three modes of heat transfer, the error was kept within 10% for all the measured values.

4.6 Conclusions

Results from actual microwave heating and numerical simulations corroborate very well, thereby confirming the potential use of this approach for simulating any proposed design of the wave guide and the microwave cavity. These results are also useful in understanding the microwave heating process, especially for in-shell eggs. Hence it will be useful in designing equipment for the microwave pasteurization of in-shell eggs. The optimal parameters were found to results in a process that was more efficient in terms of both heating time and uniformity. Microwave heating appears to be a viable alternative for the pasteurization of in-shell eggs.

4.7 Acknowledgements

The financial support from Canadian International Development Agency (CIDA) and Natural Science and Engineering Research Council (NSERC) is acknowledged.

4.8 References

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Connecting text

The FDTD and FEM simulations along with the optimization studies led us to the conclusion that a waveguide applicator specifically designed for the microwave pasteurization of shell eggs would accomplish the task with minimal quality tradeoffs. The performance of such a waveguide applicator will also depend on various parameters viz, microwave frequency, power density, orientation of the eggs and the residence time of the egg in the cavity.

Placing a second egg in the regular multimode microwave cavity changes the field distribution significantly. Therefore to make this process more suitable for easy industrial application, an easily scalable multimode cavity which can handle multiple eggs without significant changes in distribution of power from one egg to the other is required. Hence, based on the optimal parameters obtained by simulations, a slotted waveguide applicator for heating shell eggs had to be designed and evaluated.

Chapter 5

DESIGN AND CALIBRATION OF A WAVEGUIDE APPLICATOR FOR MICROWAVE PASTEURIZATION OF SHELL EGGS

5.1 Abstract

The design of a slotted waveguide applicator for heating shell eggs is presented in which the applicator consists of an array of S–Parabolic slots surrounded by a perfect electrically conducting reflector. The issue of non-uniformity in microwave heating was overcome by optimizing the power density used for the process and by rotating the egg during the heating process. Finite element method was applied to approximate the electric field within the biological medium and a closed form expression is presented for the electromagnetic coupling problem, which enables an optimisation procedure to be performed. A power density of 1.5 W/g and an angular velocity of π/6 rad/s were found to be optimal. The results of the simulation were used to fabricate a waveguide applicator for 2450 MHz frequency with S-parabolic slots with a total power density adjusted to 1.5 W/g of load inside the cavity, rotated with a pair of rollers and a motor. The applicator enhanced both penetration and focusing, as well as provided the necessary temperature gradient from the egg yolk to the shell.

Keywords: Waveguide design, slotted waveguide, pasteurization, microwave

5.2 Introduction

Studies of electromagnetic field interactions with biological systems date back at least to the 1700s when Galvani and Volta, among others, investigated electrical effects in frogs' legs, and Mesmer used magnets to treat patients (Durney, 1992). Since that time, electrical processes were inherent to biological

systems. Various medical and biological applicators of electromagnetic fields have been studied extensively.

Conventional heating (convective/conductive) is also very non-uniform (usually only the surface is heated and the heat must conduct to the interior), and will produce a uniform temperature distribution only if the heating is done very slowly. One of the main advantages of microwave heating is that the heat is generated in the interior of the sample, avoiding the delay in heat transmission to the interior caused by low thermal conductivity (Orsat et al., 2005).

The usual problem with microwave applicators (both domestic and industrial) is that the heating pattern is not uniform, and thus the final temperature distribution is not uniform. The reasons for this are as follows, and demonstrate the problems encountered in applicator design.

1. The electric field spatial distribution (i.e., the source of the heat) is inherently sinusoidal (i.e., non-uniform) and has peaks at specific locations which change positions as the dielectric constant of the material changes.

2. The strength of the electric field (and thus the heating) is reduced in the interior of a sample because the microwaves are absorbed on the way in.

3. The dielectric constant and the microwave absorption of the material change as the temperature increases, meaning that both the previously mentioned effects also change with the temperature increase.

For the above reasons, understanding and predicting the temperature distribution in microwave heated material depends upon knowing the temperature dependence of the complex dielectric constant i.e., the real and absorptive parts (Dev et al, 2008).

To reduce the electric field non-uniformity problem, it is common practice to move the sample around in the electric field to do some averaging. In batch processing, this averaging is done either by rotating the (solid) material (as in the household oven), or by stirring the (granular or liquid) material during the heating period. In continuous feed, continuous processing mode, the averaging is usually accomplished by moving the material into the oven , pass it through and out at a steady speed, so that each piece sees the same integrated amount of heating. e.g., a conveyor belt for solids or a microwave transparent tube for liquids and granular material (Meredith, 1998).

However, although faster and more uniform, microwave heating is not inherently uniform, and to make use of its high speed in industrial processing usually requires a custom shaped applicator which produces electric field distributions which take into account the material dielectric properties at the processing temperature (Metexas 1983).

Conventional methods of heat pasteurization using hot air and hot water bath severely affect the functional quality of the eggs. Protein denaturation is a complex process which primarily depends on the time-temperature combination of the heating process. Therefore denaturation can be reduced by rapid heating. Considering microwaves as a viable alternative for the pasteurization of in-shell eggs, preliminary trials performed had confirmed that microwaves at 2450 MHz can be successfully used to raise the temperature of in-shell eggs to the required pasteurization temperatures (57.5°C for egg white and 61.1°C for yolk) (FSIS-USDA, 2006) in a few minutes (Dev et al, 2008).

Though heating uniformity can be an issue in microwave heating, it can be overcome with the proper orientation of the egg and a specially designed

waveguide, which is an engineering issue (Fleischman 2004) and also by the precise design of the egg susceptors in the microwave chamber (Yakovlev 2001).Therefore a unique S-parabolic slotted waveguide applicator was designed for 2450 MHz frequency for the in-shell egg pasteurization with the help of finite element modelling and simulation.

5.3 Mathematics of slotted waveguides

A slot cut in the waveguide wall in the direction transverse to the current lines produces significant perturbation of the current sheet, which results in coupling of the internal field to space (Silver 1949). These type of slots are called radiating slots and the degree of coupling with space depends on the current density intercepted by the slot and the component of the length of the slot transverse to the current lines. Thus coupling at a given position can be adjusted by changing the dimensions and orientation of the slot.

In general the field components of a Transverse Electric TE mode waveguide of order $a = mn$ can be written as:

$$\left.\begin{aligned} H_z &= jH_{az}\exp(\mp j\beta_a z) \\ E_t &= E_{at}\exp(\mp j\beta_a z) \\ H_t &= \pm H_{at}\exp(\mp j\beta_a z) \end{aligned}\right\} \qquad (5.1)$$

where, E_t and H_t are the transverse electric and magnetic field vectors and the signs are taken according to the position of the wave in the z direction. For a TE mode waveguide the slots act as transverse magnetic (TM) mode exits for the coupled E field from the waveguide. Therefore the general form of the TM mode field components is the same set of equations as (5.2) with H_z replaced by:

$$E_z = jE_{az} \exp(\mp j\beta_a z) \tag{5.2}$$

If β_a is real, the functions E_{az}, H_{az}, E_{at} and H_{at} are all real and depend only on a, x, and y. The component vector functions, E_{at} and H_{at} have the orthogonality property as given by:

$$\begin{aligned} \int (E_{at} \times H_{bt}) \cdot i_z dS &= 0, \quad a \neq b \\ &= S_a, \quad a = b \end{aligned} \tag{5.3}$$

where,

S_a, is twice the Poynting energy flux for a freely propagated mode, and

i_z is a unit vector in the direction O_z.

The normal modes of the guide form a complete set in terms of which an arbitrary field distribution over the wall of the guide can be expressed in the form of a Fourier expansion.

Considering a slot from z_1 to z_2 in the wall of the infinite guide and assuming that the guide is to be excited by a known field distribution along the slot, the field in the guide, which is denoted by subscript I, will consist of outgoing waves on either side of the slot. That means it will contain only waves going to the right for $z > z_2$ and only waves going to the left for $z < z_1$ as denoted by equations (5.4).

$$\left.\begin{aligned} E_{lt} &= \sum_a A_a E_{at} \exp(-j\beta_a z), \quad z > z_2 \\ E_{lt} &= \sum_a B_a E_{at} \exp(j\beta_a z), \quad z < z_1 \\ H_{lt} &= \sum_a A_a H_{at} \exp(-j\beta_a z), \quad z > z_2 \\ H_{lt} &= -\sum_a B_a H_{at} \exp(j\beta_a z), \quad z < z_1 \end{aligned}\right\} \tag{5.4}$$

The amplitude of the waves going to the right and left are not necessarily the same, and are denoted by A_a and B_a in the set of equations (5.4)

5.4 Simulation of the e-field inside the waveguide.

The design of a unique slotted waveguide requires placing the slots at the locations of the maximum electric field. Therefore the following Maxwell's equations (5.5)were solved in a 3D space using COMSOL Multiphysics version 3.5, for the dimensions of a standard WR284 waveguide, to determine the distribution of the electric field inside the waveguide.

$$\left.\begin{aligned} \frac{\partial H_x}{\partial t} &= \frac{1}{\mu_0}\left(\frac{\partial E_y}{\partial z} - \frac{\partial E_z}{\partial y}\right) \\ \frac{\partial H_y}{\partial t} &= \frac{1}{\mu_0}\left(\frac{\partial E_z}{\partial x} - \frac{\partial E_x}{\partial z}\right) \\ \frac{\partial H_z}{\partial t} &= \frac{1}{\mu_0}\left(\frac{\partial E_x}{\partial y} - \frac{\partial E_y}{\partial x}\right) \\ \frac{\partial E_x}{\partial t} &= \frac{1}{\varepsilon_0\varepsilon'}\left(\frac{\partial H_z}{\partial y} - \frac{\partial H_y}{\partial z}\right) - \frac{2\pi f\varepsilon''}{\varepsilon'}E_x \\ \frac{\partial E_y}{\partial t} &= \frac{1}{\varepsilon_0\varepsilon'}\left(\frac{\partial H_x}{\partial z} - \frac{\partial H_z}{\partial x}\right) - \frac{2\pi f\varepsilon''}{\varepsilon'}E_y \\ \frac{\partial E_z}{\partial t} &= \frac{1}{\varepsilon_0\varepsilon'}\left(\frac{\partial H_y}{\partial x} - \frac{\partial H_x}{\partial y}\right) - \frac{2\pi f\varepsilon''}{\varepsilon'}E_z \end{aligned}\right\} \tag{6 .5}$$

5.4.1 Assumptions for the simulation

The fundamental assumptions on which the simulations were based are the following:

1) The walls of the guide are perfectly conducting and of negligible thickness.
2) The slot is narrow; to be more precise, we assume that

   2 log (length of slot/width of slot)>> 1.

3) In considering the field outside the guide, the penetration of the field into the region behind the face containing the slot is neglected. In other words, the problem was treated as if the guide-face containing the slot had an infinite perfectly conducting flange on it.
4) The guide transmits only the H_{01}-wave, and the length of the slot is near that of the first "resonance" (i.e., near $\lambda/2$).

Figure 5.1 gives the electric field distribution inside a standard WR284 waveguide. Figure 5.2 gives the simulated temperature profile inside the egg heated while rotating with an angular velocity of π/6 radians/second under a standard straight slot of dimensions 56 mm X 5 mm. This results show a relatively high non uniformity within the egg.

5.4 Design of an S-Parabolic slotted waveguide

In light of the above discussions, as the eggs have a continuously varying diameter along their long axis in 2D and from the simulation results (Figure 5.1) it is obvious that the electric field strength decreases radially or rather sinusoidally from the centre point of its peak value. Thus a slot design that radiates power with relatively uniform power density to the thickest portion along the central long axis of the egg as well as the thinnest edges of the eggs needs consideration. By continuously changing the orientation of the slot the amount of microwave coupling into the space can be varied continuously.

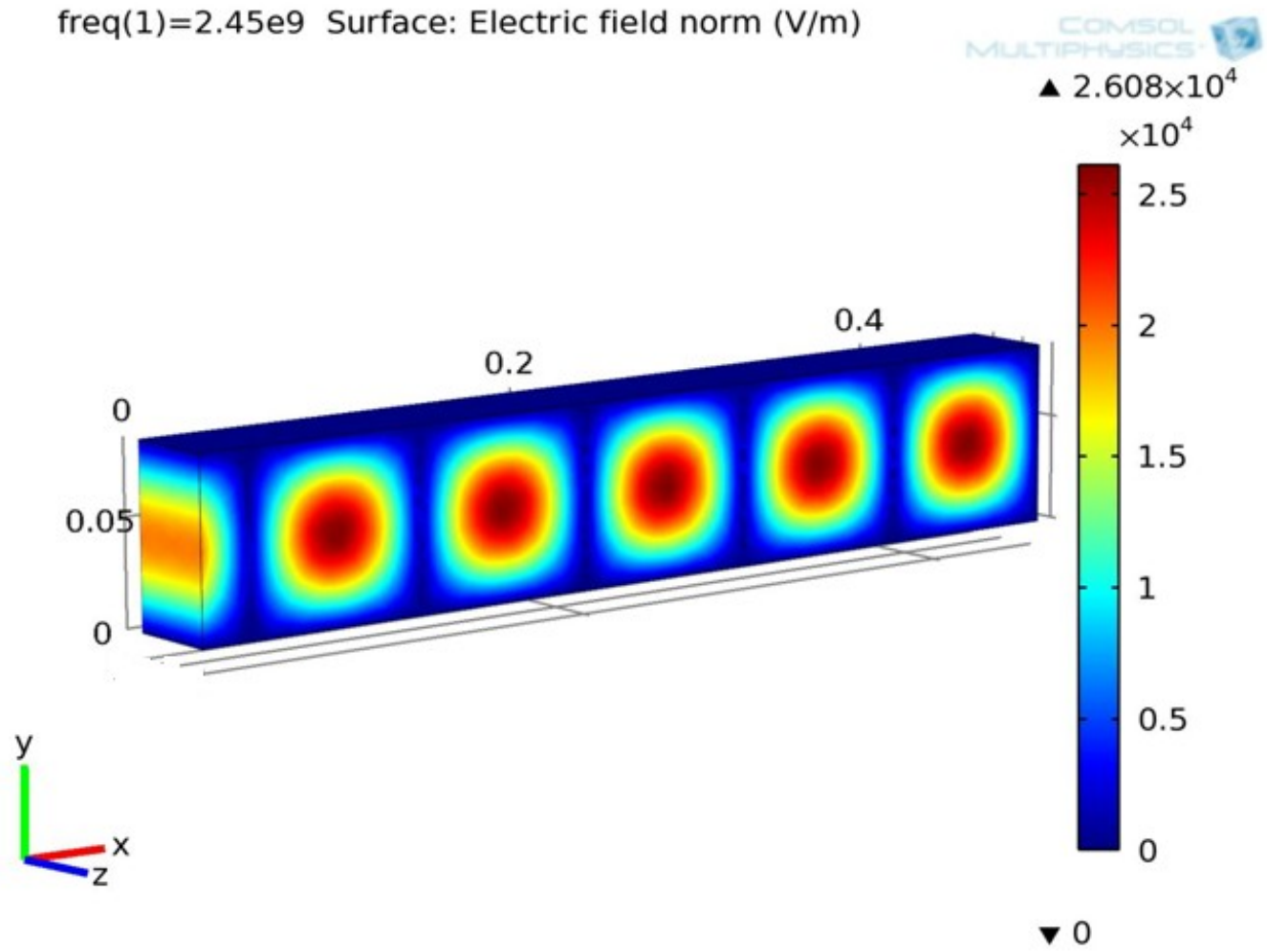

Figure 5.1 Electric field distribution along the Z axis and the XY plane

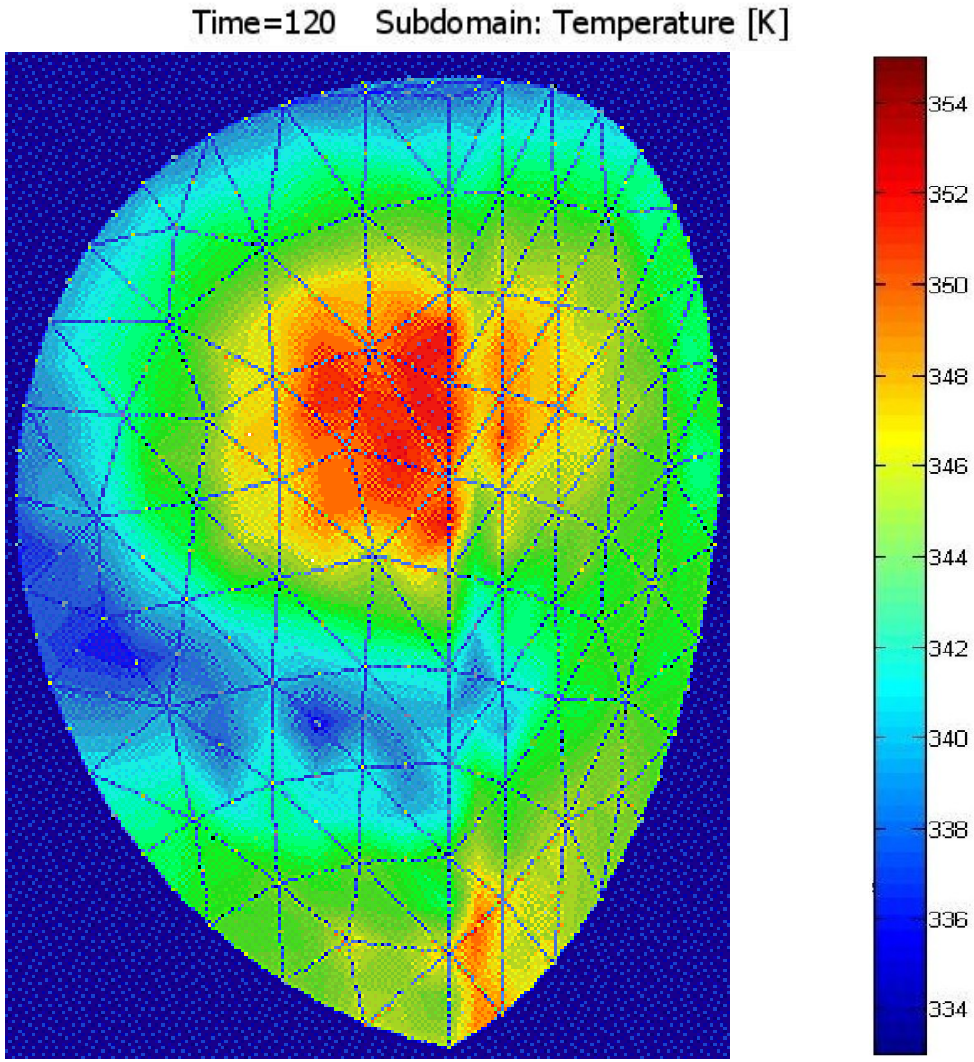

Figure 5.2 Simulated temperature profile inside the egg (quartered for better visualization) rotating under a straight slot.

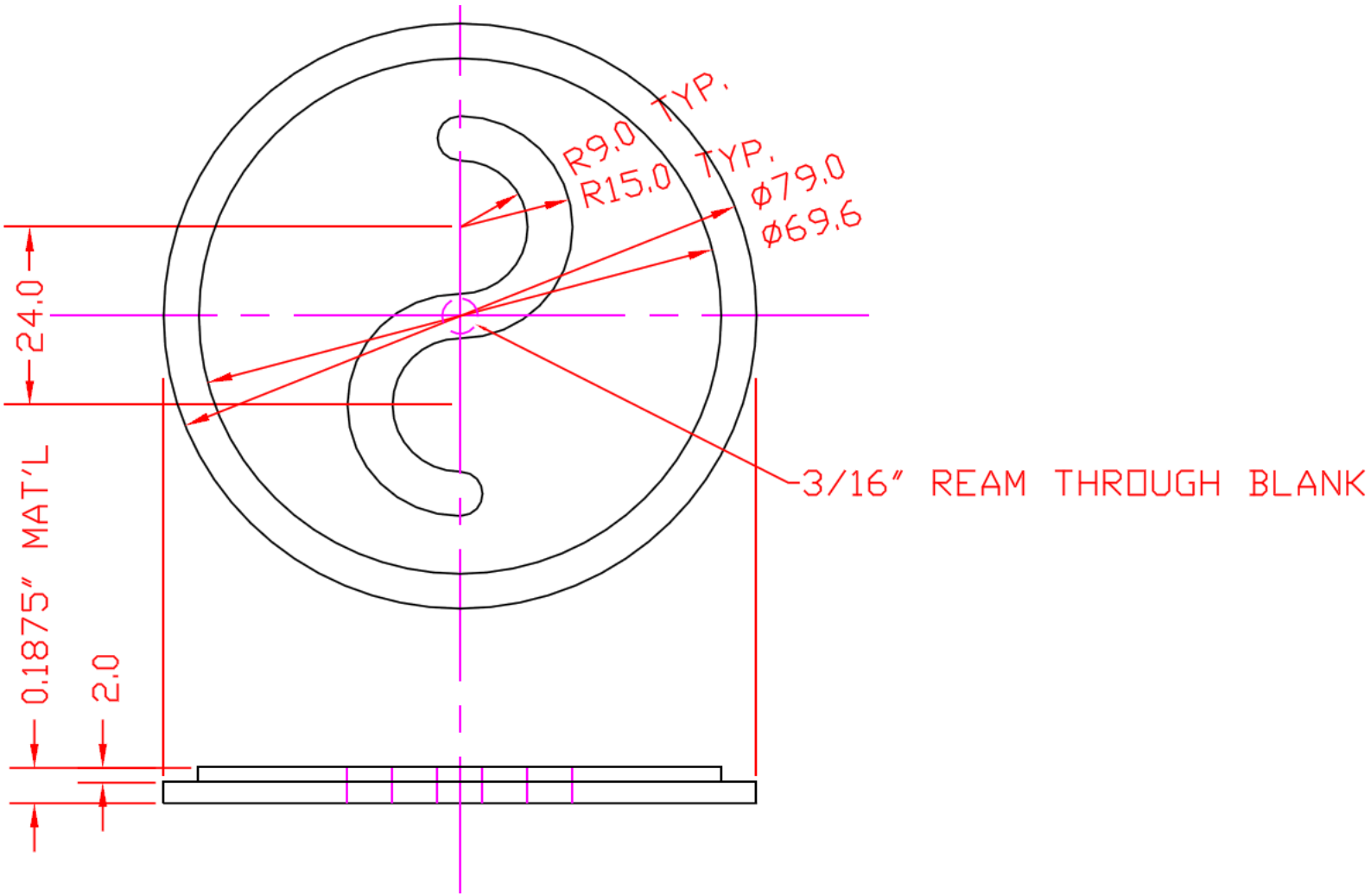

Figure 5.3 Dimension of the S-parabolic slot

Therefore, by making an S shaped slot made by two semicircular slots curved in the opposite direction, one can achieve variable output within the length of the slot. Figure 5.3 shows the dimensions of an S-parabolic slot designed for the pasteurization of in-shell eggs.

The variation of the E-field output through the S-shaped slot, follows the equation of a parabola. Hence the name S-Parabolic slotted waveguide. Also, rotating the egg while subjecting it to microwave treatment enhances the uniformity of the temperature distribution. The simulated temperature profile inside the egg rotating under an S-Parabolic slot (Figure 5.4) shows a gradient in temperature, which is ideal for the pasteurization of in-shell eggs. These simulation results were validated after fabricating the device using fibre optic probes inside the shell egg. The difference was found to be less the 0.5°K.

5.5 Fabrication of the microwave egg pasteurization equipment

An instrumented and computer controlled laboratory scale microwave (MW) oven was custom built in the laboratory for this part of the study. Figure 5.5 presents a schematic of the complete setup with all dimensions. Its main components were a 2450 MHz microwave generator (Gold Star 2M214, South Korea) with adjustable power from 0 to 400 W, waveguides, a three-port circulator, a manual three-stub tuner to match the load impedance, microwave couplers to measure forward and reflected power, a carbon load to absorb reflected power and a microwave cavity made of brass, (47 x 47 x 27 cm) in which the egg samples were processed. The wave guides were standard rectangular WR284 (72 x 35 mm) and a TE_{10} mode of application was used.

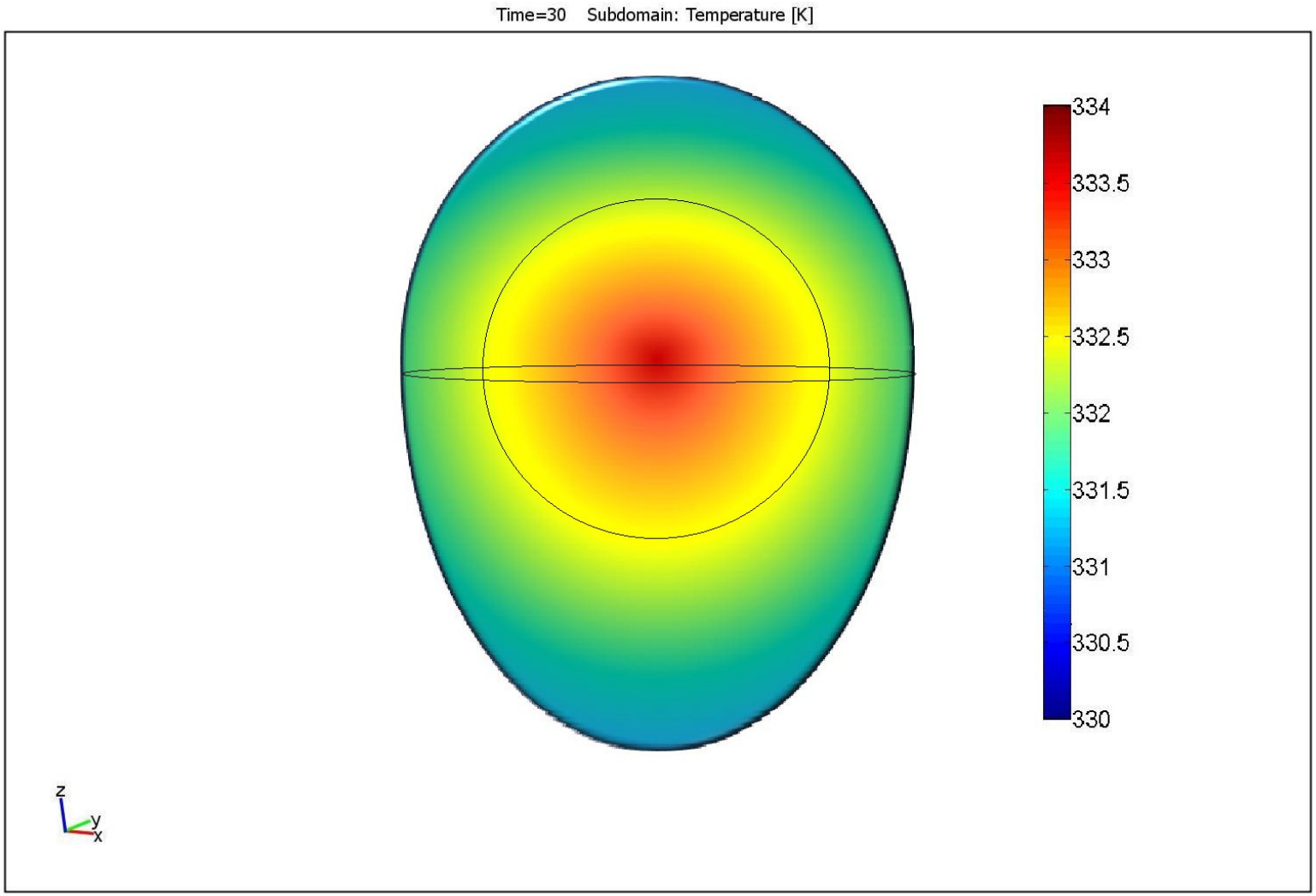

Figure 5.4 Simulated temperature profile inside the egg rotating under an S-Parabolic slot

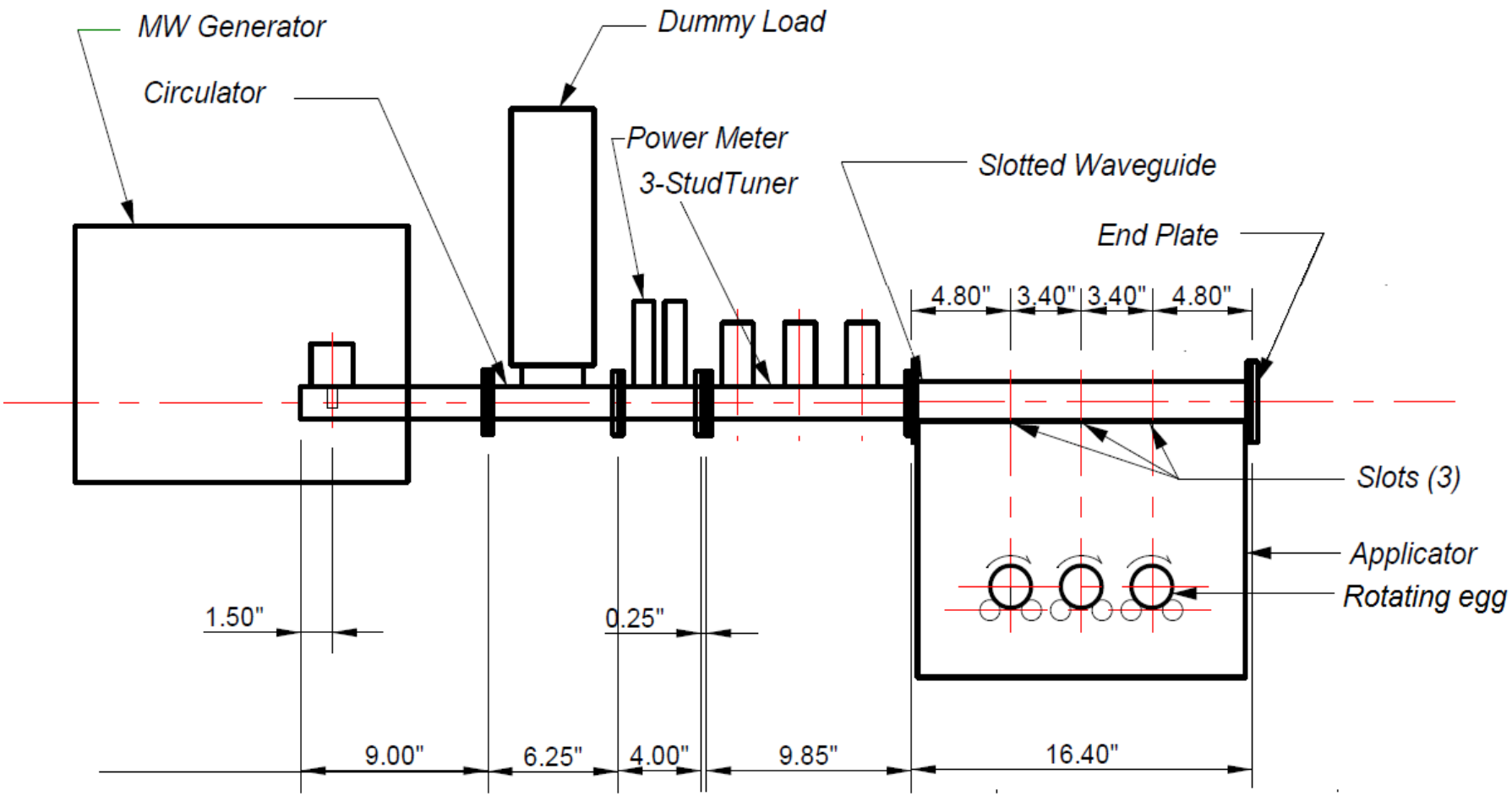

Figure 5.5 Schematic of the custom built microwave pasteurization setup

The microwave generator (magnetron) produced microwaves with varying power densities based on the power supplied. The microwaves generated were guided using the waveguides into the microwave cavity via the above mentioned sequence of components. The manual three-stub tuner was used to adjust the reflected power, thereby keeping it at the minimum possible value (<10% of the incident power).

Figures 5.6, 5.7 and 5.8 show the complete setup as well as a close up of the slots. The temperatures were measured using fiber optic probes (Nortech EMI-TS series, Quebec City, Canada). The probes were connected to a data acquisition unit (Agilent 34970A, Santa Clara, USA) which was itself connected to a computer. The entire setup was monitored and controlled using the HPVEE (Agilent, Santa Clara, USA) object-oriented programming language. The treatments were done in triplicates (each replicate obtained from an individual egg). A microwave power density of 1.5 W g^{-1} was used.

Fibre optic probes were introduced through the shell of the in-shell eggs (one for the white and one for the yolk) tentatively, assuming that the yolk was located at the center of the in-shell egg and egg white along the sides surrounding the yolk. The eggs were then heated in the microwave chamber till the yolk reached 62°C. As 62 °C was set as the microwave cut off temperature, several cycles of microwave heating occurred during the pasteurization holding time of 2.5 mins. The microwave generator was set to turn on when the temperature fell to 61°C. Eggs were rotated using cylindrical Teflon rollers as shown in Figure 5.7.

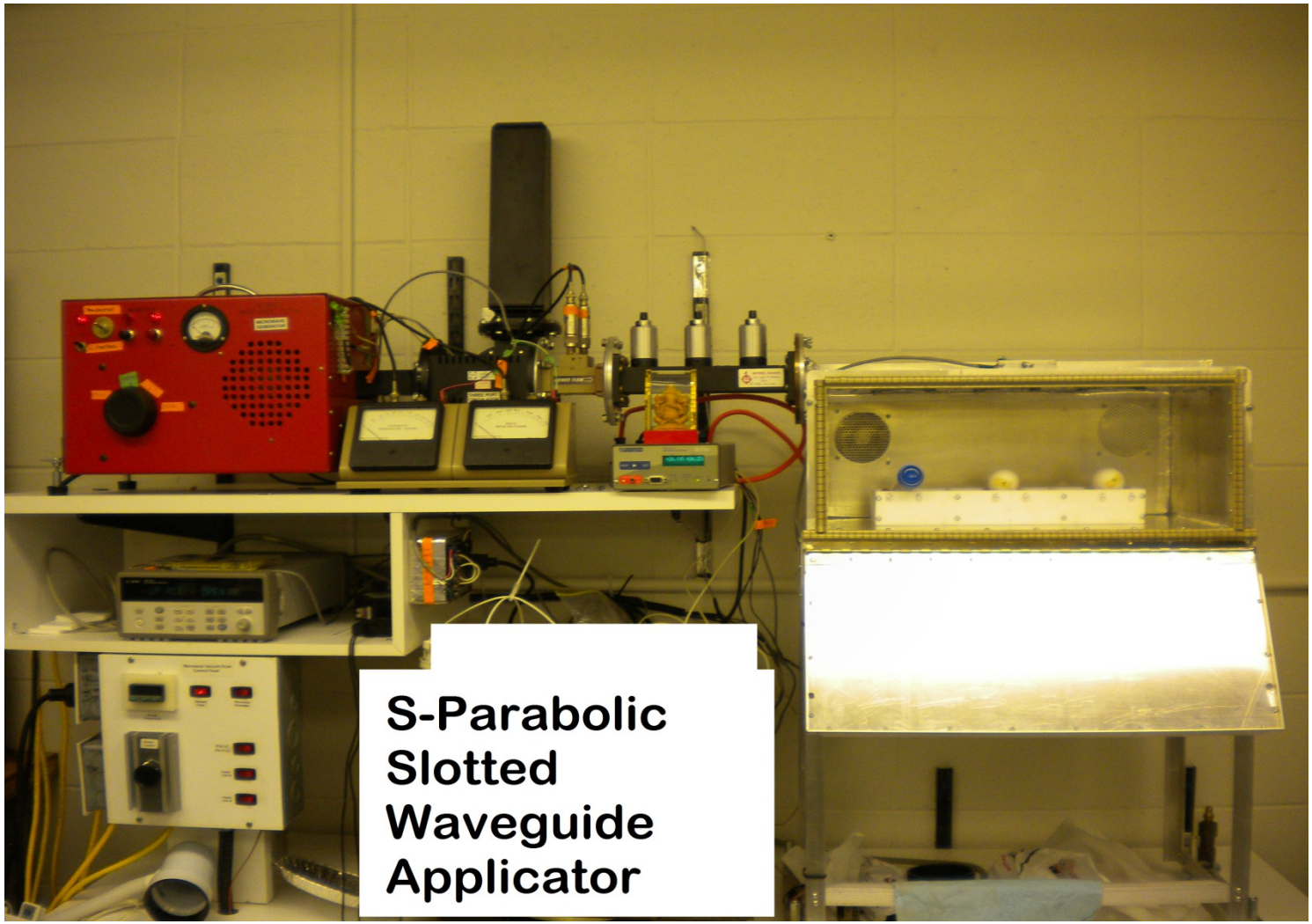

Figure 5.6 S- Parabolic slotted waveguide applicator - complete setup

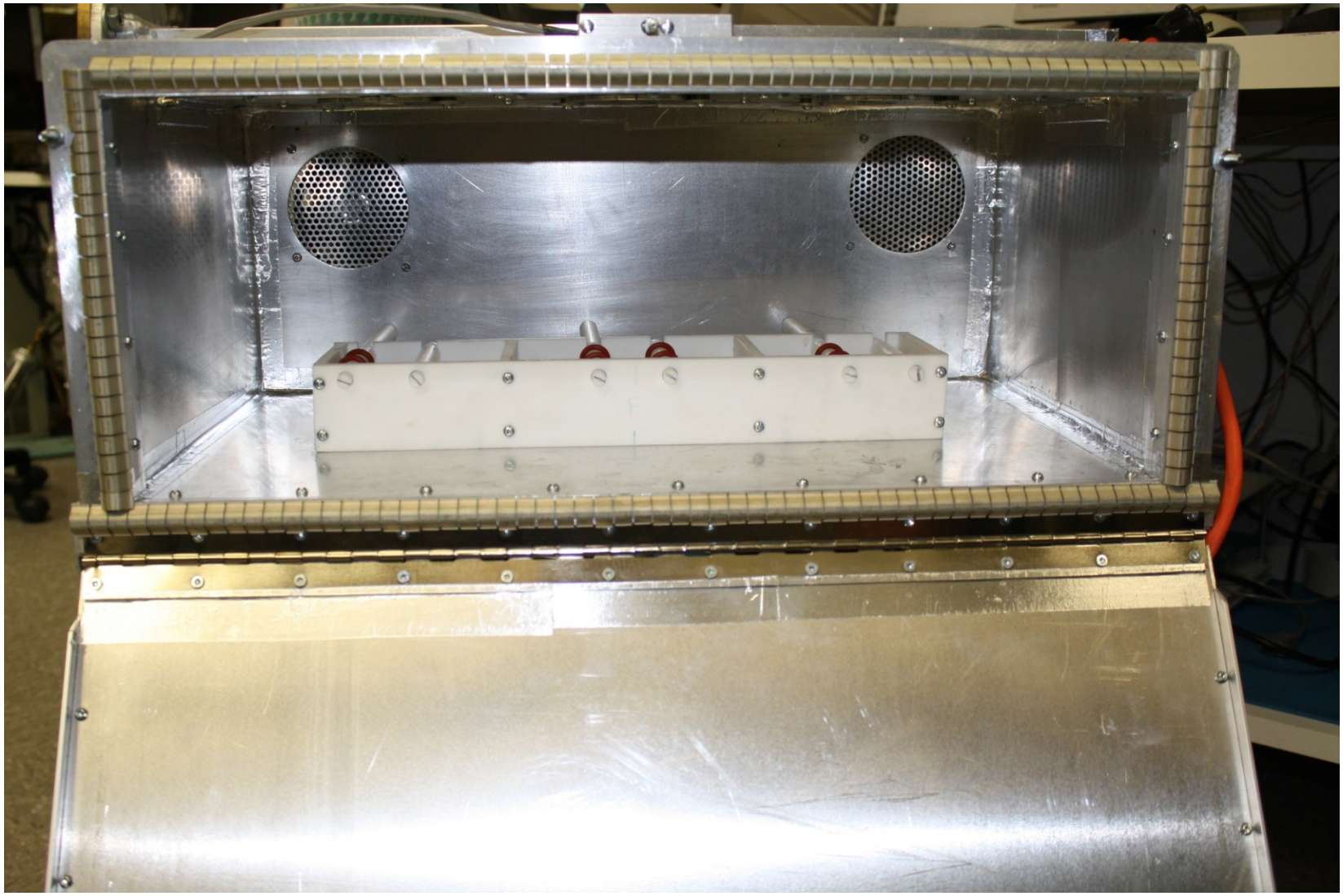

Figure 5.7 Special microwave cavity with an S-Parabolic slotted waveguide

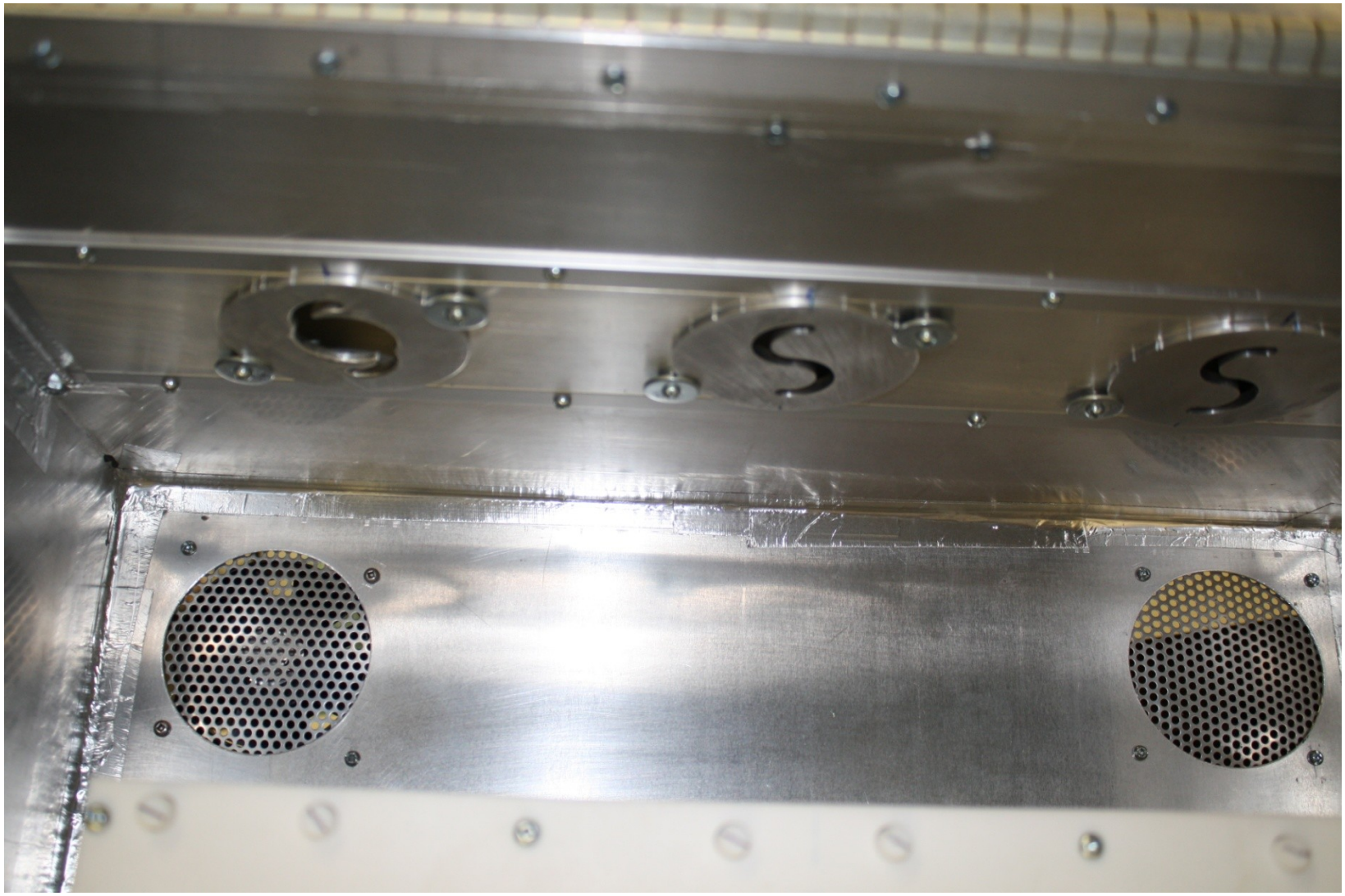

Figure 5.8 S- Parabolic slotted waveguide applicator with a galactic slot

5.6 Calibration of the microwave pasteurization setup

The calibration of the microwave pasteurization setup was done by heating 50 ml of water in microwave-transparent polypropylene tubes under each slot. As the microwaves progress through a slotted waveguide, the power radiated through consecutive slots decreases exponentially, as each slot radiates a certain percentage of the remaining power in the waveguide. This results in decreased power output in consecutive slots Therefore a 25 mm diameter circular slot was cut at the centre of the first slot forming a unique shape (Figure 5.8) which we named a “galactic slot” as it resembles a spiral galaxy. This resulted in significant distortion of the EM field by radiating 23±2% of the total power and provided a discontinuity in the E-field. This discontinuity of the sheet of electric current along one wall resulted in a shift in the position of the maximum E-field. Thus the second slot was radiating 46±2% of the available power in the waveguide and the third slot was radiating 87±2% of the available power in the waveguide as the reflected power from the terminal end of the waveguide also added up to significant portion of the radiated by this slot. Thus the distribution of the incident power was 25%, 35% and 35% respectively for the three slots with approximately 5% reflected power.

Since the galactic slot radiated lesser power compared to the other slots, a water load (50 ml of distilled water at 5℃ in a microwaveable polypropylene tube) instead of an egg was placed under this slot to absorb the power radiated. The eggs under the other slots then were exposed to an uniform power distribution and hence heated up uniformly. This was validated by experimental trials using fibre optic probes.

This setup would not be a problem to recreate at the industrial scale, as a large magnetron can be used and each slot can radiate a maximum of 215 W only. Therefore passing large amounts of power into the waveguide automatically provides equal distribution of power across several slots and the total number slots radiating uniform power will depend on the total power input.

5.7 Conclusions

Thus a slotted waveguide with a unique S-parabolic slot was designed, fabricated and calibrated. A power density of 1.5 W g^{-1} and an angular velocity of $\pi/6$ rad s^{-1} were found to be optimal. The results of the simulation were used to fabricate a waveguide applicator for 2450 MHz frequency with S-parabolic slots with a total power density adjusted to 1.5 W g^{-1} of load inside the cavity, rotated with a pair of rollers and a motor. The applicator enhances both penetration and focusing, as well as provides the necessary temperature gradient from the egg yolk to the shell. Industrial scale up of this is relatively simple but requires further research. The results obtained in this study can readily be used in building a scaled up version for application in the industry.

5.8 Acknowledgements

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Connecting text

After designing and fabricating a slotted waveguide applicator, it was necessary to verify the pasteurization performance of the applicator. Pasteurization being a thermo-biological process requires inoculation of microbial cultures, incubation, heat treatment and assessment of the microbial load after heat treatment. Handling pathogens like *Salmonella* requires great care and additional safety equipment. Therefore using surrogate non-pathogenic bacteria is the best practical approach. Hence a microbial validation with such non-pathogenic surrogate bacteria could validate the effectiveness of the designed slotted waveguide applicator.

Chapter 6

MICROBIAL VALIDATION OF MICROWAVE PASTEURIZATION OF EGGS

6.1 Abstract

To validate the effectiveness of a novel microwave egg pasteurization process, non-pathogenic *Escherichia coli* K12 was used as a surrogate for pathogenic *Salmonella enteritidis* in eggs. *Escherichia coli* K12 (ATCC 23716) was cultured in *E. coli* broth for 2 days. Grade A shell eggs were inoculated with the 10^5 CFU ml^{-1} cultured *E. coli* K12 and incubated below 5℃ for 5 days. Microwave pasteurization of eggs was carried out using a laboratory scale controlled microwave cavity setup, a regular domestic microwave oven with a turn table, a special microwave cavity fitted with an S-Parabolic slotted waveguide and a hot water bath. The eggs were then broken and plated in EC agar and incubated for 2 days at 37℃. The inoculated but thereafter untreated eggs had a count of 10^6 CFU ml, whereas both types of microwave-pasteurized eggs had no detectable colonies. This indicated that microwave pasteurization is an effective way of pasteurizing in-shell eggs.

Keywords Microbial Validation, Microwave pasteurization, In-Shell egg pasteurization

6.2 Introduction

Eggs remain a potential host for different pathogens. Contamination of eggs with one serotype of the bacteria *Salmonella* namely *Salmonella enteritidis* has adverse economic implications for the poultry industry (Bruce & Drysdal,

1994; Wong & Kitts, 2003). There are 1.4 million infections, with more than 16,000 hospitalizations and nearly 600 deaths each year, due to food-borne salmonellosis in the United States. Incidences of egg salmonellosis has increased steadily from 1976 to 2001 (Shah, Bradshaw, & Peeler, 1991; CDC, 2001).

More than 90 percent of food-borne Salmonellosis, caused by *Salmonella enteritidis*, occurs through shell eggs (Schroeder, Naugle, Schlosser, Hogue, Angulo et al. 2005; Woodward, Khakhria, Johnson, 1997). Most *Salmonella enteritidis* outbreaks involved Grade A eggs that were washed and disinfected and also met the State requirements for shell quality (St. Louis, Morse and Potter, 1988). The probability of fresh eggs having Salmonella varies from 0.005% (Mermelstein, 2001) to 1 % (Grifiths, 2005) depending on various factors involved in the egg production. In particular, the risk of illness increases when egg is used as an ingredient in meals prepared for the general public (Todd, 2001). As a result, the US Department of Agriculture (USDA) regulations mandate that commercial egg products must be subjected to pasteurization processes to reduce pathogens to a reasonably acceptable level.

Egg white is used as a foaming, leavening, gelling and/or binding agent in numerous food preparations. Egg white proteins are the most heat sensitive components of an egg. Egg yolk has good emulsifying and binding properties (Li-Chan, Powrie and Nakai 1995), but these properties are severely affected by high temperatures (Van der Plancken et al., 2006). The conventional methods of thermal processing of foods result in peripheral over heating before the material in the centre reaches the required temperature. This is potentially a great problem in pasteurization, especially when it comes to shell eggs.

S.R.S. Dev *et al.* (2008) had demonstrated that microwave heating is an excellent alternative to overcome the problem of peripheral overheating during shell egg pasteurization. With this heating system the FSIS recommendation of heating up the yolk to a higher temperature (61.1ºC) was rendered possible without heating the egg white beyond its recommended pasteurization temperature (57.5ºC). The risk of great pressure build-up within the egg shell when heated using microwaves is absolutely preventable within the pasteurization temperatures (Fleischman, 2005; Rehkopf, 2005).

Non-pathogenic surrogate microorganisms can be used in the place of the pathogens to validating thermal processes like pasteurization. The thermal tolerance of the surrogate microorganism must be equivalent to or higher than the targeted pathogen (Eblen et al, 2005). Jin et al. 2008 had observed that the non-pathogenic *E. coli* K12 exhibited similar kinetic behaviour, but higher thermal resistance than *S. enteritidis* in both liquid egg white and liquid whole egg. Thus E. coli K12 can serve as an appropriate surrogate in evaluating the efficacy of thermal pasteurization for reducing and/or eliminating *S. enteritidis* in eggs.

Therefore this study was conducted with the objective of evaluating the microbial destruction efficiency of the microwave pasteurization process for in-shell eggs using the non-pathogenic *E. coli K12* in the place of the pathogenic *S. enteriditis.*

6.3 Safety Emphasis

The study was conducted with a non-pathogenic strain of bacteria (*E.coli* K12*).* The inoculation and plating operations were conducted in an UV-sterilized laminar flow chamber (Fisher Scientific, USA) equipped with a Bunsen burner. A biological safety cabinet (Fisher Scientific, USA) was used for storing the plates.

6.4 Materials and Methods

In order to evaluate the microwave pasteurization efficiency the *E.coli* K12 was cultured, inoculated, incubated, and subjected to two different microwave pasteurization treatments, and then plated to assess the surviving population (CFU – Colony Forming Unit).

6.4.1 The Culture

The *E.Coli* K12 ATCC 23716 was obtained in lyophilized form in vials from Cedarlane® Laboratories Limited, ON, Canada. This was rehydrated using EC broth (Oxoid Canada) and cultured for 48 hours to obtain an initial population of about 3.2×10^8 CFU ml^{-1}.

6.4.2 Egg samples

The fresh whole eggs, within 3 days of grading and packing (identified from the best before date stamped on the eggs, which is usually 35 days from date of packing), (Li Chan et al., 1995) used in this study were procured from the local market and kept in a refrigerator at 5˚C until used. They were all Canadian Grade A eggs, size large, with a mean mass of 60±2 g.

6.4.3 Inoculation and Incubation

Inoculation was done in triplicate for the two microwave treatments and for the untreated control. The egg samples were inoculated by drilling a tiny hole of less than 1 mm in diameter using a drill bit sterilized with alcohol and injecting 100 µl of the above mentioned *E.coli* culture into the egg yolk. This is done to prevent the inoculums from getting in the egg white, as the egg white contains lysozyme, which is antibacterial in nature. Comparatively egg yolk is more

conducive to the growth of bacteria (Fleishman et al. 2003). The hole made for inoculation was sealed with sterile masking tape. Three sets of inoculated eggs in triplicate (9 eggs in total) were incubated at ambient temperature (23±2℃) for 2 days to allow the bacteria to grow and spread within the yolk.

6.4.4 Heat treatments for pasteurization

Three heat treatments for the microwave pasteurization of in-shell eggs were investigated: the standard hot water method, the instrumented and computer controlled laboratory setup, and a regular domestic microwave oven.

6.4.4.1 Computer Controlled Laboratory Microwave Setup

The first treatment consisted of heating in-shell eggs in a laboratory scale microwave oven working at 2450 MHz using a power density of 1 W g^{-1}. A custom built instrumented and computer-controlled laboratory-scale microwave (MW) oven (Figures 6.1, 6.2 and 6.3) was used for this part of the study. Its main components were: a 2450 MHz microwave generator (Gold Star 2M214, South Korea) with adjustable power from 0 to 750 W, waveguides, a three-port circulator, a manual three-stub tuner to match the load impedance, microwave couplers to measure forward and reflected power, a carbon load to absorb reflected power and a microwave cavity made of brass, (47 x 47 x 27 cm) in which the egg samples were processed. The wave guides were rectangular (72 x 35 mm) and the TE10 mode of application was used.

The microwave generator (magnetron) produced microwaves with varying power densities based on the supplied power. The generated microwaves were guided using the waveguides into the microwave cavity via the above mentioned components in a sequence. A manual three-stub tuner was used to adjust the

reflected power, thereby keeping it at the minimum possible value (<10% of the incident power). The temperatures were measured using fiber optic probes (Nortech EMI-TS series, Quebec City, Canada). The probes were connected to a data acquisition unit (Agilent 34970A, Santa Clara, USA) which was itself connected to a computer. The entire setup was monitored and controlled using the HPVEE (Agilent, Santa Clara, USA) object-oriented programming language.

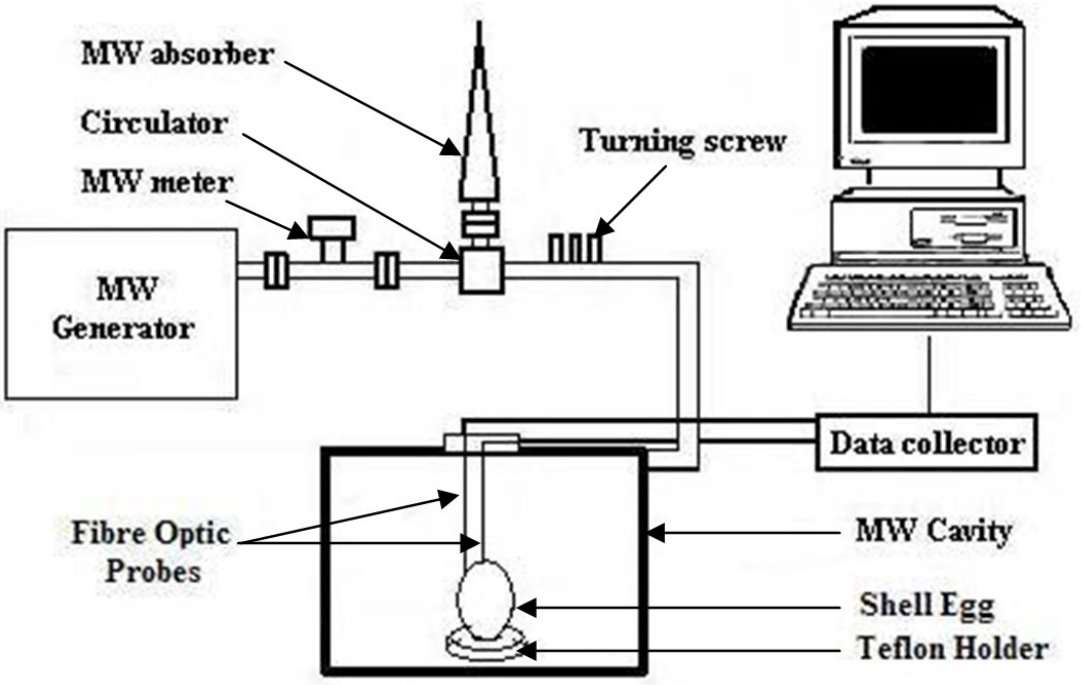

Figure 6.1 Laboratory controlled microwave setup

Figure 6.2 Experimental setup for Microwave pasteurization

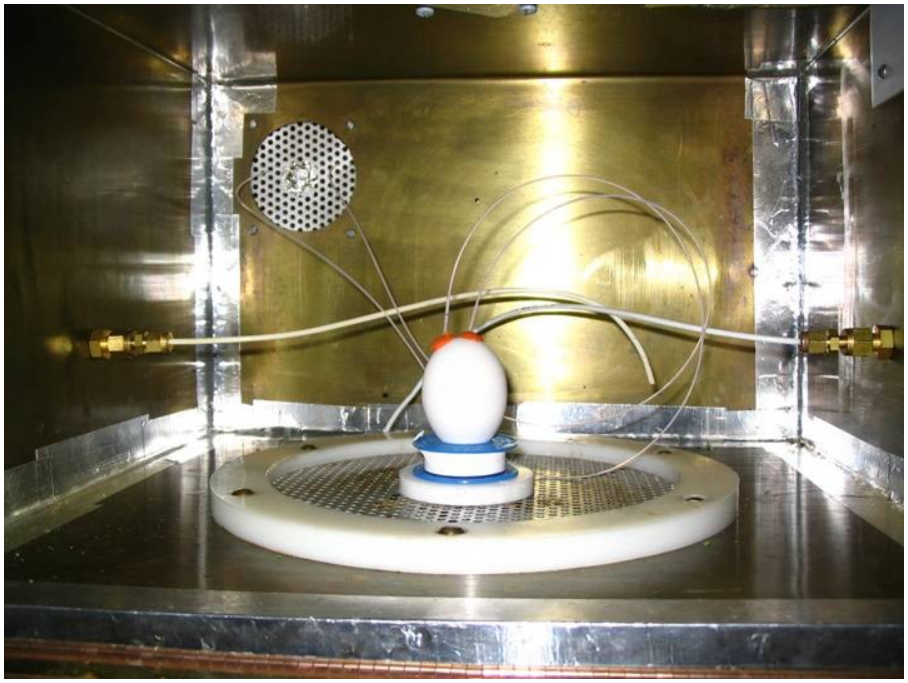

Figure 6.3 Shell egg with fibre optic probes in the microwave cavity

The treatments were done in triplicates (each replicate obtained from an individual egg). A microwave power density of 1.5 W g^{-1} was used. The hole made for inoculation was used for the insertion of probes for temperature control during the pasteurization process and one more hole was similarly drilled 1.5 cm away from the previous hole for inserting a probe into the egg white.

Fibre optic probes were introduced through the shell of the in-shell eggs (one for the white and one for the yolk) tentatively, assuming that the yolk was located at the center of the in-shell egg and egg white along the sides surrounding the yolk. The eggs were then heated in the microwave chamber with the broad end of the egg facing upwards, till the yolk reached 62°C. As 62 °C was set as the microwave cut off temperature, several cycles of microwave heating occurred during the pasteurization holding time of 2.5 min. The microwave generator was set to turn on when the temperature fell to 61°C. Eggs were held upright using a cylindrical Teflon holder (Figure 6.3).

Hot spots and cold spots which are characteristic of microwave heating were ignored in placing the probes, as the standard deviation of the heating time for consecutive measurements was very small (<5%).

6.4.4.2 Regular Domestic Microwave Oven Setup

A Panasonic NNSN968B full size regular domestic microwave oven with turn table was used at power level 1 (89 W calibrated power). The same Teflon egg holder (Figure 6.1) was used to hold the egg upright with the broad end up. The heating time to reach the yolk pasteurization temperature of 62°C from 5°C was calculated to be 1 minute and 48 seconds, based on an optimised finite element model developed by Dev et al. (2008) for microwave heating of in-shell eggs. The eggs were heated in the microwave cavity for the above mentioned

time and the recommended holding time for pasteurization (2.5 mins) was maintained by momentarily transferring the egg to a water bath maintained at 65°C.

Immediately after these treatments the shell eggs were immersed in a cold water bath (5°C) for 10 minutes, in order to ensure that the extent of heat damage to the proteins did not continue beyond the duration of the pasteurization.

6.4.4.3 Special microwave cavity with an S-Parabolic slotted waveguide Setup

A microwave cavity specifically designed for the pasteurization of shell eggs (Figures 6.4, 6.5 and 6.6) was used for this part of the study. This was similar to the computer-controlled laboratory domestic microwave setup (section 6.4.4.1), except for a specially designed cavity and waveguide applicator. The waveguide has uniquely-designed S-parabolic slots (Figure 6.6).

As the power dissipation decreases exponentially from the first slot to the n^{th} slot in a regular slotted waveguide, the dimensions of the first slot were adjusted to form a “galactic slot” (named after its shape), which provided distortion of the E field, making it possible to have a uniform power distribution among the slots.

The power emitted by the galactic slot was only half of the others, which would have led to unequal heating in the eggs. Hence instead of placing an egg, a water load was placed under the first slot to absorb the excess power. A power density of 1.5 W g^{-1} was applied. The eggs were continuously rotated at a speed of 5 rpm during the treatment with the help of a pair of rollers for each egg, controlled by a switch. The unit had a cooling fan attached to it, in order to vent any heat generated during the process.

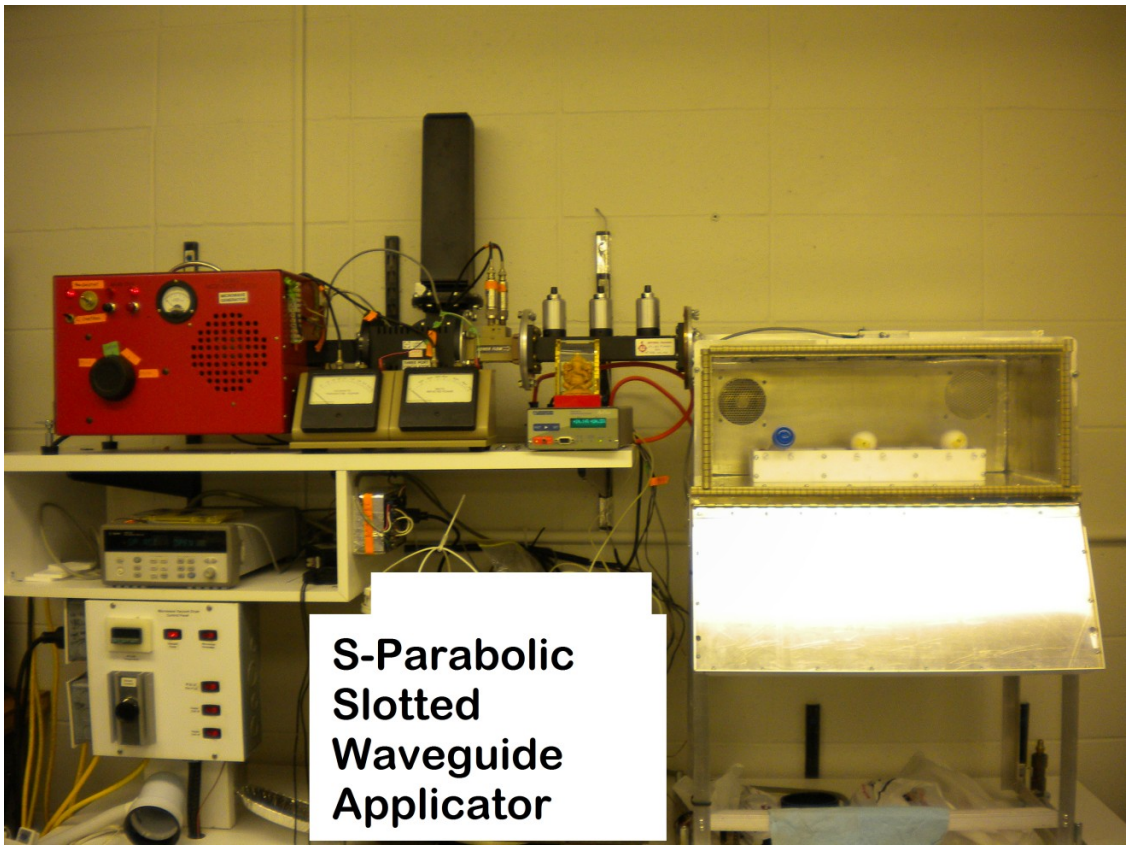

Figure 6.4 S- Parabolic slotted waveguide applicator - complete setup

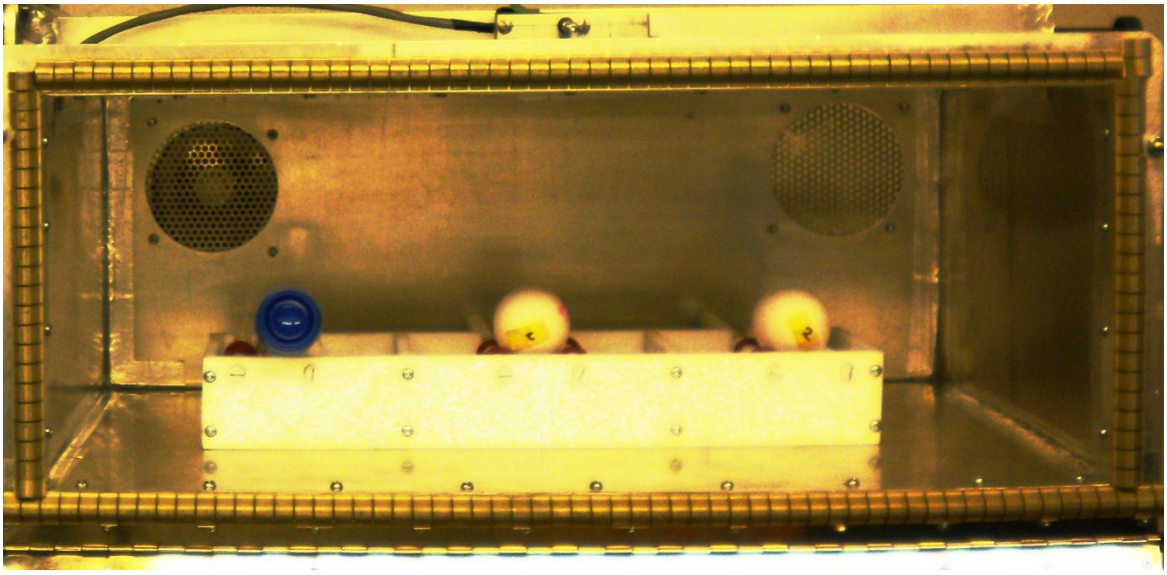

Figure 6.5 Special microwave cavity with an S-Parabolic slotted waveguide

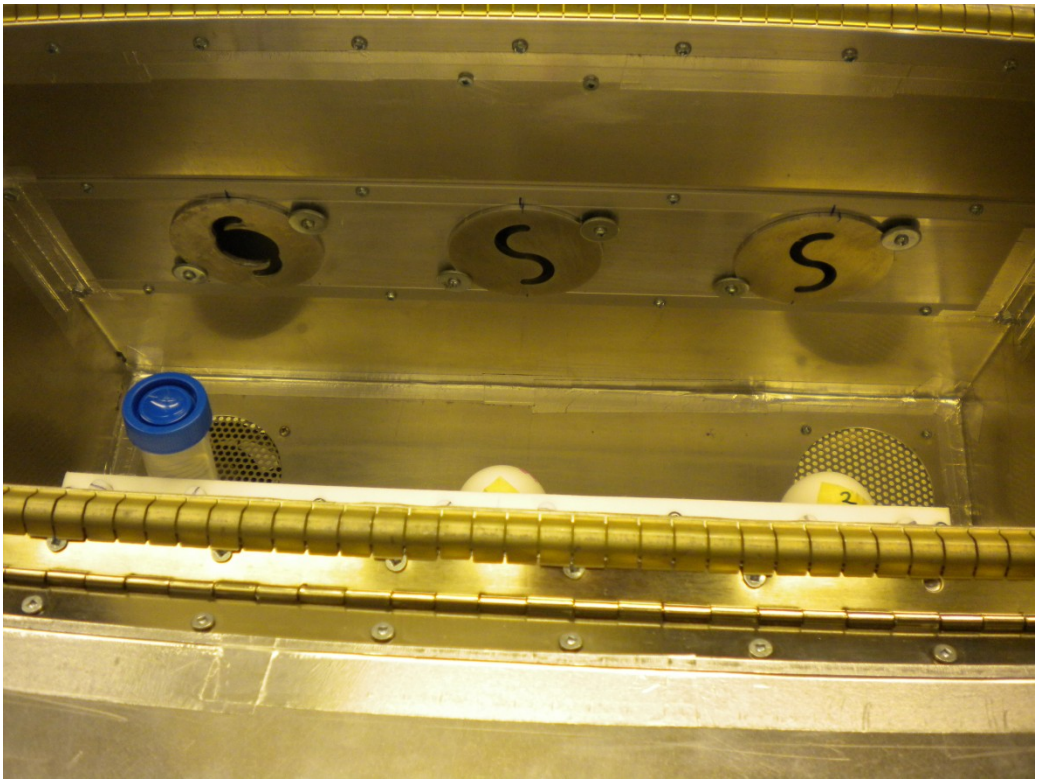

Figure 6.6 S- Parabolic slotted waveguide applicator

6.4.5 Estimation of Microbial Population

Three un-inoculated eggs and three inoculated ones were broken immediately after inoculation and plated in duplicates on EC agar (prepared in the laboratory by adding Agar to the EC broth) to obtain the initial plate count.

Yolk (100 µl) from raw un-inoculated eggs was plated without any dilution as there was no *E. coli* expected to be present initially. Dilutions of 5 and 6 logs made with sterile water were used in plating the inoculated eggs. Similarly 100 µl of the diluted sample was used for plating thereby resulting in 10^6 and 10^6 dilutions respectively.

After two days' incubation, the inoculated eggs were broken and plated. The three untreated eggs were diluted to 5 and 6 logs and 100 µl were plated in duplicates before the microwave treatment. This resulted in the effective dilutions

of 10^6 and 10^7 being plated respectively. Comparatively, 100 µl of both the microwave-treated egg samples were plated without dilution and with 2 log dilutions.

6.5 Results

6.5.1 Growth curve

The initial revitalization/revival of the bacteria from the lyophilized culture was done by measuring optical density of the culture medium at 600 nm, plotting a growth curve and correlating it with the population in CFU ml^{-1}. The results of the growth curve modelling (Figures 6.7 and 6.8) illustrate the CFU ml^{-1} in the inoculums. These were plated and confirmed.

6.5.2 Initial Population

The bacterial population present at the time of inoculation and before the pasteurization process (Table 6.1) show that, as expected, there were no initial CFUs of *E. coli* present in any of the three un-inoculated raw egg yolks. Also there was no CFU detected in 10^7 dilutions of the inoculated eggs. All the 10^6 dilutions of the inoculated eggs plated showed an average initial population a little over 5 logs.

6.5.3 Final Population

The incubation for 2 days at 24 ± 2°C resulted in nearly two log increase in the bacterial population resulting in a little over 10^8 CFU/ml, which is evident from the plate count after incubation.

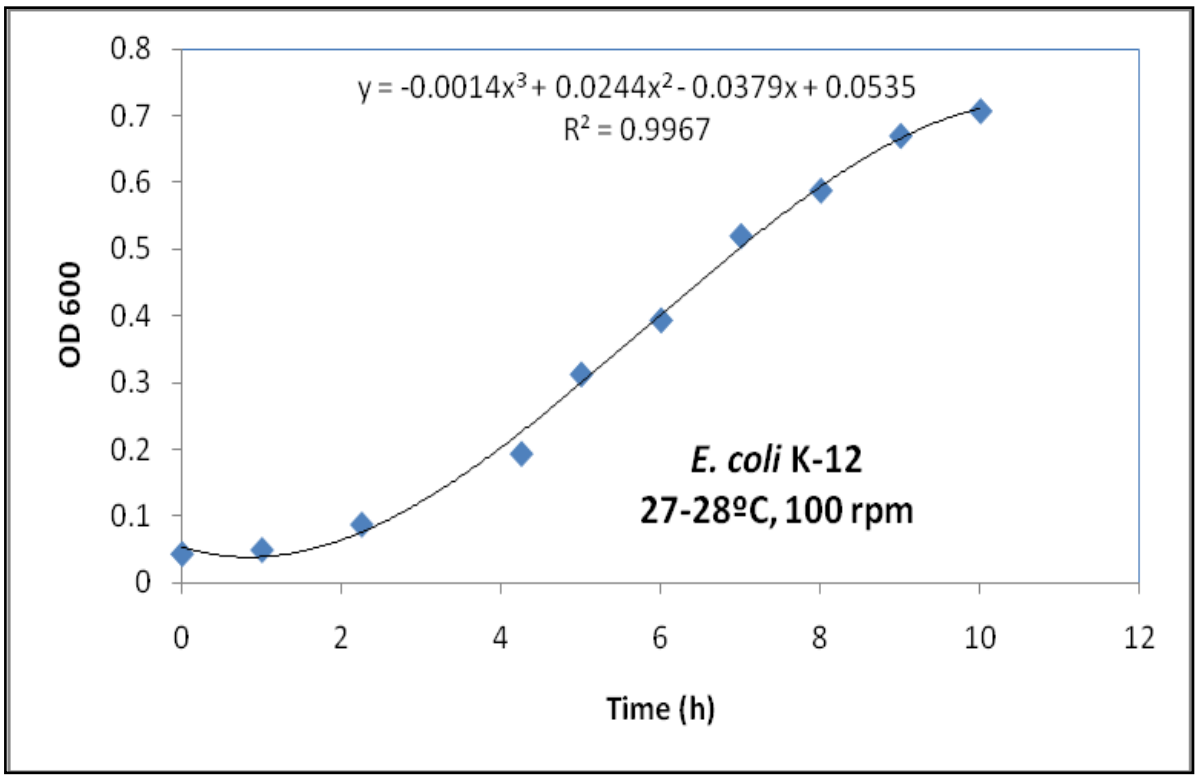

Figure 6.7 Change in Optical Density (OD at 600 nm) over time for *E.coli* K-12

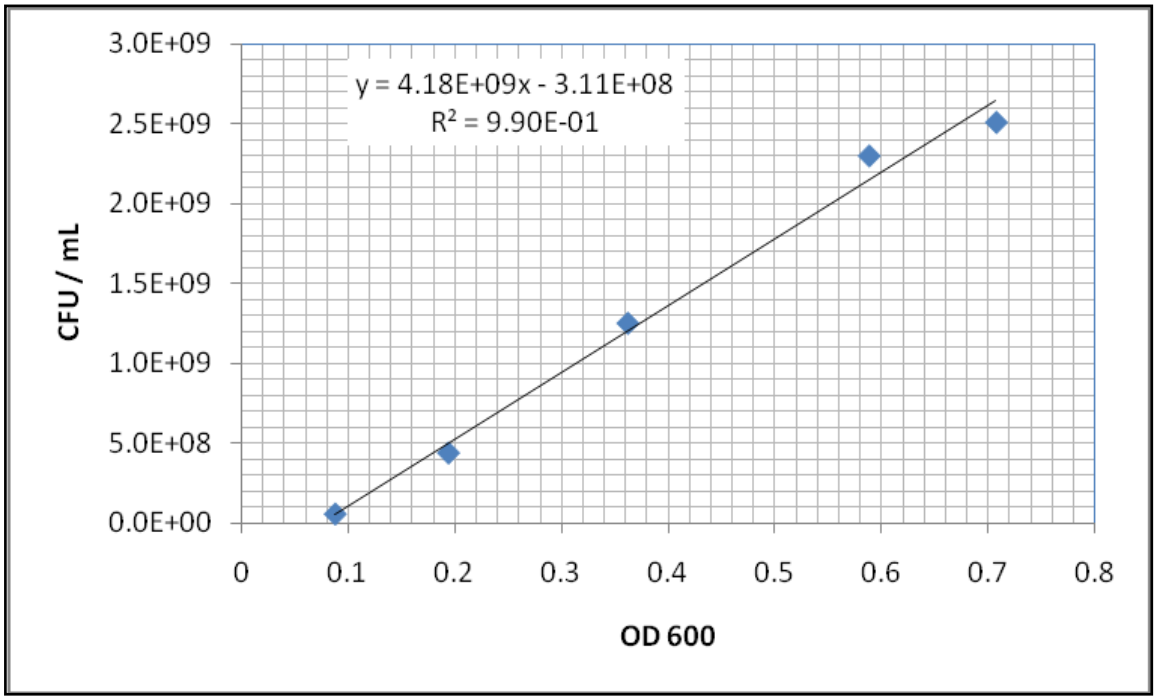

Figure 6.8 Correlation of OD to CFU ml^{-1}

Table 6.1. Bacterial population before and after incubation

	Initial			Final (After incubation @ 24 ± 2°C for 2 days)		
Treatment	Effective Dilution	Mean Plate Count	Mean Population CFU/ml	Effective Dilution	Mean Plate Count	Mean Population CFU/ml
Control -ve	10	0	0	10	0	0
Control +ve	10^6	3.24 ± 0.58	3.24 ± 0.58	10^8	3.33 ± 1.15	3.33 ± 1.15
	10^7	0	x 10^5	10^9	0	x 10^8

Figure 6.9 gives the CFU ml^{-1} after heat treatment using different microwave setups. There were less than 10 CFU ml^{-1} of *E. coli* present in the S-parabolic waveguide applicator microwave pasteurized samples which corresponds to a 7 log reduction, far above the FSIS-USDA pasteurization requirements. The laboratory microwave-heated samples showed a little over 10^2 CFU ml^{-1} which barely meets the FSIS-USDA pasteurization requirements and the domestic microwave oven also had a little over 10^3 CFU ml^{-1} but with coagulations indicating uneven heating (Figure 6.10).

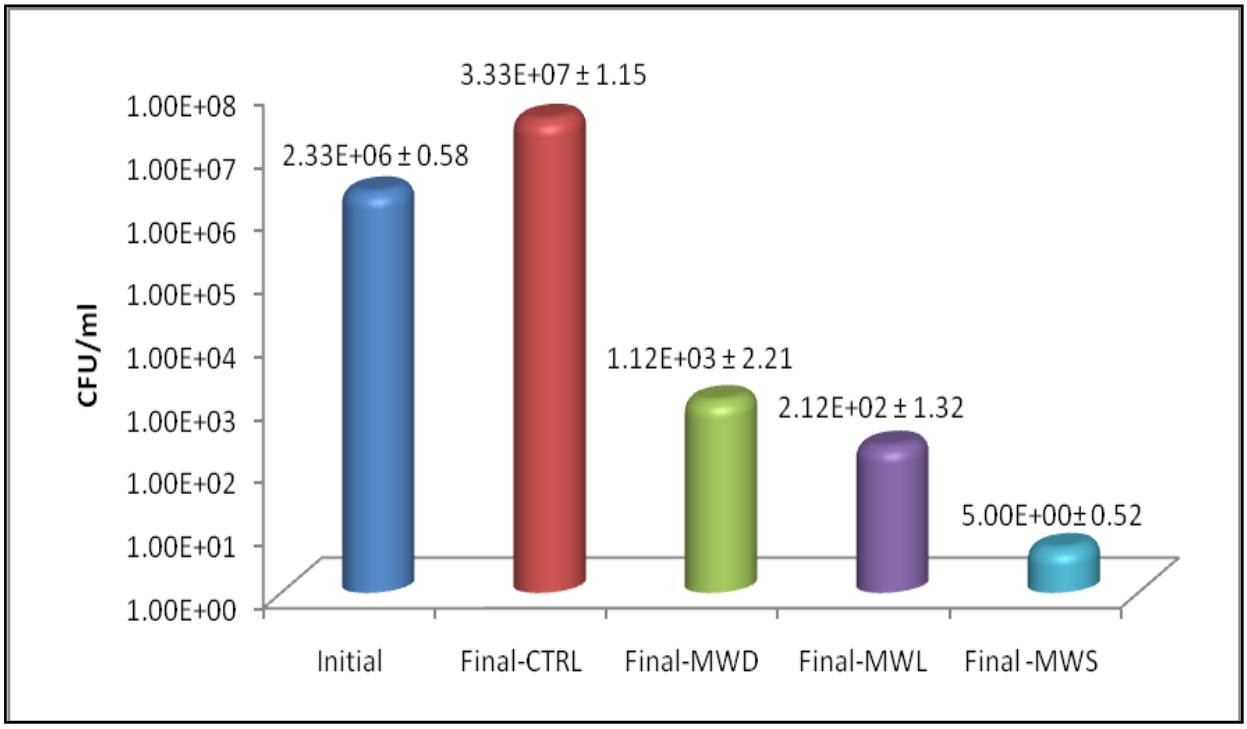

Figure 6.9. CFU ml^{-1} of egg yolk after heat treatment using different microwave setups

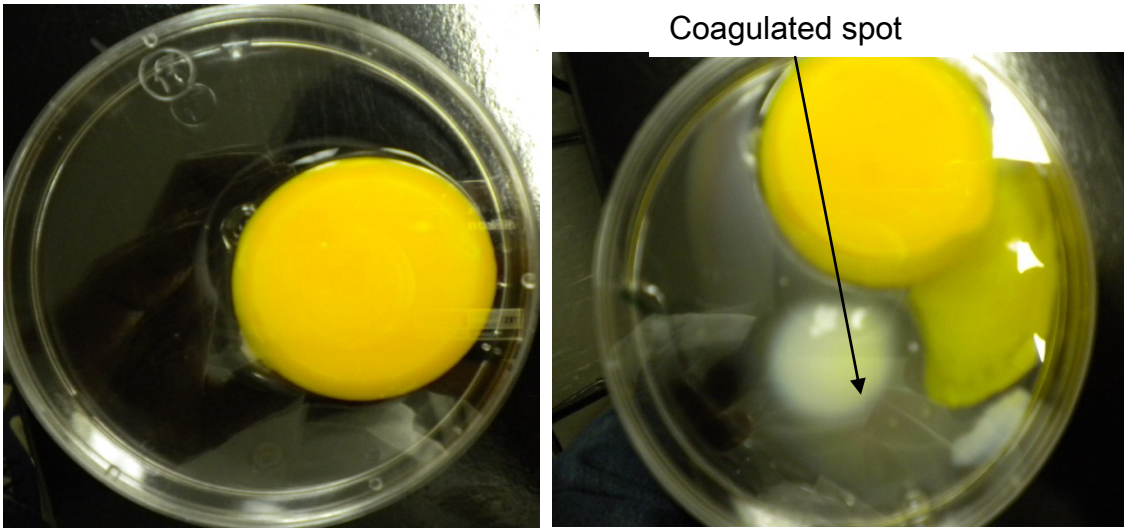

Figure 6.10. Coagulation produced by heat treatment (right) compared to the control (left)

6.6 Discussion

All the microwave pasteurization methods accomplished the target of five log reduction of pathogens which is the target for the pasteurization of eggs (FSIS-USDA, 2006). Comparison among the three microwave heat treatments reveal that the S-parabolic waveguide applicator microwave treatment was much more effective than the laboratory microwave treatment and the domestic microwave heating, as there was less than 10 CFU ml^{-1} after pasteurization using the laboratory setup. This difference may be due to non uniformity of heating in the domestic microwave oven and a few colonies must have survived due to the cold spot generated in the domestic microwave oven.

The difficulty in the monitoring and maintenance of the temperature throughout the pasteurization holding time could be another possible cause of this lower efficacy of the domestic microwave treatment. The simulations of Dev et al (2008) show that the temperature gradient formed within the egg might have equilibrated while being maintained in the water bath at 65°C. However, the shell being a bad conductor of heat, the conduction of heat would not have been effective from the water into the eggs over the holding time of 2.5 minutes.

Lakins et al. (2008) had reported that applying directional microwave technology resulted in a 2-log reduction of S. Enteriditis. Their study involved using directional microwave technology for a 20 s treatment which provided rapid heating of the yolk to 48 ± 4°C depending on egg position. They suggest that the differences due to position of the egg inside the chamber may be decreased with modeling programs available that can indicate proper positioning of the magnetrons to ensure uniform electromagnetic rays throughout the entire testing

area. These results indicate that providing a high temperature for a short time may be an effective strategy for reducing bacterial populations in shell eggs.

Maintaining the required temperature gradient throughout the pasteurization duration is critical for effective pasteurization of the eggs. The computer controlled S-parabolic waveguide applicator microwave setup performed well due to its unique design and also due to the ability to maintain the temperature throughout the pasteurization time.

6.7. Conclusion

The microwave heating of eggs was very efficient both in terms of time and energy, as the entire pasteurization process including the required holding time can be completed within 5 minutes. This helps retain the raw quality of the eggs, as protein denaturation is minimized. The microwave pasteurization technique for in-shell eggs had proven to be very efficient. But it requires specifically-designed equipment for efficiently performance, as uniformity is always an issue while using microwaves. Further research needs to be done in identifying and designing other efficient configurations of microwave waveguides to perform similarly at the industrial scale.

6.8 Acknowledgements

The financial support of Natural Sciences and Engineering Research Council of Canada and Le Fonds Québécois de la Recherche sur la Nature et les Technologies of Quebec is gratefully acknowledged.

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Connecting text

Egg proteins are highly heat sensitive. Any thermal treatment of eggs usually results in significant changes to the functional properties due to the denaturation of proteins and this is more pronounced in the egg white more than the egg yolk. Quantification of these changes could act as a good index for the protein damage sustained by the eggs during pasteurization. After microbiological validation of the designed waveguide applicator and the process parameters, the effect of the same process parameters on the physical properties affecting the functional quality of the egg white needs to be measured and compared in order to quantify the quality tradeoffs for microbial safety.

Chapter 7

QUALITY ASSESSMENT OF MICROWAVE PASTEURIZED IN-SHELL EGGS

7.1 Abstract

In-shell eggs were pasteurized using a custom-built microwave cavity with a slotted waveguide compared with a conventional hot water-bath at 60°C. The quality of albumen and yolk samples from microwave pasteurized, water-bath pasteurized and unpasteurized in-shell eggs (not inoculated) were assessed through visual attributes (turbidity-UV-Spectrometry), viscosity (22°C), thermal analysis (enthalpy of denaturation), and dielectric spectroscopy (200 MHz to 40 GHz). The microwave pasteurized eggs had superior quality in all parameters analysed and also had a much longer keeping quality than unpasteurized eggs.

Keywords: Post-Processing Quality, Egg quality, Microwave, Pasteurization, Shell eggs.

7.2 Introduction

Eggs are popular for the exceptional functional properties of their two major components: the egg white and the yolk. Egg white is used as a foaming, leavening, gelling and/or binding agent in numerous food preparations. Egg white proteins are the most heat sensitive components of an egg. Egg yolk has good emulsifying and binding properties (Li-Chan et al, 1995). The physical properties like whipability, foamability, foam stability, viscosity etc., which contribute to egg's functional properties, and make them essential ingredients in various food

products, are severely affected by high temperatures treatments (Van der Planken et al., 2006).

Due to its extraordinary nutritive value, eggs remain a potential host for pathogens like *Salmonella enteritidis*. More than 90% food-borne Salmonellosis, caused by *Salmonella enteritidis* occurs through shell eggs (Schroeder et al, 2005; Woodward et al, 1997). Most of the *Salmonella enteritidis* outbreaks have involved Grade A eggs that were washed and disinfected and also met the requirements of the State for shell quality (St Luis et al., 1988). The probability of fresh eggs having Salmonella varies from 0.005% (Mermelstein, 2001) to 1% (Griffiths et al. 2005) depending on various factors involved in the egg production.

Thermal processing methods are the most widely used technique for destroying microorganisms and imparting foods with a lasting shelf-life, amongst which pasteurization has its own prominent and specific applications. Pasteurized foods are safety-assured for the consumer within the recommended storage period and storage conditions. Today various techniques are applied for the pasteurization and thermal processing of foods. The conventional methods of thermal processing of foods result in peripheral over heating before the food in the centre reaches the required temperature. This is potentially a great problem in pasteurization, especially when it comes to the quality of shell eggs.

The Food Safety and Inspection Service (FSIS) of United States Department of Agriculture (USDA) recommends heating the egg white and the egg yolk to 57.5°C and 61.1°C, respectively, for 2.5 minutes to ensure egg safety against Salmonella and other food borne pathogens (FSIS-USDA, 2006). The existing method of pasteurizing the shell eggs uses immersion in hot water at

60°C for 20 minutes, results in overheating of the egg white and partially cooked eggs (Mermelstein, 2001; Hou et el., 1996). Eggs contain different protein fractions namely conalbumin, ovalbumin, ovotransferin, ovomucoid, ovomucin, globulins, lyzozyme, etc. that contribute to the functional properties of the egg white as a whole (McDonnell et al, 1955; Cunningham et al, 1995). The denaturation of some of these proteins starts at temperatures as low as 45°C.

Studies on the physico-chemical changes arising from heat treatment of egg white have revealed that at lower temperatures (< 50°C) these changes were only temperature dependent, but at higher temperatures (> 50°C) the time factor, also plays an important role, indicating a time-temperature-dependent level of denaturation with an equilibrium (denaturation-saturation) time (Van der Plancken et al. 2006). Therefore minimizing total time of heating is crucial in generating a better quality pasteurized egg white.

Dev et al. (2008) demonstrated that microwave heating is an excellent alternative to overcome the problem of peripheral overheating during shell egg pasteurization. Also the FSIS recommendation of heating up the yolk to a higher temperature (61.1ºC) was achievable without heating the egg white beyond its recommended pasteurization temperature (57.5ºC). The risk of pressure build-up within the egg shell when heated using microwaves is not inevitable within the pasteurization temperatures (Fleischman, 2004; Rehkopf, 2005). A comprehensive assessment of the functional quality of the microwave-heated eggs can be done by examining the changes in the physical properties responsible for that quality.

Heat induced changes are more pronounced in the egg white than the egg yolk within the pasteurization temperature limits and hence egg white would be a

suitable indicator for the comparison of such changes (Li-Chan et al., 1995). Therefore this paper focuses on comparing the physical properties of microwave and water bath in-shell pasteurized egg white with that of raw egg white for any heat induced changes. The targeted physical properties were turbidity, viscosity, enthalpy of denaturation, and parameters quantified by dielectric spectroscopy.

7.3 Materials and Methods

In-shell eggs were pasteurized using a custom-built laboratory microwave oven setup with a specially-designed slotted waveguide applicator or using a hot water bath maintained at 60°C. Effects of heat treatments on the physical properties affecting the functional quality of the egg white recovered from the treated eggs were measured and compared to those of fresh untreated egg white.

7.3.1 Egg samples

Fresh whole eggs, within 3 days of grading and packing (identified from the best before date stamped on the eggs, which is usually 35 days from the date of packing) (CEMA, 2004), used in this study were procured from a local market and kept in a refrigerator at 5°C until used. They were all of Canadian Grade A eggs, size large, each with a mean mass of 60±2 g. Prior to pasteurization, the eggs were brought to room temperature of about 24°C by placing the opened carton on the laboratory counter for a period of 3 to 4 hours (tested by breaking and measuring inner temperatures of 3 representative samples) before applying the heat treatments. This was done to replicate the possible use of this technique in the industry, wherein significant amount of energy can be saved by following such a procedure.

7.3.2 Heat treatments for pasteurization

Two heat treatments for the pasteurization of in-shell eggs were investigated and compared. Each treatment was done in triplicate (i.e.) three eggs were used for each treatment for the measurement of each parameter within the scope of this study. The first treatment consisted of heating in-shell eggs in a custom-built laboratory microwave oven setup with a specially-designed slotted waveguide applicator working at 2450 MHz, using a power density of 2 W g^{-1}. In-shell egg white was heated for 1.25 minutes to raise the temperature to 58℃ and held at 58±0.5℃ for 2.5 minutes, by periodically turning the microwave cycles on and off, as per FSIS-USDA (2006) recommendations. Temperature measurements were not done during the treatments as an optimised algorithm developed using a microbial validated finite element method for the microwave pasteurization of eggs was used to control the on/off cycles (Dev et al, 2008a and 2009) and the microwave operation was controlled by the computer running HPVEE (Agilent) object-oriented programming language to maintain the desired process temperature. The schematic of this setup is shown in Figure 7.1.

The second treatment consisted of immersing the in-shell egg in a temperature-controlled water bath maintained at 60°C for a period of 20 minutes (Schuman et al., 1997). These eggs were left intact without any inserted probes as this was already a commercially practiced technique, approved by FSIS-USDA.

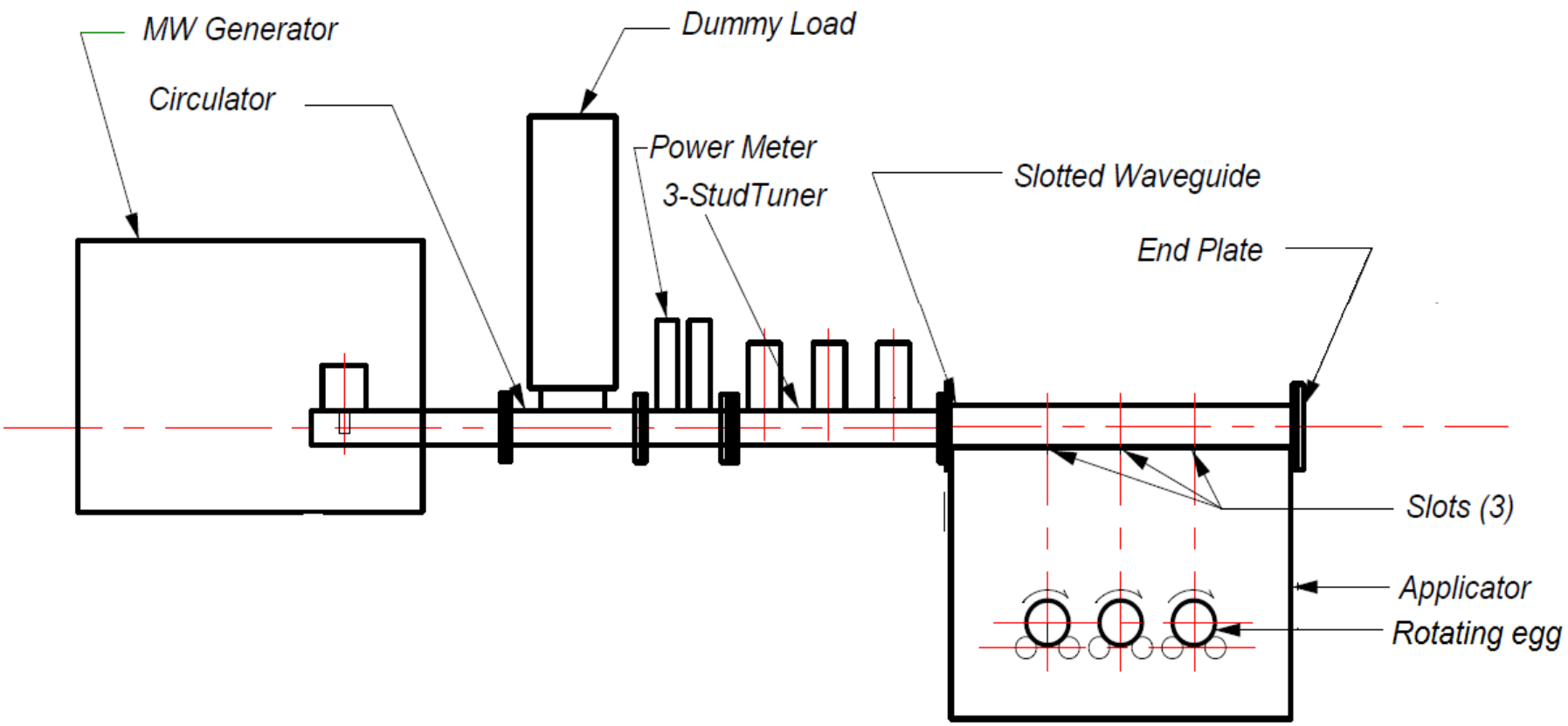

Figure 7.1 Schematic of a slotted waveguide microwave pasteurization setup

It is clear that the temperatures reached by the egg components using the microwave and water bath heating are not identical. But the objective of this study was to compare the properties of the egg white pasteurized in-shell by the proposed (microwave) technique with that of the commercially practiced (water bath) technique, both meeting the FSIS-USDA pasteurization requirements/recommendations. Immediately after both these heat treatments, the shell eggs were immersed in a vessel of cold water (5°C) and left there for 10 minutes. This was done to ensure that the extent of heat damage to the proteins did not continue after the pasteurization treatment.

7.3.3 Measurements of the egg white physical properties

The physical properties which can be related to the functional quality of the egg white of in-shell heat treated and untreated eggs were measured and compared. Eggs were cracked carefully and the egg white was collected in small beakers. Shell and yolk was discarded. All measurements took place in triplicate. Parameters measured to assess the functional properties of egg white were: enthalpy of protein denaturation, foam density and foam stability, viscosity, turbidity and dielectric properties.

7.3.3.1 Enthalpy of protein denaturation

The enthalpy of denaturation is the net value of the combination of endothermic reactions, such as the disruption of hydrogen bonds, and of exothermic processes, such as the breakup of hydrophobic interactions and protein aggregations. The resulting residual enthalpy has been correlated to the remaining content of ordered secondary structure of a protein (Van der Plancken et al, 2006). Comparative analysis of thermograms between pasteurized and unpasteurized egg constituents may thus indicate damage to proteins.

The instrument used to measure the enthalpy of denaturation was a TA Instruments Q100 Differential Scanning Calorimeter (NewCastle, DE, USA) (Figure 7.2) operated with the TA Instruments Q100 DSC 7.0 Build 244 software. Untreated and heat-treated samples were first placed in aluminium pans (20 µl per pan) and then hermetically sealed. The pans were transferred to the instrument pan holder and heated from 20°C to 120°C at a constant rate of 10°C min^{-1}. An empty pan was used as a reference. The sample residual enthalpy was the recorded at the denaturation temperature of 83°C (Van der Plancken et al., 2006).

Figure 7.2 TA Instruments Q100 Differential Scanning Calorimeter

7.3.3.2 Viscosity

Viscosity was measured at 22°C with a computer-controlled TA Instruments AR2000 Advance Rheometer (NewCastle, DE, USA). Any minute change in viscosity can be detected and attributed to changes in protein structure (denaturation). Measurements of viscosity could not be made above 45ºC as any further increase in temperature might lead to further denaturation. For each sample, measured viscosity values were plotted against temperature and analyzed.

7.3.3.3 Foam density and foam stability

Foam density is a measure of the thickness of the foam, which gives a clear picture of the quantity of air incorporated in the egg white foam. This is an important factor that represents the aerating properties of the egg white in its food applications. The stability of the egg foam is a crucial parameter for the functional quality of the egg white. The use of egg white is highly dependent on its foam stability in many of its commercial applications in the food industry (McDonnell et al. 1955).

Each egg white sample was foamed in a graduated cylindrical beaker (500 ml) with a Braun 60 Egg Beater (USA). The foaming process consisted of beating 50 g of egg white for 2 minutes at a speed of 2000 rpm. The foam density was then measured by weighing the mass of the known volume of foam in the beaker and the foam stability was taken as the quantity of liquid drained as a function of time from the completion of foaming. Foam stability measurements were taken for 180 minutes after foaming. Figure 7.3 shows the experimental setup for the determination of foam stability.

Figure 7.3 Experimental setup for measurement of foam stability

7.3.3.4 Turbidity

Turbidity is a direct measure of the extent of protein coagulation, as coagulated proteins are opaque and reduce the transmittance of light through the egg white. The amount of light absorbed (absorbance) is a function of the turbidity of a liquid. The absorbance of the heat-treated and untreated egg white samples was measured at 24ºC, at 650 nm (Van der Plancken et al, 2006) using a Biochrom Ultra spec 2100 Pro spectrophotometer. Plain demineralised water was used for calibration, such that an absorbance (turbidity) of 0% corresponded to a totally clear solution.

7.3.3.5 Dielectric properties

The change in dielectric properties is considered to be a good indicator of the extent of denaturation of the egg white proteins (Bircan et al, 2002). An open-ended coaxial probe technique was used to measure and compare the dielectric properties of heat-treated and untreated samples (Figure 7.4). An Agilent Network Analyzer Model 8722ES equipped with an 85070E Dielectric Probe Kit and an Electronic Calibration Module (Agilent, Palo Alto, CA, USA) was used to measure the dielectric properties from 200 MHz to 40 GHz of egg white and yolk samples from treated and untreated eggs. Measurements were made on six eggs per treatment and the data was analyzed using standard statistical procedures. This instrument was controlled with the Agilent 85070D Dielectric Probe Kit Software Version E01.02.

7.3.3.6 Keeping quality of eggs

Viscosity at 20°C, turbidity and foam density of both the microwave pasteurized and waterbath-pasteurized eggs stored at 5°C were measured at an

interval of 7 days for 8 weeks and compared with those of the unpasteurized eggs.

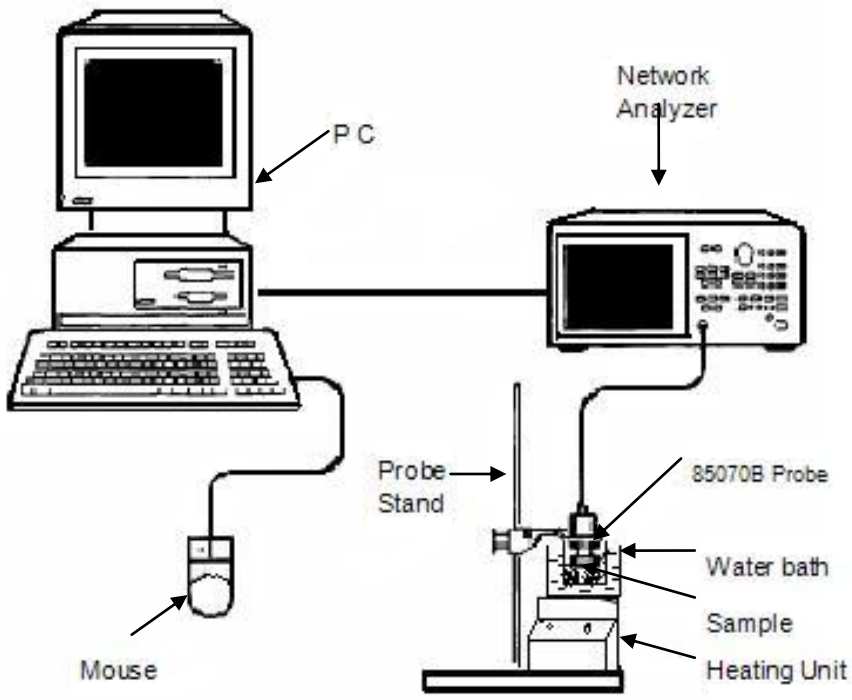

Figure 7.4. Dielectric properties measurement setup

7.3.4 Data analysis

All the data obtained were statistically analyzed using MATLAB 7.8 software from Mathworks. Analyses of variances followed by Duncan's multiple range tests were conducted to locate significant differences among means. In all comparisons, significant deviations from mean values obtained from untreated egg white were considered to have an effect on egg white functional properties.

7.4 Results and Discussion

7.4.1 Enthalpy of protein denaturation

As shown in the Figure 7.5, reductions in residual enthalpy indicated that the heat treatments had partially denatured the egg white proteins of all heat treated samples. However, microwave-treated samples exhibited less such reductions than samples heated in the hot water-bath. This implied that less denaturation occurred in the microwave-heated in-shell egg. These results corroborate the work of Van der Plancken et al. (2006) with heat-treated egg white.

The difference in mean enthalpy of the egg white between microwave-heated and the untreated (raw) in-shell eggs was not significant ($P>0.01$), whereas the water-bath-heated in-shell eggs showed significant ($P\leq0.01$) lower enthalpy than the others.

7.4.2 Viscosity

Egg white viscosities of heat-treated eggs were lower than those of untreated eggs (Figure 7.6), and decreased with temperature and the level of protein denaturation. This is due to fact that the denatured proteins de-solubilise resulting in a lower viscosity (Pitsilis et al., 1975). At higher temperatures, differences among the treatments were lower than at lower temperatures (Figure 7.6), where all the three samples had a considerable difference amongst themselves.

The results of the statistical analysis (ANOVA & Duncan's Test) were similar to that of the enthalpy of denaturation. The viscosity of in-shell microwave-heated egg white was not significantly different ($P<0.05$) from viscosity of the untreated eggs, while the water-bath-heated samples were significantly different ($P<0.05$) from either of the other two.

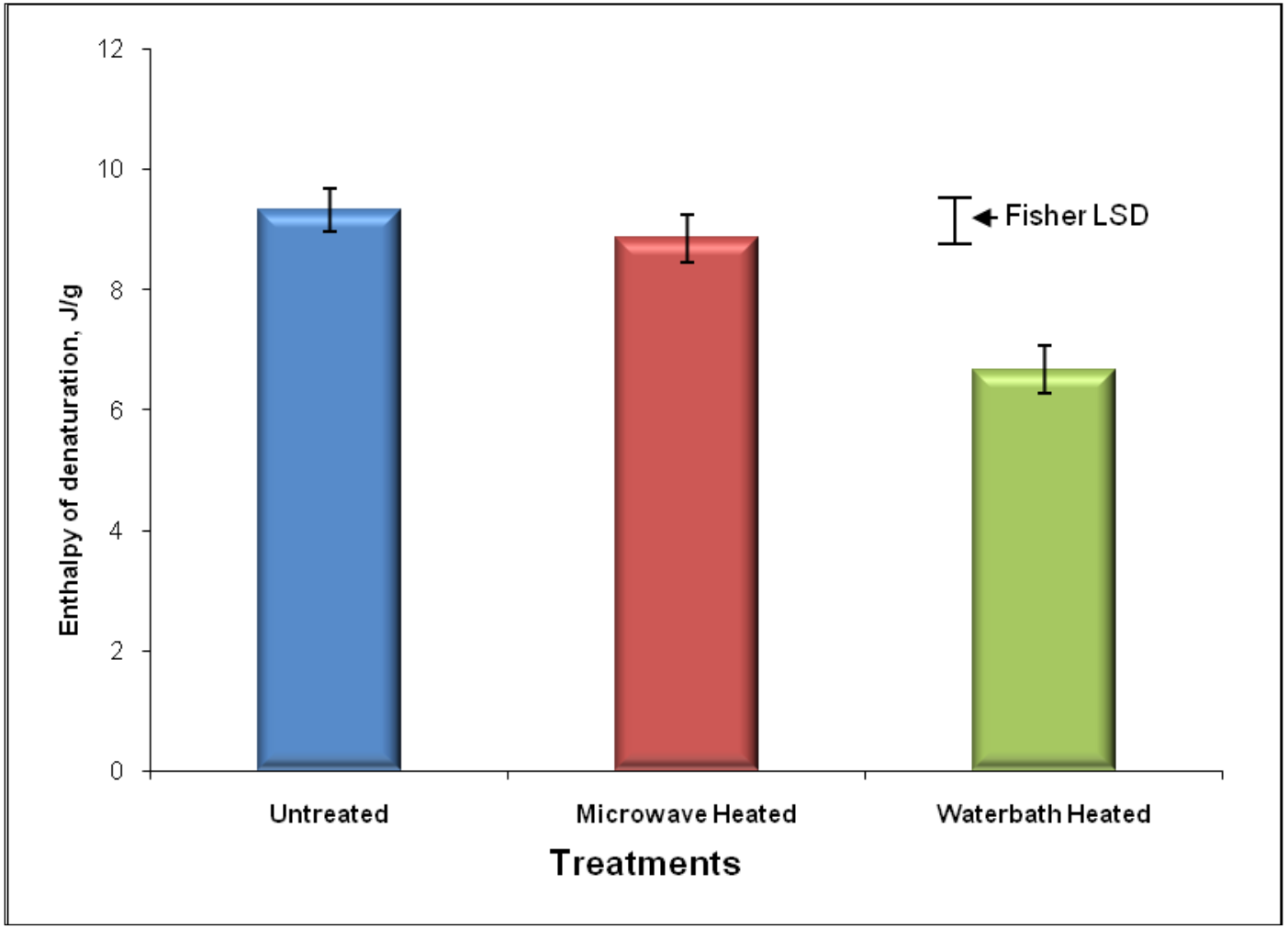

Figure 7.5 Enthalpy of denaturation of untreated and in-shell pasteurized egg whites

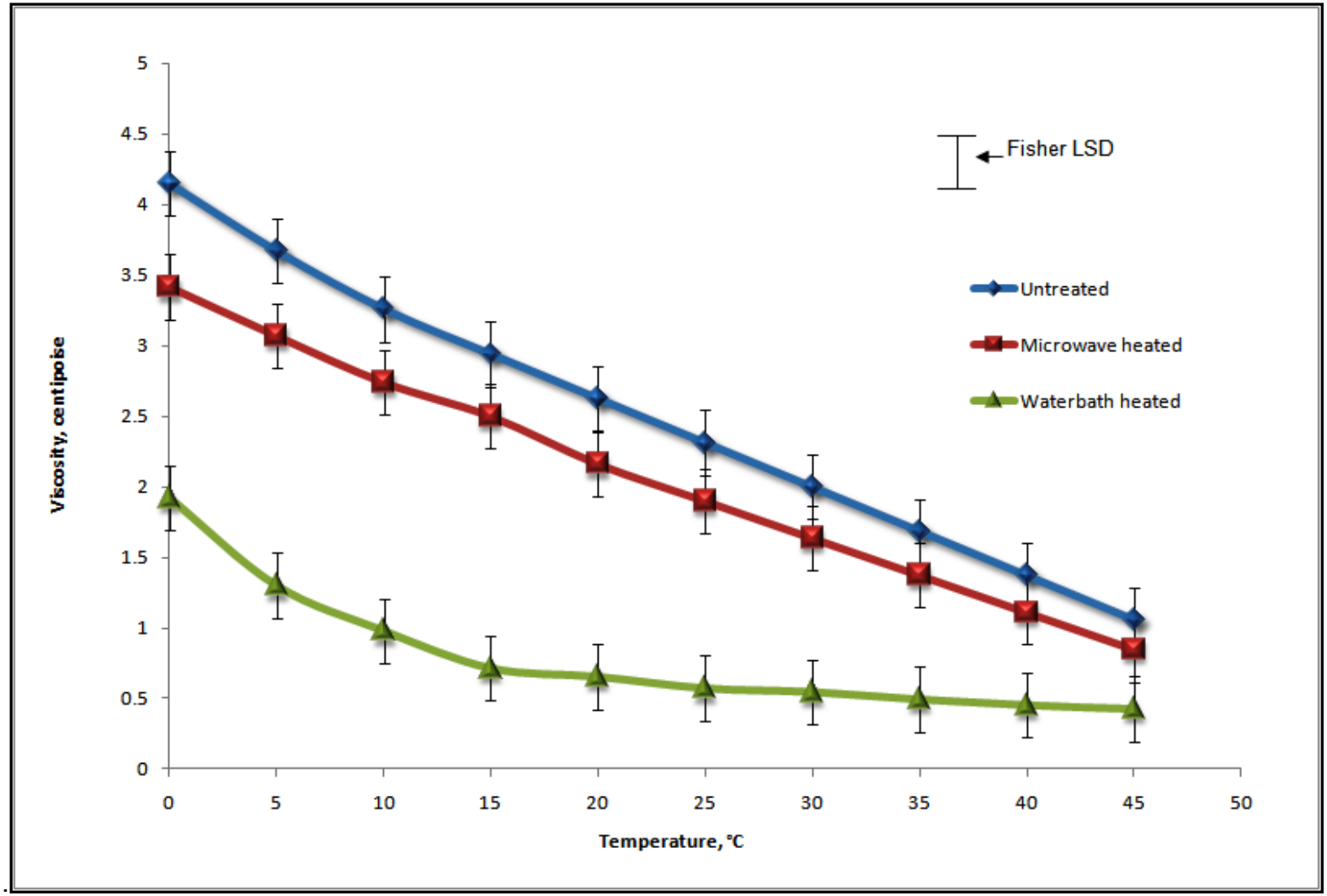

Figure 7.6 Viscosity of untreated and in-shell pasteurized egg white

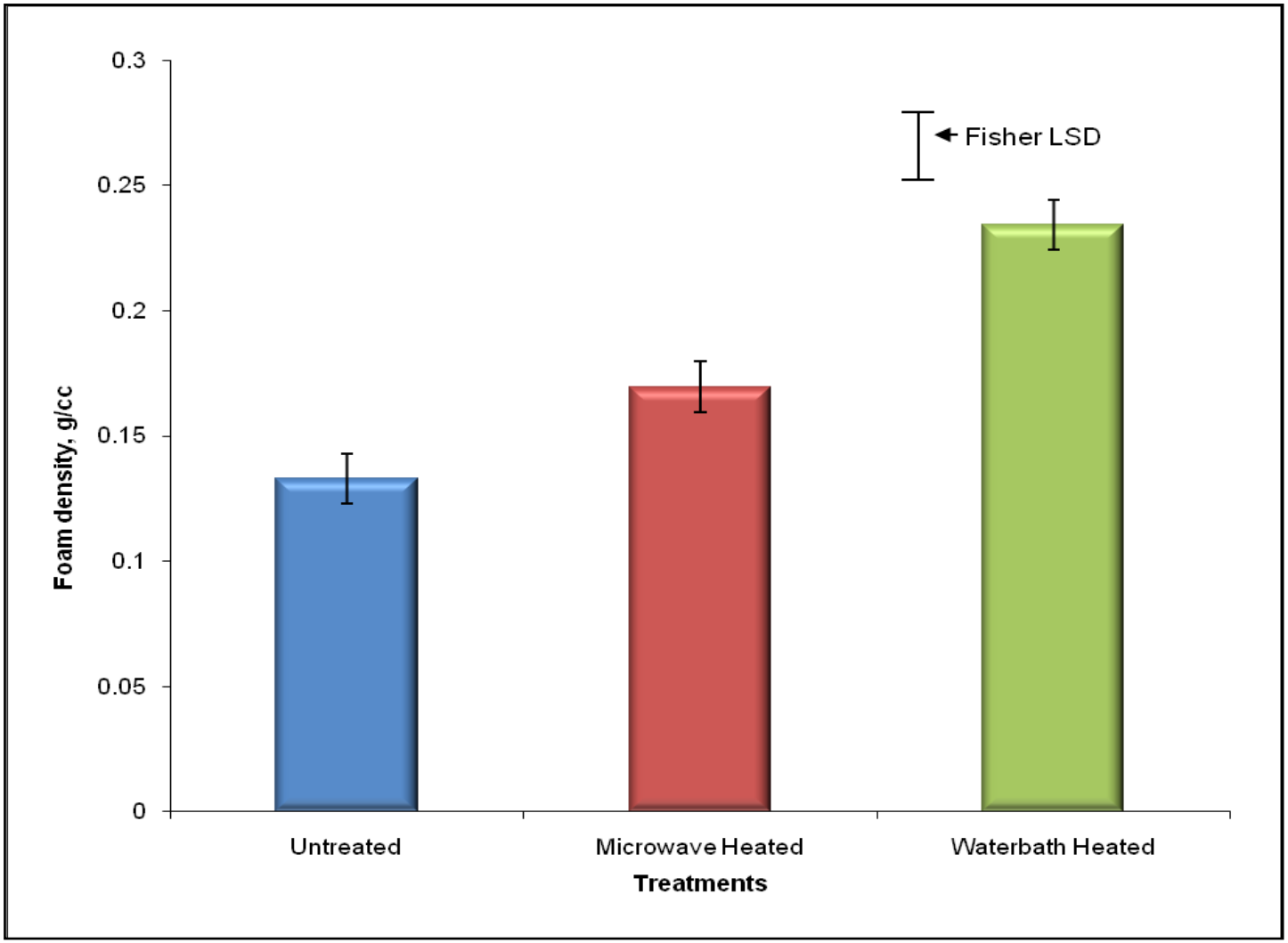

Figure 7.7 Foam density of the egg white of untreated and in-shell heated eggs.

7.4.3 Foam density and foam stability

The foam density of the egg white from microwave-heated in-shell egg was lower than that of water-bath-heated in-shell eggs making them more suitable for commercial applications.. The foam stability, reported as the volume of drained liquid as a function of time, indicated that microwave-heated samples had a foam stability similar to that of untreated eggs (Figure 7.8), whereas the waterbath-heated samples had poor foam stability. This is due to the fact that the de-solubilisation of the denatured proteins results in a colloidal suspension with macroscopic particles that interfere with the surface tension of the bubbles formed making the foam less dense and less stable

Statistical analysis (ANOVA & Duncan's Test) revealed that the differences in foam stability between the microwave-heated in-shell egg and untreated ones was not significant ($P<0.05$). The stability of the foam made with the egg white of the eggs heated in the waterbath was significantly lower than that of the two others.

7.4.4 Turbidity

The analysis performed on the turbidity of the egg white samples measured as the absorbance at 650 nm indicated that the microwave-heated in-shell egg white showed greater transmittance than waterbath-heated ones (Figure 7.9). This implies that the extent of denaturation was much less in the microwave-heated samples as coagulation due to denaturation increases turbidity (Van der Plancken et al, 2006).

Differences in mean turbidity values for the egg white taken from untreated, microwave-heated or waterbath-heated eggs were all significant at the 0.01 level. Turbidity of the microwave-heated samples was closer to that of untreated eggs than to waterbath-treated eggs.

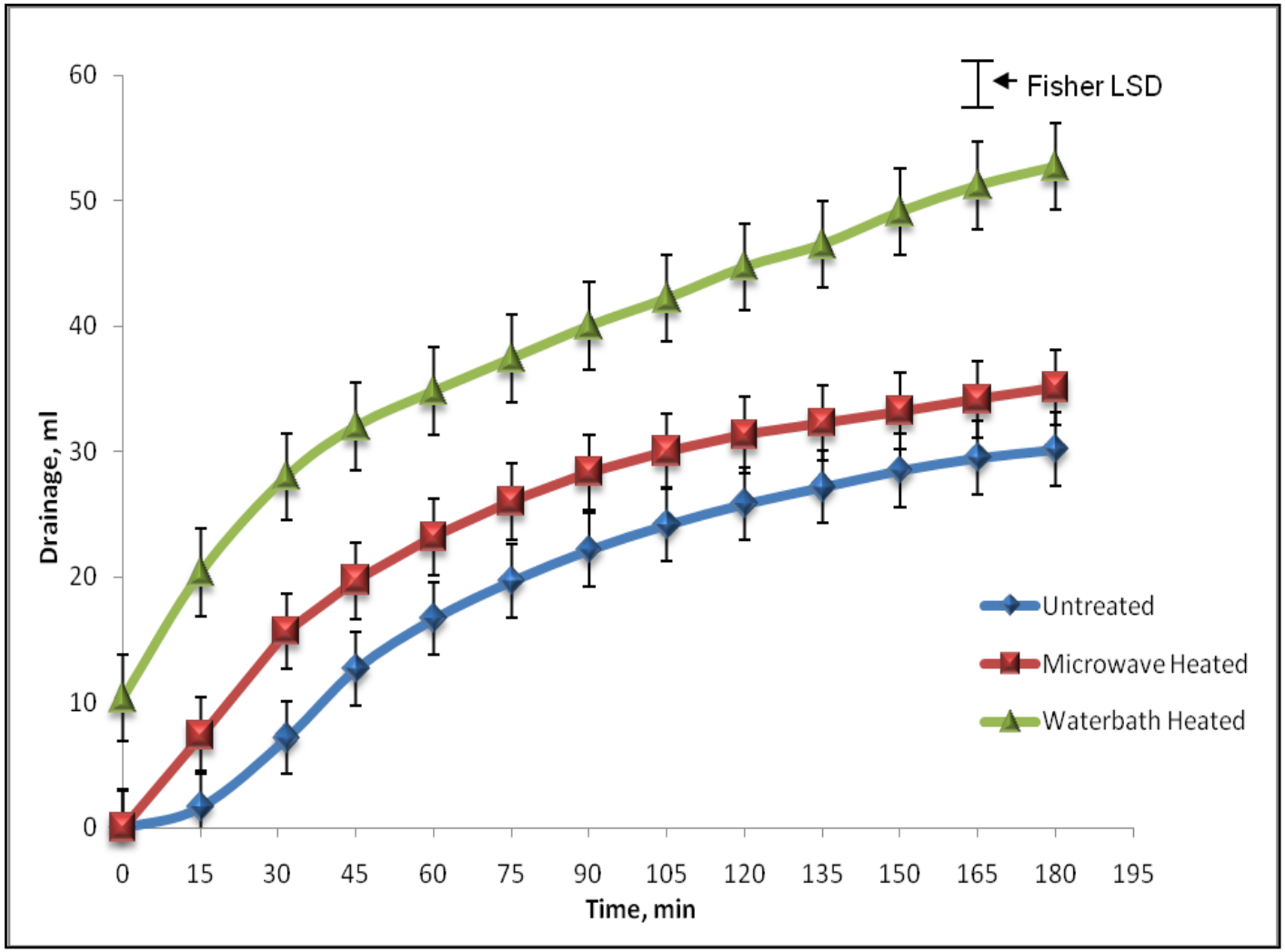

Figure 7.8 Foam stability of the egg white of untreated and in-shell heated eggs.

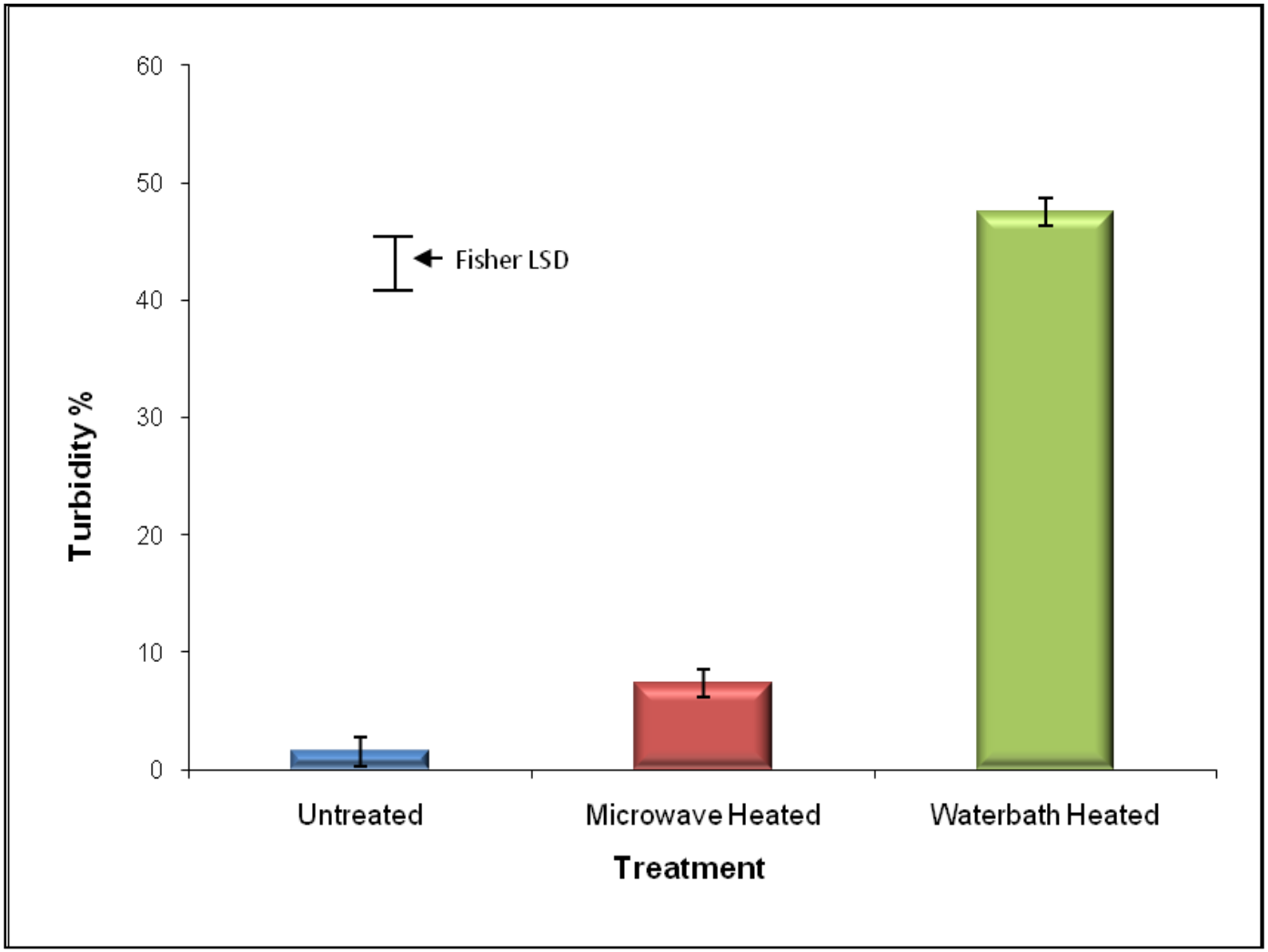

Figure 7.9. Percent turbidity (650nm) of untreated and in-shell heated egg white

7.4.5 Dielectric properties

The dielectric constants and loss factors of the egg white of untreated eggs and of eggs heated in a microwave oven or in a hot water bath as a function of temperature are presented in Figures 7.10 and 7.11. The dielectric properties of the egg white of microwave-heated in-shell eggs, measured at 2450 MHz, were similar to those of untreated eggs.

The dielectric properties egg whites from hot water-treated eggs showed a completely different trend as they were directly proportional to the temperature. This behaviour was associated with a greater denaturation of proteins in these samples. The dielectric properties which were inversely proportional to the temperature when raw (untreated) eggs were tested became directly proportional to temperature when the eggs were denatured (Bircan, 2002).

Statistical analysis (Generalized Linear Model on curves and ANOVA and Duncan's test on the slopes and intercepts) revealed significant differences ($P<0.05$) among all the samples.

7.4.6 Keeping quality of pasteurized eggs

7.4.6.1 Change in viscosity of the egg white over time

In general, the viscosity of the egg white decreases over storage time (Jones, 2007). But the change in viscosity is more pronounced with unpasteurized eggs compared to pasteurized ones (Figure 7.12). Thus pasteurization acts as a means of extending the shelf life of eggs. Kemp et al. 2006 found similar results with the storage of eggs for 8 weeks.

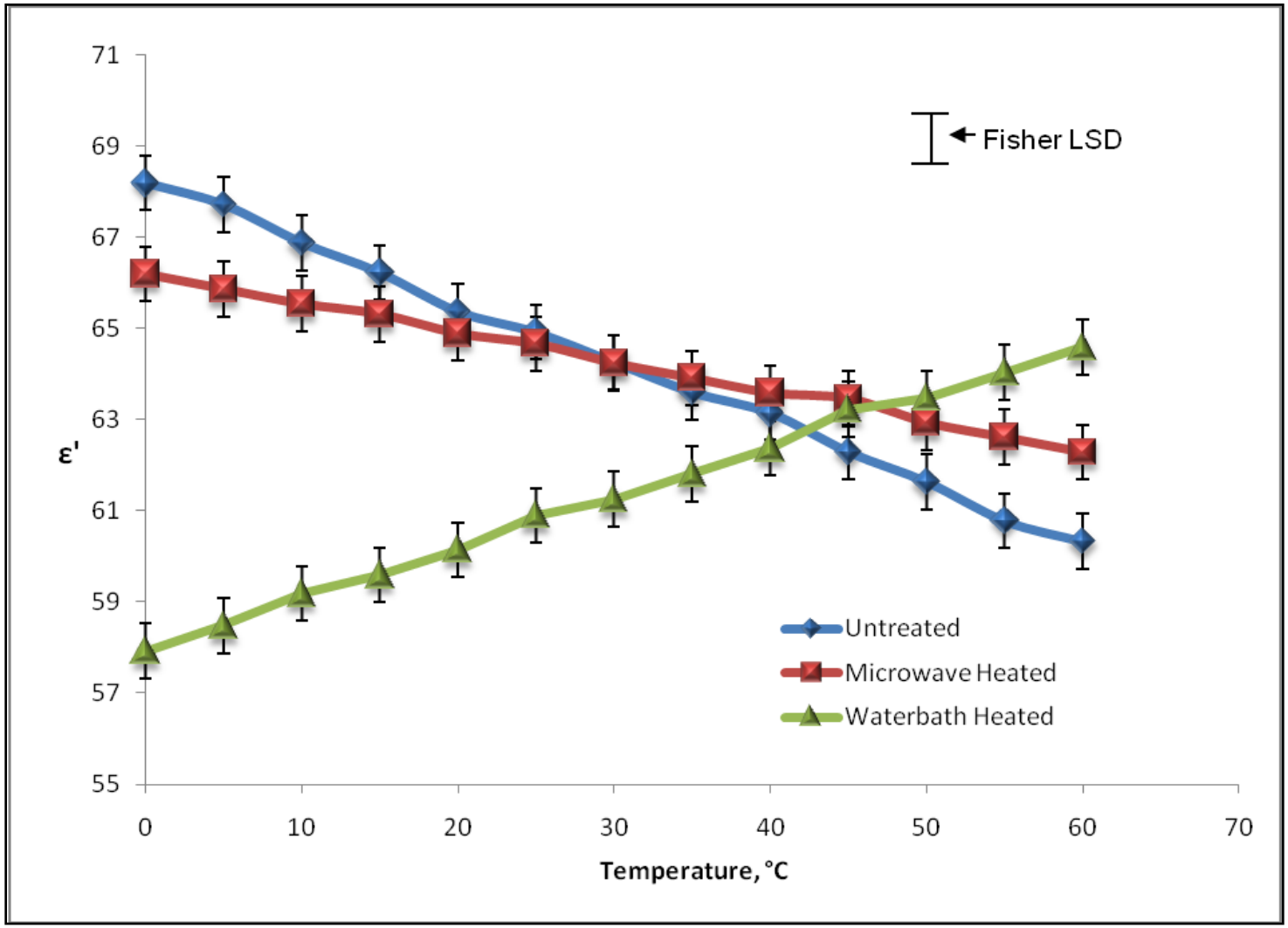

Figure 7.10 Dielectric constant (ε') of the egg white of untreated and in-shell heated eggs

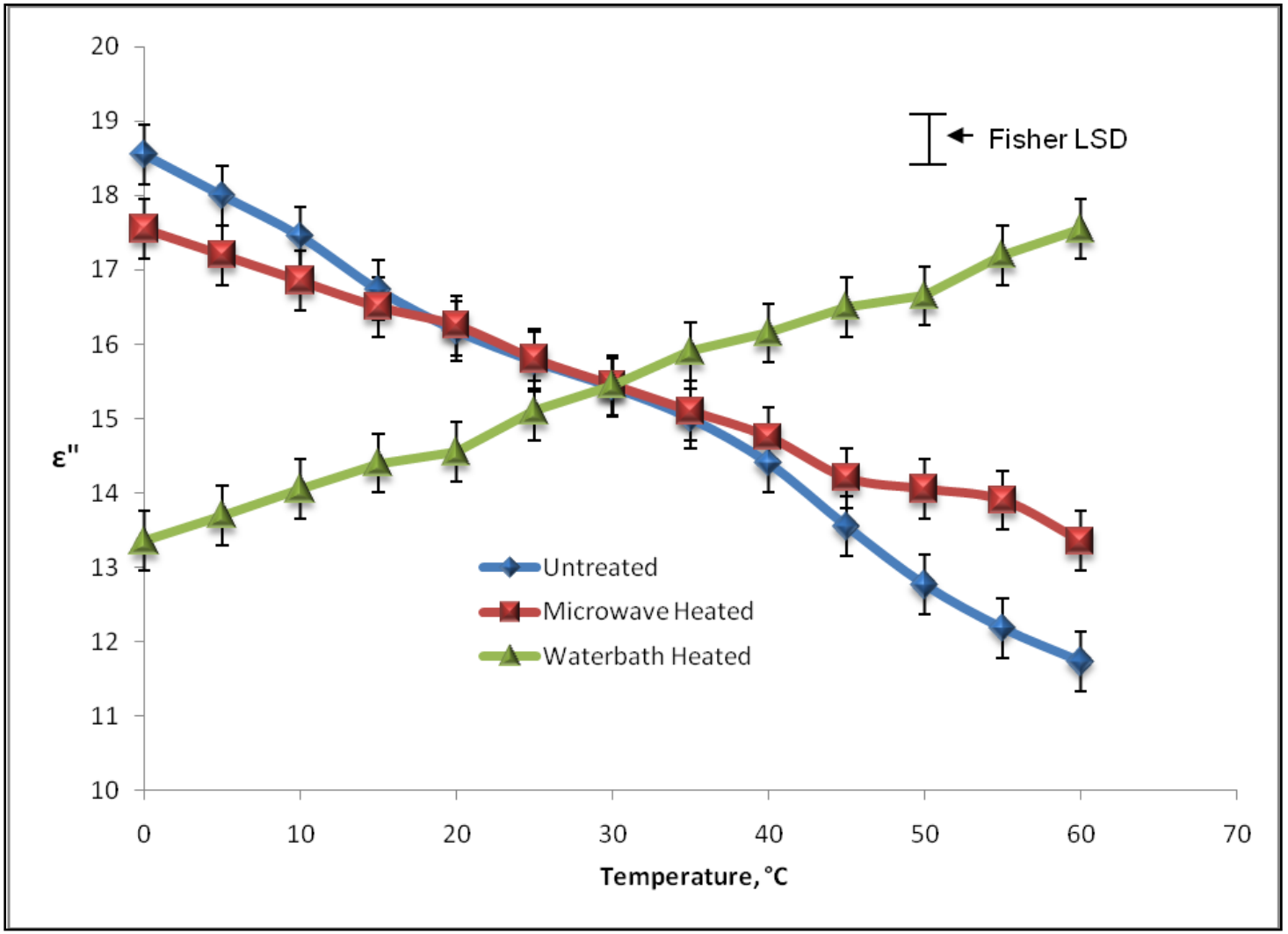

Figure 7.11 Dielectric loss factor (ε") of the egg white of untreated and in-shell heated eggs.

7.4.6.2 Change in turbidity with time

The turbidity of fresh eggs also has a general tendency to decrease with time mainly due to the release of carbon dioxide into the air sac. Interestingly in both the heat-treated samples, turbidity increased slightly after the first week. But after the first week, there was no significant further change in turbidity of any of the treatments (Figure 7.13). This slight initial increase may be due to some heat induced biochemical reactions that continued at a very slow rate after the heat treatment. Abdel-nour et al. (2009a and 2009b) reported an increase in absorbance of the eggs at the NIR wavelengths using hyperspectral imaging during the first two weeks of storage.

7.4.6.3 Change in foam density with time

The foam density of all the samples increased with time (Figure 7.14) as the foam density has an inverse relation to the viscosity. From the beginning, there was no significant ($P<0.05$) difference in the foam density of microwave pasteurized samples compared to the unpasteurized ones. The rate of increase in foam density was lower compared to that of the unpasteurized and waterbath-pasteurized samples. Li Chan et al (1995) state that because of this tendency of the fresh eggs to lose their foam density and foam stability with time they are not preferred for usage in cakes and other bakery products after two weeks of storage.

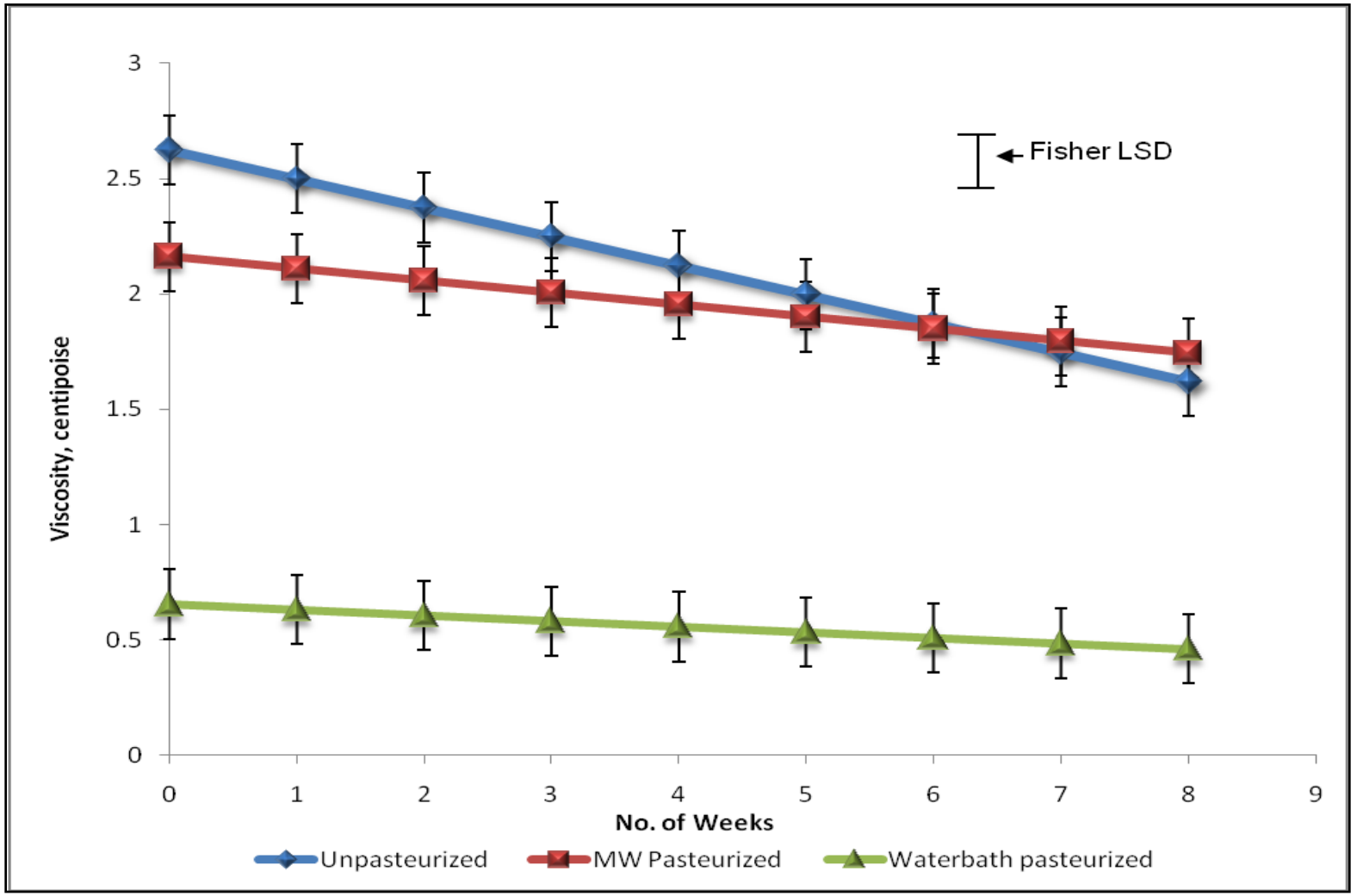

Figure 7.12 Change in viscosity with time

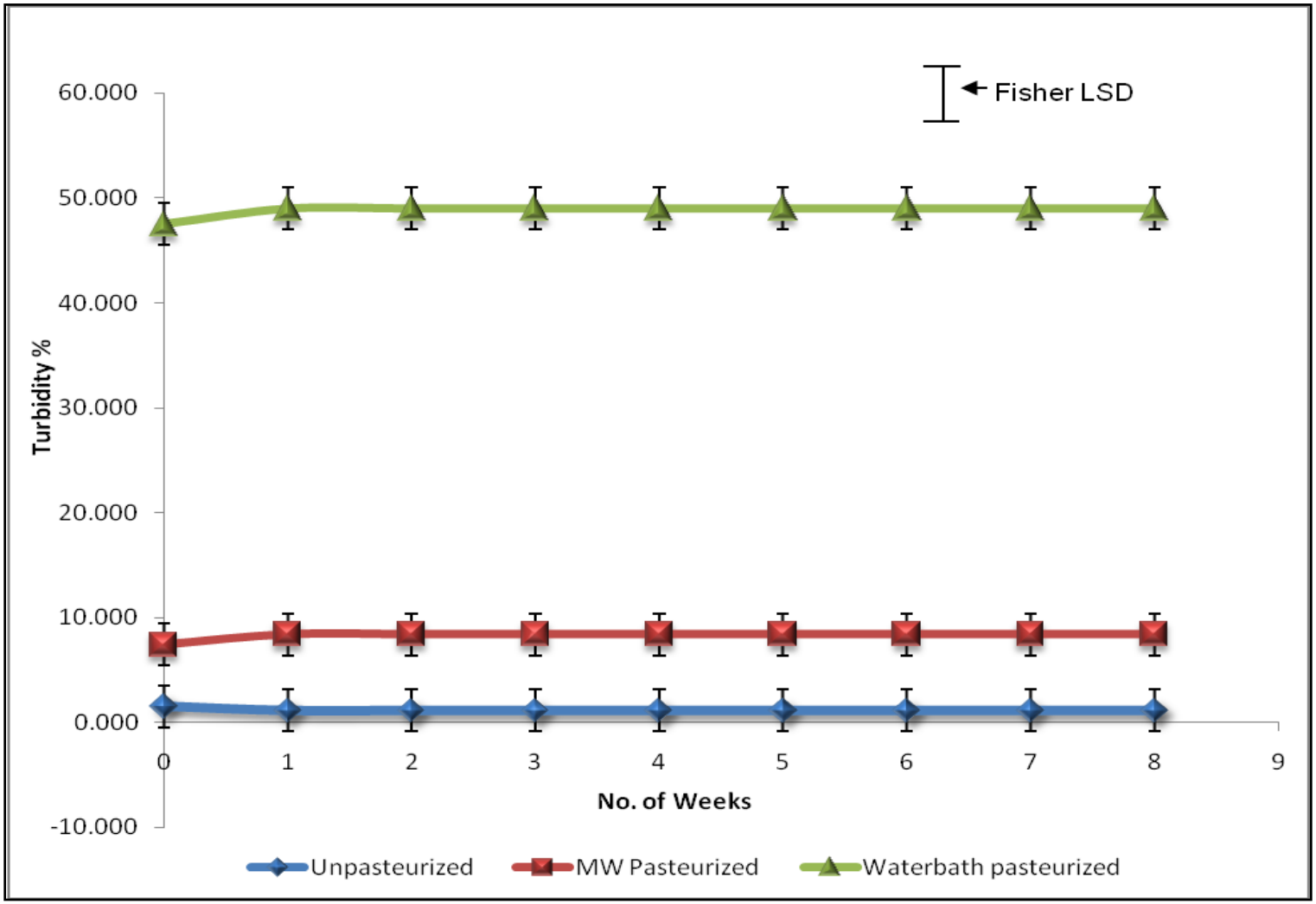

Figure 7.13 Change in Turbidity over time

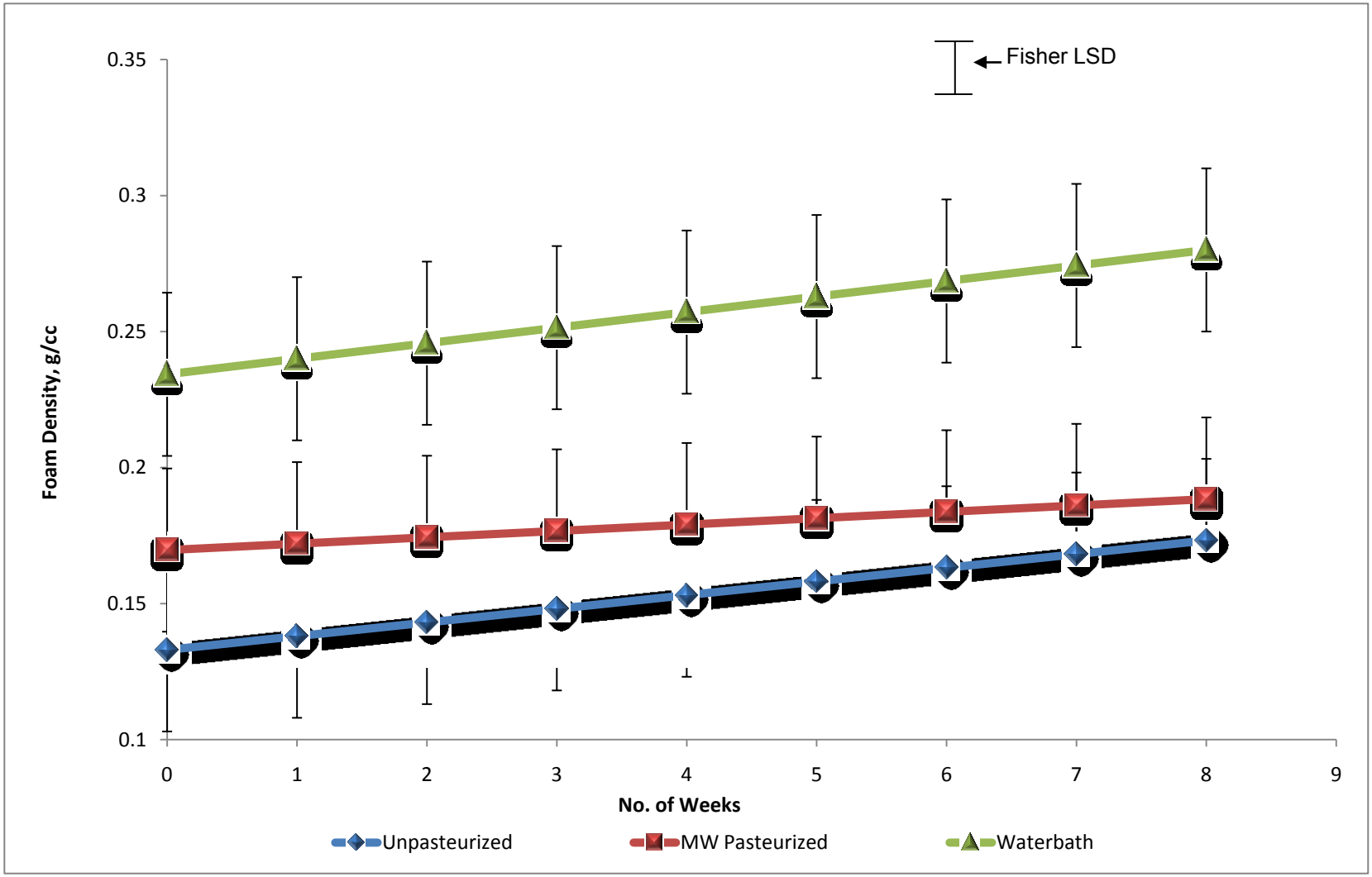

Figure 7.14 Change in foam density over time

7.5 Conclusions

The effects of microwave-heating and hot waterbath-heating of in-shell eggs on the functional properties of the egg white was assessed and compared to that of untreated eggs. It was demonstrated that enthalpy denaturation was much higher for the microwave-heated in-shell egg white similar to that of the untreated egg white and it was also clearer and had a greater viscosity. The microwave-heated egg white produced more stable foam of a lower density than its counterparts. The egg white's dielectric properties gave a good idea of the extent of denaturation in all the three samples.

The tests confirmed that though there was a considerable change in all the above tested parameters in the microwave-heated in-shell egg white, the changes were much less when than those of waterbath-heated eggs, the microwave-heated eggs's properties being more similar to those of the raw (untreated) egg white.

Thus microwaves were proven to be a viable and better alternative for the in-shell heating and pasteurization of shell eggs than conventional hot water methods.

7.6 Acknowledgements

The financial support of Natural Sciences and Engineering Research Council of Canada and Le Fonds Québécois de la Recherche sur la Nature et les Technologies of Quebec is gratefully acknowledged.

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Connecting Text

As we all know in any process, quality control of the end product plays a major role in the marketability and consumer acceptability of the product. Microwave heating is a relatively complex phenomenon. Keeping perfect control of the process parameters for each and every egg at the industrial scale is practically impossible. In order to make sure the final product is of acceptable quality to the consumers, a real-time inline product monitoring and sorting system for removal of any defective/coagulated eggs from mixing with good quality pasteurized eggs in the packaging line, need to be in place. The use of Vis/NIRS has been very well evaluated for the prediction of shell pigmentation, freshness, blood and meat spots and hatching eggs for years. Hence the potential use of hyperspectral imaging in the Vis/NIRS spectral range needs to be investigated.

Chapter 8

HYPERSPECTRAL IMAGING FOR ASSESSMENT OF IN-SHELL PASTEURIZED EGG QUALITY

8.1 Abstract

The potential use of Vis/NIRS in real-time assessment of microwave pasteurized egg quality in terms of variation in transmittance was investigated. Transmittance characteristics in the spectral range of 400 to 1700 nm for microwave-pasteurized eggs treated with three different power densities was compared with that of the waterbath-pasteurized and unpasteurized eggs. Informative wavelengths were identified using subset selection by multiple linear regression analysis. An unsupervised k-means classification was performed to classify the spectral data within a 95% confidence interval. Thus it was established that the presence of any heat damage to proteins inside the shell egg can be quantified in terms of its reduction in transmittance. Also microwave in-shell pasteurized eggs with low power density treatment had transmittance values not significantly ($P<0.05$) different from those of unpasteurized eggs, indicating that microwave pasteurization is ideally suited for shell eggs. A protocol for in-line monitoring of the process quality during microwave in-shell egg pasteurization was developed.

Keywords Hyperspectral Imaging, Unsupervised classification, Protein denaturation.

8.2 Introduction

Spectroscopy has the advantage of being a fast and non-contact, non-invasive method, making it particularly suitable for egg quality assessment. Minimizing contact enhances hygiene. Also large number of eggs can be graded in a short period of time with virtually no sample preparation (De Ketelaere et al., 2004).

Visible/ Near Infrared Spectroscopy (Vis/NIRS) is a rapid, non-invasive and in-line method, increasingly being used for testing the quality of many agricultural products. This technique has been found to be quite effective in assessing the internal quality of fruit and vegetables. Research has been carried out to determine the dry matter in onions (Birth et al., 1985), the quality characteristics of mandarin (Gómez et al., 2006) and the quality of kiwi fruit (Slaughter and Crisosto, 1998).

The use of Vis/NIRS has also been evaluated for the prediction of shell pigmentation, freshness, blood and meat spots, and hatching eggs (Wei and Bitgood, 1989; Narushin et al., 2004; Gielen et al., 1979; Das and Evans, 1992a; Das and Evans, 1992b; Bamelis et al., 2002; Abdel-Nour et al., 2009a).

Studies related to prediction of egg albumen quality using NIRS have obtained varying results. The feasibility of using visible transmission spectroscopy as a non-destructive method to assess the freshness of an egg was investigated by Kemps et al. (2006). The spectral data of 600 white-shelled eggs were compared with the pH and the HU (Haugh unit, a unit for describing egg freshness, based on the thickness of the albumen) and showed that the light transmission spectrum of an egg can provide quantitative information about egg freshness. Kemps et al. (2007) combined visible and near-infrared transmission

spectroscopy with low resolution nuclear magnetic resonance (LR-NMR) and concluded that combining the two spectroscopic techniques did not improve the assessment of egg quality when compared to the use of the transmission spectroscopy alone.

NIR spectral data was used by Schmilovitch et al. (2002) to predict the number of days after laying, the size of the air chamber, weight loss and pH value of eggs with an $R^2 > 0.90$. This high value refers to group means and not to the individual egg. Liu et al. (2007) measured the internal quality of chicken eggs using transmission spectroscopy, finding the egg freshness-relevant transmittance spectral data to be found between 400 and 600 nm.

Vis/NIR spectroscopy is capable of providing detailed chemical, moisture, and other descriptions of constituent parts of an item with the help of vital spectral response information (Casasent and Chen, 2003). A key role in the success of hyperspectral target detection and classification is played by feature extraction, which is the reduction of data dimensionality by extracting features from original spectral space or transformed feature spaces (Cheriyadat and Bruce, 2003).

The choice of the wavelengths is needed to establish a proper protocol for classification. Wavelength selection has many benefits such as the stability of the model to the collinearity in multivariate spectra as well as the interpretability of the relationship between the sample composition and the model. Bangalore et al. (1996) investigated the feasibility of coupling genetic algorithm methods for the selection of wavelengths with partial least squares regression for analysing spectral data. Their study showed that the results obtained after selection were better than those obtained with no spectral range selection. Du et al. (2004)

using the changeable size window partial least squares and searching combination moving window partial least squares found that the combination of these two methods improved the prediction ability of the PLS model. Todeschini et al. (1999) proposed the use of Kohonen artificial neural networks (K-ANN) for selecting a set of wavelengths. Ventura et al. (1998) used a multiple linear regression (MLR) procedure to select the best wavelengths for determination of soluble solids in apple.

A Partial Least Square (PLS) regression model was built by Kemps et al. (2007) in order to link spectral data with the measured albumen pH and HU. They reported that the correlation coefficients between the measured and predicted albumen pH and HU were 0.86 and 0.82, respectively. Furthermore, their study reported that the relevant information concerning egg freshness was in the range of 570 to 750 nm. However, in their studies the spectra ranged from 200 to 1100 nm. These studies demonstrate that the selection of relevant wavelengths is important and can be used as a good strategy to avoid the inclusion of uninformative wavelengths in the predictive model. The selection of informative wavelengths makes the prediction of quality and freshness in shell eggs less complicated.

Therefore in this study, the usefulness of the Vis/NIR transmittance spectroscopy (400-1700 nm) as a non-destructive method for the in-line post-pasteurization sorting of eggs by correlating the change in transmittance to protein damage/ denaturation due to the thermal process was investigated.

8.3 Materials and methods

In-shell eggs were pasteurized using a custom-built laboratory microwave oven setup with a specially-designed slotted waveguide applicator or using a hot water bath maintained at 60°C. Effects of heat treatments on the physical

properties affecting the functional quality of the egg white recovered from the treated eggs were measured and compared to those of fresh untreated egg white.

8.3.1 Egg samples

Fresh whole eggs, within 3 days of grading and packing (identified from the best before date stamped on the eggs, which is usually 35 days following the date of packing) (CEMA, 2004), were procured from a local market and kept in a refrigerator set at 5°C until used. They were all of Canadian Grade A, size large, each of a mean mass of 60±2 g. Prior to pasteurization, the eggs were brought to room temperature of about 24°C by placing the opened carton on the laboratory counter for a period of 3 to 4 hours (tested by breaking and measuring inner temperatures of 3 representative samples) before giving the heat treatments. This is done to replicate the possible use of this technique in the industry, where a significant amount of energy could be saved by following this procedure.

8.3.2 Heat treatments for pasteurization

Two heat treatments for the pasteurization of in-shell eggs were investigated and compared. Three eggs were used for each treatment for the measurement of each parameter. The first three sets of treatments consisted of heating in-shell eggs in a custom-built laboratory microwave oven setup with a specially-designed slotted waveguide applicator operating at 2450 MHz, using power densities of 0.75, 1.5 and 3 W g^{-1}. In-shell eggs were heated for 3, 1.25, or 0.5 minutes corresponding to the above mentioned power densities in order to raise the temperature to 58 ℃ and held at 58±0.5℃ for 2.5 minutes, as per FSIS-USDA (2006) recommendations, by periodically turning the microwave cycles on and off. Temperature measurements were not done during the treatments as an

optimised algorithm developed using a validated finite element method for the microwave pasteurization of eggs was used for the on/off cycles (Dev et al, 2008a and 2009) and the microwave operation was controlled by the computer running HPVEE (Agilent) object-oriented programming language to maintain the desired process temperature. The schematic of this setup is shown in Figure 7.1 in the previous chapter.

The second treatment consisted of immersing the in-shell egg in a temperature-controlled water bath maintained at 60°C for a period of 20 minutes (Schuman et al, 1997). These eggs were left intact without any inserted probes as this was already a commercially practiced technique, approved by FSIS-USDA.

It is clear that the temperatures reached by the egg components using the microwave and water bath heating are not identical. However, the objective of the present study was to compare the properties of egg white pasteurized in-shell by the proposed (microwave) technique with those of eggs subjected to the commercially practiced (water bath) technique, both of which meet the FSIS-USDA pasteurization requirements/recommendations.

Immediately after heat treatments the shell eggs were immersed in cold water (5ºC) for 10 minutes, ensuring that heat damage to the proteins did not continue any longer after the pasteurization.

8.3.3 Hyperspectral Imaging

The hyperspectral imaging system used for the study consisted of 2 line-scan spectrographs namely: ImSpector (ImSpector, V10E, Spectral Imaging Ltd., Finland) with the spectral range of 400 to 1000 nm (Figure 8.1) and HyperspecTM (Headwall Photonics Inc. USA) with a spectral range of 900 to 1700 nm (Figure 8.2).

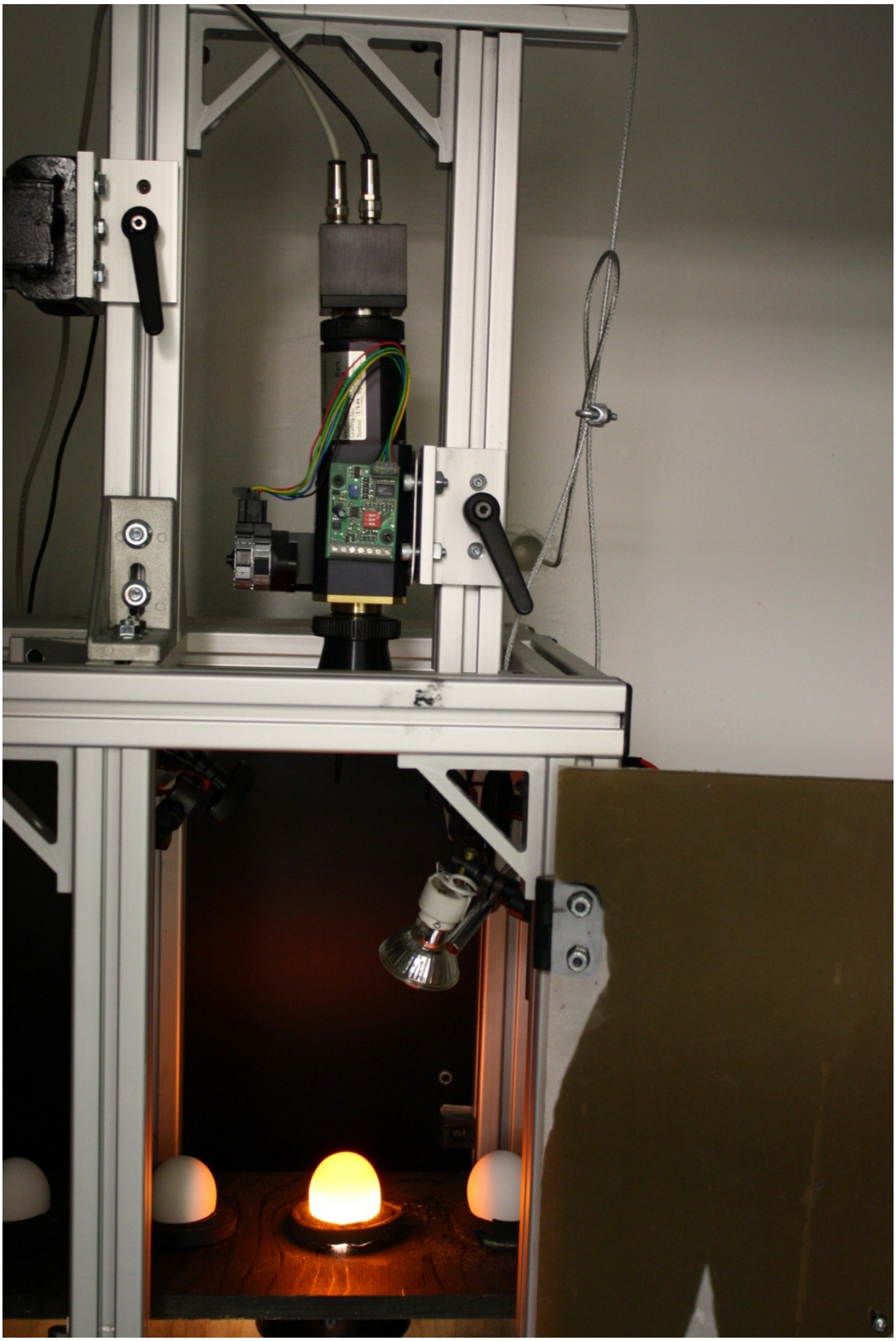

Figure 8.1 ImSpector - 400 to 1000 nm Hyperspectral imaging setup

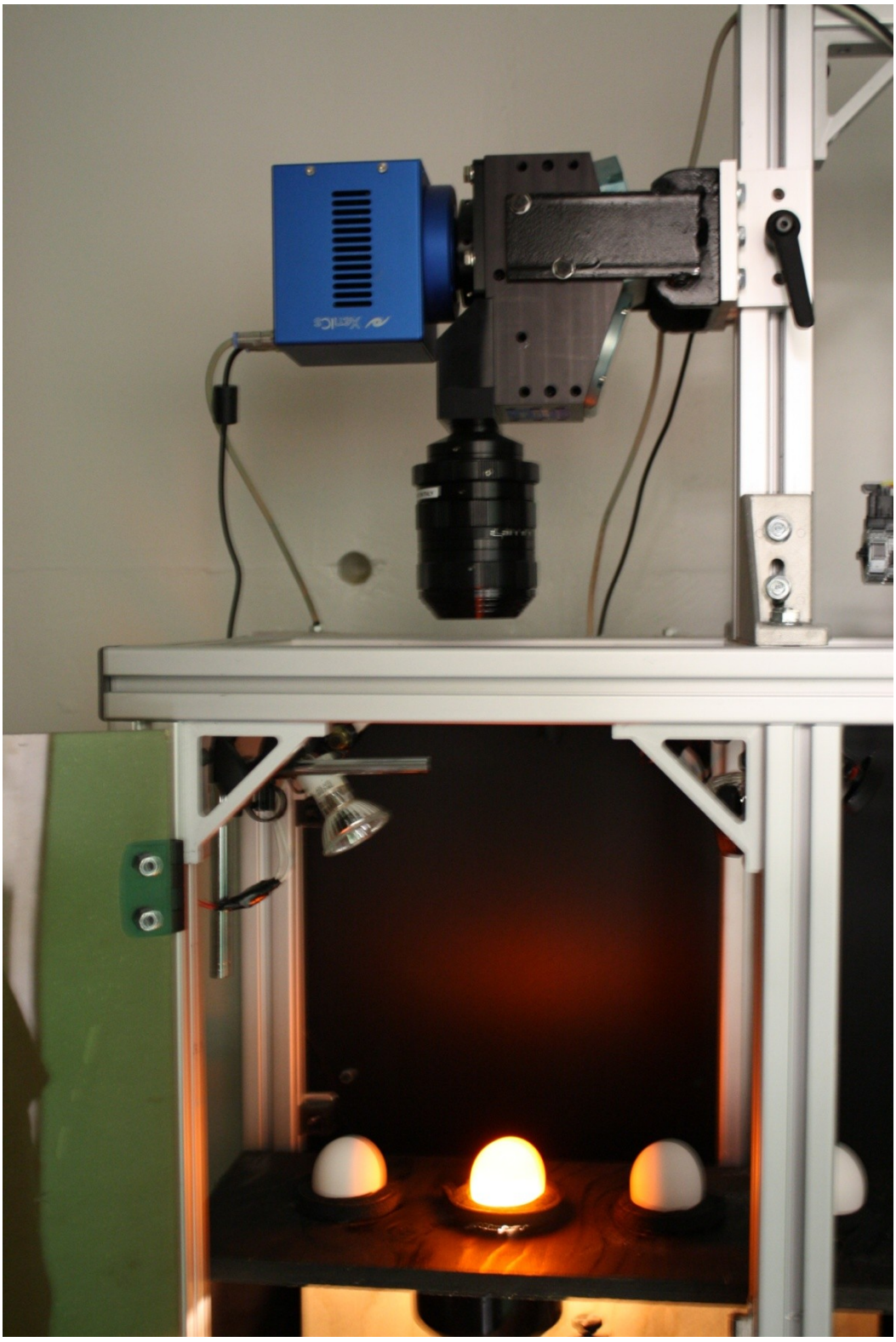

Figure 8.2 HyperspecTM - 900 to 1700 nm Hyperspectral imaging setup

The ImSpector and HyperspecTM were connected to a CMOS camera and InGaAs cameras, respectively, both mounted above a moving conveyor driven by a stepping motor with a user-defined speed (MDIP22314, Intelligent motion system Inc., USA). A tungsten halogen lamp was used to back illuminate the eggs as they are moved across the cameras' field of view.

8.3.4 Data Analysis

MATLAB Version R2010a (Mathworks Inc, USA) was used in merging the hypercubes and multiple linear regression for subset selection. An unsupervised k-means classification of the spectral data was performed using the ENVI version 4.7 software (ITT Visual Information Solutions, CO, USA).

8.4 Results and discussion

The two spectral data hypercubes obtained from the two cameras were merged using MATLAB R2010a and multiple linear regression analysis was done for subset selection (Ventura et al., 1998). From 2151 wavebands scanned, 10 wavelengths (5 from the Visible spectral range and 5 from the NIR range - given in Table 8.1) were chosen as informative wavelengths as they have an $R^2 > 0.90$ in the multiple linear regression analysis for maximum R^2.

The results do not corroborate those of Kemps et al. (2006) who have shown that the relevant information in terms of albumen pH and HU was restricted to the interval between 570 and 750 nm. Kemps et al. (2007) also found that the spectral region between 500 to 900 nm was valuable for the prediction of albumen freshness and egg quality, which is again different from the results obtained in this study. These differences can be due to the method of selection of relevant wavelengths and due to the range of spectral data studied.

Abdel-Nour et al. (2009b) found similar results to this study in choosing the wavelengths for the prediction albumen pH and HU.

Table 8.1 Informative wavelengths for hyperspectral classification of egg quality

S. No	Wavelength (nm)	R^2
1	411	0.96
2	444	0.93
3	484	0.91
4	530	0.96
5	661	0.97
6	936	0.93
7	1196	0.96
8	1345	0.92
9	1402	0.96
10	1719	0.97

Thus the accuracy of prediction can be improved by choosing appropriate wavelengths. This is attributed to the method of selection of relevant wavelengths used for building the predictive model and selection of the equipment.The wavelengths in this study ranged from 400 to 1700 nm which provides better

accuracy, whereas, in the study of Kemp et al. (2006 and 2007), the wavelengths ranged from 200 to 1100 nm.

Figure 8.4 shows an unsupervised k-means classified mosaic made from two eggs from each treatment. It is clear that the waterbath-pasteurized eggs and the high power density (3 W/g) microwave-pasteurized eggs were clearly classified into groups different from that of the unpasteurized eggs and low power density (0.75 and 1.5 W/g) microwave pasteurized eggs.

8.5 Conclusions

In this research, the ability of Vis/NIR spectroscopy to assess egg pasteurization quality in terms of variation in transmittance at 10 informative wavelengths was developed. The results presented above have shown that the transmission spectral data of the egg contains information about egg quality which can be exploited to determine the damage/denaturation of proteins. The protocol developed can be used for non-destructive, real-time in-line monitoring and sorting following in-shell pasteurization of eggs.

8.6 Acknowledgements

The financial support of Naural Sciences and Engineering Research Council of Canada and Le Fonds Québécois de la Recherche sur la Nature et les Technologies of Quebec is gratefully acknowledged.

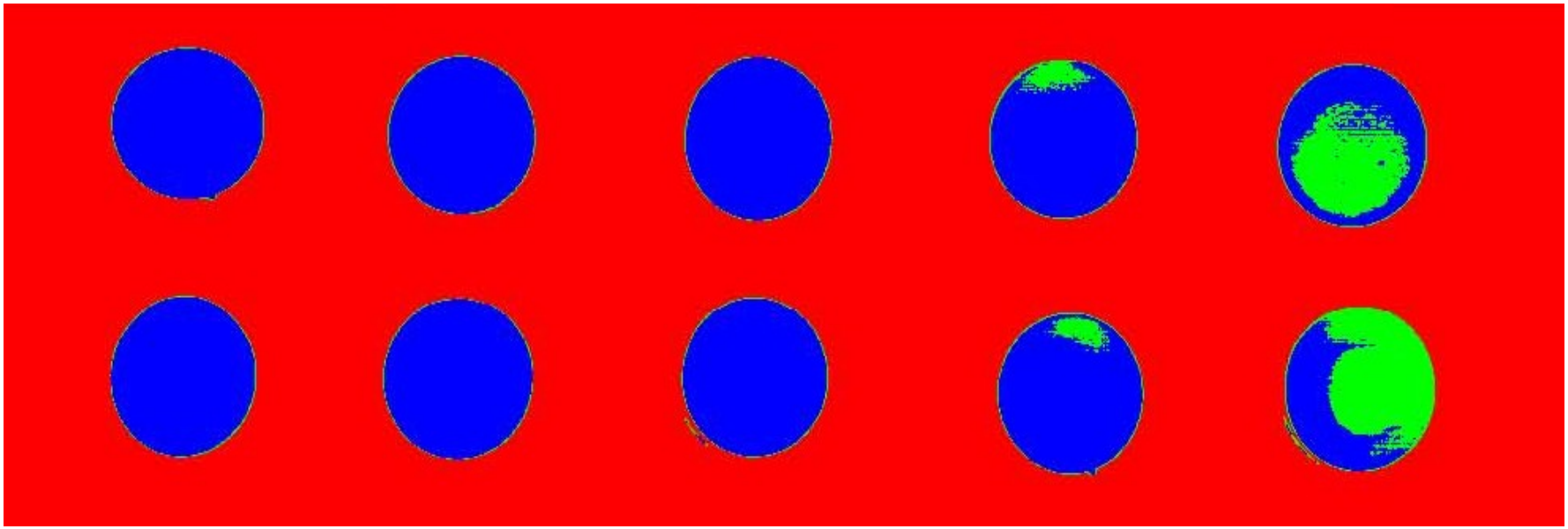

Figure 8.3 Unsupervised k- means classified mosaic made from two eggs from each treatment

From left to right (5 Columns): Column 1 - unpasteurized eggs,

Column 2 - Microwave pasteurized 0.75 W/g,

Column 3 - Microwave pasteurized 1.5 W/g,

Column 4 - Microwave pasteurized 3 W/g,

Column 5 - Waterbath pasteurized

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Chapter 9

GENERAL SUMMARY AND CONCLUSIONS

In a broad-spectrum, this study has shed more light on the least understood aspects of microwave processing. It has provided a deeper insight into the behaviour of a complex heterogeneous food material like in-shell eggs in a microwave environment. Novel simulation techniques were developed and implemented by writing appropriate computer codes. These codes took into consideration various parameters like the mass, geometry, power density and orientation to determine the energy distribution and heating rate of the shell eggs in a multimode cavity. These new approaches along with the code written can be used to simulate the microwave heating of any complex and heterogeneous object. These types of simulations will be very useful in microwave process equipment design and future development of industrial applications.

Finite Difference Time Domain modelling showed that the non-uniformity of heating gets more pronounced at higher power levels, thereby suggesting lower power levels for better quality pasteurized product. A finite element method was applied to approximate the electric field within the biological medium and a closed form expression is presented for the electromagnetic coupling problem, which enables an optimisation procedure to be performed.

A slotted waveguide with an array of unique S-parabolic slots was designed, fabricated and calibrated. A power density of 1.5 W g^{-1} and an angular velocity of $\pi/6$ rad s^{-1} were found to be optimal. The optimal parameters set forth were found to be specifically more efficient in terms of heating time and uniformity. The applicator enhances both penetration and focusing, as well as

providing the necessary temperature gradient from the egg yolk to the shell. Industrial scale up of this is relatively simple but requires further research. The results obtained in this study can readily be used in building a scaled up applicator version for application in the industry.

This study also confirmed that the microwave pasteurization technique for in-shell eggs is very efficient, but requires specifically-designed equipment for efficiently performance, as heating uniformity is always an issue while using microwaves. Tests conducted confirmed that though there was a considerable change in all the above-tested parameters in the microwave heated in-shell egg white, the changes were much less when compared to those caused in the water bath heated eggs. Microwave-heated eggs' whites were much more similar to raw (untreated) egg white than those of hot water-treated eggs.

The ability of Vis/NIR spectroscopy to assess egg pasteurization quality in terms of variation in transmittance was assessed and a protocol for classification of processed eggs using 10 informative wavelengths was developed. The protocol developed can be used for non-destructive, real-time in-line monitoring and sorting of in-shell pasteurization of eggs.

Principally, this study has elucidated the different parameters and the conditions under which in-shell eggs can be successfully pasteurized using microwave energy at 2450MHz without compromising quality. This study had also explored new techniques like hyperspectral imaging and dielectric spectroscopy for the quality assessment of in-shell pasteurized eggs.

Thus the process of microwave pasteurization of shell-eggs is a winning solution to the problem of potentialfood poisoning through raw eggs.

1. The outcomes of this research will improve the safety of in-shell eggs

2. It will help eradicate egg *Salmonellosis* and thereby reduce the direct and indirect economic losses to the poultry industry.

3. The shelf life of the eggs will improve considerably thereby allowing transport to further distances.

4. The breaking stock can be decreased considerably thereby increasing the profits for the Canadian farmers.

9.1 Contribution to knowledge

The outcomes of this comprehensive study on the microwave pasteurization of eggs had contributed to knowledge in many different ways. The following are a few of the several commendable contributions of this research.

1. The novel simulation approaches which were developed and implemented through appropriate codes, can be used to simulate the microwave heating of any complex and heterogeneous object. These types of simulations will be very useful in microwave process equipment design and development.

2. A new method for process optimization using validated simulation techniques was introduced, which can be used to optimize virtual configurations of microwave applicators before fabrication.

3. A break-through in slot design was made in a world that was always thinking straight in terms of the shape of the slot. A slotted waveguide with an array of unique S-parabolic slots was designed, fabricated and calibrated during this research that enhances both penetration and focusing, as well as providing the necessary temperature gradient for pasteurization from the egg yolk to the shell. This can be scaled up for application in the industry.

4. A protocol for classification of processed eggs using Vis/NIR spectroscopy was developed. A novel method exploiting the variation in transmittance of the pasteurized eggs for their real-time classification was developed. Real-time non-destructive in-line monitoring and sorting of in-shell pasteurization of eggs was made possible through this research.

9.2 Recommendations for further research

As any research is incomplete before the public gets the benefit of the research, there are several areas within this field of study that requires further research, in order to be able to apply this commercially.

1. Further research needs to be done in identifying and designing other efficient configurations of microwave waveguides to perform the same in industrial scale as scaling up is a big challenge for a microwave process for which uniformity is the primary factor.

2. Heat treatments affect the nutritional quality of any food material. The effect of the microwave pasteurization process on the nutritional quality of the eggs needs to be investigated.

3. There are existing commercial egg pasteurization techniques like the hot water treatment. The energy efficiency of the process compared to the commercially available techniques needs research.

4. The effect of storage temperature on the pasteurized eggs needs to be studied as storability under non refrigerated conditions will be energy efficient as well eco friendly.

5. Research into the hyperspectral imaging techniques to monitor the storage life of microwave pasteurized eggs will help better energy utilization and will reduce spoilage of eggs.

6. Change in other properties like enthalpy of denaturation and dielectric properties over time need to be investigated in order to be able to associate the effect of storage conditions on the structural integrity of the egg proteins.

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