

Eradication of storage insect pests in maize using microwave energy and the effects of the latter on grain quality

By

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DECLARATION

I declare that the dissertation herewith submitted for the degree MSc Food Science at the University of Pretoria, is my work and has not previously been submitted by me for a degree at any other university or institution of higher education.

Moelo Patience Fakude

November 2007

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ABSTRACT

Eradication of storage insect pests in maize using microwave energy and the effects of the latter on grain quality

By

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To combat insect infestation of maize and maize products during storage without using chemical fumigants, a possible physical treatment method, microwave technology was investigated. Through its selective heating between cereals and insects, microwave technology is a possible physical treatment for eradication of insects and their eggs. Eradication of five insect species, namely *Sitophilus zeamais*, *Rhizopertha dominica*, *Ephestia cautella*, *Cryptolestes ferrugineus* and *Tribolium confusum* was studied. Different microwave parameters such as power dosage, microwave mode, length of microwave cavity, maize exposure method and exposure time were investigated. The effective microwave treatment conditions were then selected, and used to treat maize kernels at laboratory scale. The effect of microwave treatment on the physicochemical properties of maize kernels was investigated.

Microwave single exposures did not result in total insect mortality when maize was dropped through the microwave cavity (free falling) as the exposure times were too short. But utilising a pulley system, total insect mortality was achieved in a single exposure of 9 sec. A long microwave cavity (728 mm) resulted in maize kernel damage in terms of swelling, popping and discolouration. Redesigning the cavity by

shortening it appeared to reduce these effects. The pulsed microwave mode was found to be better than continuous mode. The selected treatment that eradicated all five insect species with no visible kernel damage was pulsed mode at 2450 MHz frequency, using a 483 mm long microwave cavity, at a power level of 1.5 kW, with an exposure time of 9 sec.

The selected conditions (normal treatment) and a more harsh treatment (2 kW power dosage, 18 sec exposure time) were applied to 4 kg samples of white and yellow maize kernels. The normal microwave treatment significantly decreased the moisture content and kernel weight of maize kernels but had no significant effect on test weight, stress cracks, germination and translucency. The harsh microwave treatment also had significant adverse effects on test weight, translucency, germinability, hardness and stress cracks. Additionally, reduced extractability of certain proteins was observed by 2D PAGE with the harsh microwave treatment.

Both normal (power dosage of 1.5 kW, for 9 sec exposure time) and harsh (2 kW, 18 sec exposure time) treatment conditions eradicated adult insects and their eggs, but only the former maintains maize quality. The use of microwave technology has potential to be used as an insect control measure of maize products prior to packaging of the products.

It is recommended that the effect of pulsed microwave disinfection on nutritional quality (starch and protein digestibility of maize products) be studied. Heat transfer phenomena should be studied and improved if possible to reduce the power usage and possibly shorten the exposure time from 9 sec.

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1. INTRODUCTION

Maize is an American Indian word, which means “that which sustains life” (FAO, 1992). According to the Food and Agriculture Organization (2002), maize is the third most important cereal grain worldwide after wheat and rice. In South Africa white maize is used by millions of people as staple food, while yellow maize is the main ingredient in animal feed (South African Department of Agriculture, 2004). In South Africa the average annual commercial production of maize during the past 10 years was 8.2 million tons (4.3 million tons white maize and 3.9 million tons yellow maize). Maize contributes approximately 42% to the gross value of field crops, and the average annual gross value of maize for the past five years up to 2003/04 amounted to R8 919 million (US\$ 131 million) (South African Department of Agriculture, 2004).

During storage of maize, insect infestation can occur. Insect infestation causes maize kernel damage, which results in economic loss (Mason and Storey, 2003). Stored maize is damaged when insects directly feed on the kernels and indirectly contaminate the grain with their waste, cast skins, and body parts. This damage or contamination reduces the quality of the maize kernels.

Currently, insect infestation is controlled by use of chemical pesticides. The disadvantages to the use of chemical pesticides include the insect’s ability to build-up resistance to the pesticides, and the chemical residue that is left on the surface of the product after treatment (Kent and Evers, 1994). The misuse of chemical pesticides, which can lead to resistant insect species, has been observed in many countries (White, 1995). Another problem is that methyl bromide, a commonly used pesticide, is being phased out by 2015, as it has showed to be being an ozone depleting substance (Bell, 2000).

The concerns relating to pesticide residues on food products, insect resistance and health hazards have led to research on the possible use of microwave energy as an alternative insect control measure (Nelson, 1996). Microwaves are part of the electromagnetic spectrum with wavelengths from 1 mm to 1 m, corresponding to frequencies between 300 MHz and 300 GHz (Thostenson and Chou, 1999). Microwaves are nonionizing forms of energy that cause a rise in temperature within

the microwaved medium due to the friction of water molecules when exposed to an electromagnetic field at high frequencies (Lewandowicz et al., 2000).

The use of microwave energy offers the possibility of selectively heating insects over the host grain material due to the difference in dielectric properties between the insects and host material. The success of dielectric heating depends on the moisture content of the host material and it becomes effective when the moisture level of the host material is significantly lower than that of the insects (Nelson, 1996).

2. LITERATURE REVIEW

2.1 THE MAIZE KERNEL

2.1.1 Structure and chemical composition

Maize can be divided into various groups differing in character, namely dent, flint, sweet, floury, popcorn and waxy (Jugenheimer, 1976). In South Africa the common type of maize produced is dent maize. According to Hosney (1994), dent maize is the largest type, with a flattened kernel weighing an average of 350 mg. In colour, maize kernels can range from white to brown/ purple, but white and yellow are the most common colours.

The maize kernel is composed of four major parts: pericarp, germ, endosperm and tip cap (Figure 2.1). According to Inglett (1970) the endosperm comprises about 82% of kernel's dry weight, the germ 12%, pericarp 5%, and tip cap 1%.

2.1.1.1 Pericarp

The pericarp also known as the "hull or the bran" is the outermost layer of the kernel (Watson, 2003). It is made up of elongated cells forming a tough, dense tissue (Inglett, 1970), and in thickness it ranges from 66 to 160 μm (Hosney and Faubion, 1992). The pericarp is divided into 5 different layers. From the outside they are the epidermis, mesocarp, cross cells, tube cells and lastly the seed coat. The inner layer of the pericarp adheres tightly to the outer surface of the aleurone layer (Watson, 2003).

The epidermis has a waxy cuticle with a thickness of 0.7 to 1.0 μm . The epidermis extends around the whole kernel except the tip cap (Eckhoff, 1995). This layer is known to restrict the entry of water to the kernel. The mesocarp is the thickest layer and amounts to 90% of the pericarp (Watson, 2003). The cells of the mesocarp are approximately 1000 μm in length and 7-10 μm in diameter. The tube cell layer is the innermost layer compressed against the seed coat membrane. The cells are thin walled, unbranched and 135-250 μm long. The seed coat layer is attached to the

outermost aleurone layer and it is considered to have semi-permeable properties by allowing selected material to enter the kernel (Watson, 2003)

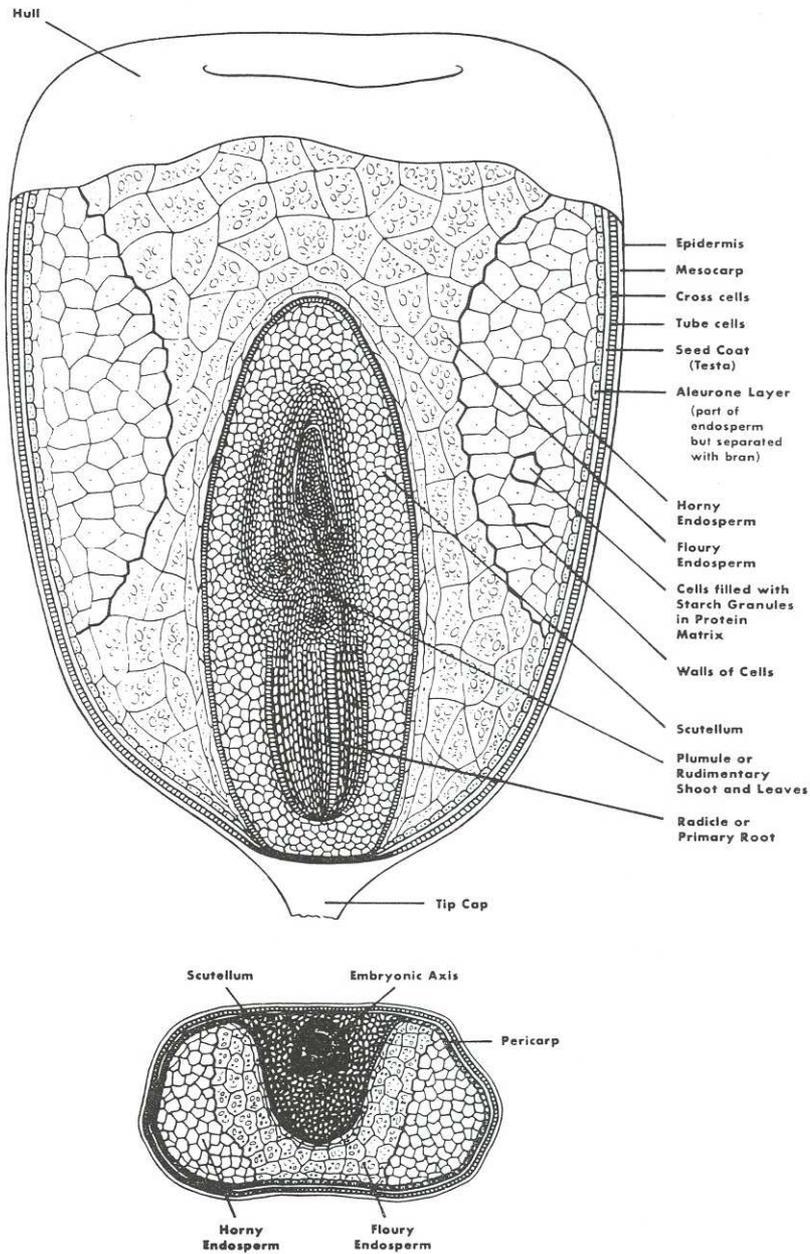


Figure 2.1: Longitudinal and cross sections of a maize kernel (Hoseney, 1994).

2.1.1.2 Germ

The germ is composed of the embryo and the scutellum. It (germ) stores nutrients and hormones, which are utilised in the stages of germination. The germ is the major source of lipids, accounting for 83% of the total kernel lipids (Watson, 2003). The lipids are normally extracted to give maize oil (Watson, 2003).

2.1.1.3 Endosperm

According to Watson (2003), the endosperm constitutes 82-84% of the kernel dry weight. The endosperm is surrounded by a single thick walled layer, the aleurone layer, which also covers the germ (Hoseney and Faubion, 1992). This layer contains protein and lipid bodies. The major constituent of the endosperm is starch, which amounts to 86-89% of endosperm by weight (Watson, 2003). The starch is stored in starch granules that are 3-25 μm in diameter.

The maize endosperm has vitreous (corneous or horny) and opaque (floury) regions in the same kernel (Watson, 2003). Kent and Evers (1994) state that vitreous kernels are translucent and appear bright against strong light, whereas floury kernels are opaque and appear dark. Light can pass through vitreous endosperm because the starch granules are tightly held together by a thick protein matrix, resulting in a dense endosperm with few or no air spaces (Hoseney, 1994). Even though the starch granules in the floury endosperm are also surrounded by a protein matrix, the matrix is thinner as compared to vitreous endosperm (Paulsen et al., 2003). The presence of small air-filled fissures between floury endosperm cells form reflecting surfaces, thereby preventing light transmission (Kent and Evers, 1994).

The protein content in maize ranges from 6 to 18%, and the average protein content of the endosperm is 8-9% (Lawton and Wilson, 2003). According to Shukla and Cheryan (2001), Osborne (1924) classified plant proteins according to their solubility. Albumins are soluble in water, globulins soluble in saline solution, glutelins soluble in dilute alkali and prolamins are soluble in alcohol. The albumins and globulins are the physiologically active proteins which are concentrated in the aleurone layer, pericarp

and the germ (Lawton and Wilson, 2003). The prolamins and the glutelins are storage proteins, and they are located in the endosperm.

2.2 INSECT PESTS

Insects infest stored products worldwide. Most of the insects associated with stored grain in South Africa are species that were imported into the country by grains or other food products (Harney, 1993). The spread of insects around the world is supported by the transportation of grains from grain-surplus countries to those who are grain-short. It is estimated that 900 Mt of cereal grains are stored around the world at one time (White, 1995). Beetles are the most common type of insects that attack stored grain followed by moths (Mason and Storey, 2003). There are nearly 40 families of beetles (Smit, 1964).

Insects are animals with external skeletons (or exoskeletons), divided into segments or rings (Smit, 1964). The segments are grouped into three parts namely the head, thorax (middle part), and the abdomen (hind part). The thorax has of three pairs of legs and two pairs of wings.

Insect species that are known to infest stored maize and maize products that will be discussed in this literature review, are:

Sitophilus zeamais (Motschulsky) – maize weevil

Rhizopertha dominica (Fabricius) – lesser grain borer

Cryptolestes furrugineus (Stephens) – rusty grain beetle

Tribolium confusum (Jacquelin du Val) – confused flour beetle

Ephestia cautella (Walker) – tropical warehouse moth.

Insects that infest stored grain are divided into two groups: namely primary and secondary insects. The former can bore the kernel and develop inside it, thereby hiding their infestations in the grain mass (Cotton and Wilbur, 1982). The latter do not have the kernel boring capability and can only develop outside the kernels by feeding on broken kernels, the germ, on grain dust, and flour. Primary insects include species

such as *S. zeamais* and *R. dominica*, while secondary insect species include *C. furrugineus* and *T. confusum* (Table 2.1).

2.2.1 *Sitophilus zeamais* – maize weevil

The maize weevil is in the Curculionidae family, which includes *S. granarius* (granary weevil) and *Sitophilus oryzae* (rice weevil) (Harney, 1993). According to Smit (1964) this family is the largest family in the animal kingdom containing nearly 40, 000 species. A maize weevil is about 3 to 4.8 mm in length with reddish to brown to black colour (Harney, 1993). The thorax is covered with small round pits and the wings are covered with four light red to yellow spots, two on each side of the body (Figure 2.2) (Smit, 1964). These four spots distinguish the maize weevil adults from rice and granary weevils. With its developed wings, it can easily fly from storerooms and silos to infest grain in the field. It has antennae which are segmented with a compact, 2 segmented clubs (Harney, 1993).

The maize weevil is classified as a primary pest as it can bore the grain kernel to feed as well as lay its eggs inside the kernel. Most insects have four development stages: namely eggs, larva, pupa and adults (Mason and Storey, 2003). The female weevil can roughly lay 350 eggs inside the grain kernel (Smit, 1964). Once the eggs hatch, the resulting larvae feed on the nutritious endosperm until they fully develop. The fully-grown larvae pupate inside the grain. Roughly after a week from pupating the adult weevil emerges out of the kernel. The life cycle of this species takes around 30 days from eggs to adult (Harney, 1993).

Table 2.1: Primary and secondary insect pests of stored maize (Mason and Storey, 2003)

Scientific Name	Common Name	Family Name
Primary pests		
<i>Sitophilus oryzae</i> (L.)	Rice weevil	Curculionidae
<i>S. zeamais</i> (Motschulsky)	Maize weevil	Curculionidae
<i>S. granarais</i> (L.)	Granary weevil	Curculionidae
<i>Rhizopertha dominica</i> (F.)	Lesser grain borer	Bostrichidae
<i>Prostephanus truncates</i> (Horn)	Larger grain borer	Bostrichidae
<i>Araecerus fasciculatus</i> (DeGeer)	Coffee bean weevil	Anthribidae
<i>Sitotroga cerealella</i> (Olivier)	Angoumois grain moth	Gelechiidae
Secondary pests		
<i>Cryptolestes ferrugineus</i> (Stephens)	Rusty grain beetle	Laemophoeidae
<i>C. pusillus</i> (Schönherr)	Flat grain beetle	Laemophoeidae
<i>Oryzaephilus surinamensis</i> (L.)	Sawtoothed grain beetle	Silvanidae
<i>Oryzaephilus mercator</i> (Fauvel)	Merchant grain beetle	Silvanidae
<i>Cathartus quadricollis</i> (Guerin-Méneville)	Squarenecked grain beetle	Silvanidae Silvanidae
<i>Plodia interpunctella</i> (Hübner)	Indianmeal moth	Pyrilidae
<i>Ephestia kuehniella</i> (Zeller)	Mediterranean flour moth	Pyrilidae
<i>Pyrallis farinalis</i> (L.)	Meal moth	Pyrilidae
<i>Cadra cautella</i> (Walker)	Almond moth	Pyrilidae
<i>Nemapogon granella</i> (L.)	European grain moth	Tineidae
<i>Tenebroides mauritanicus</i> (L.)	Cadelle	Trogossitidae
<i>Trogoderma variabile</i> (Dermestidae)	Ballion Warehouse beetle	Dermestidae
<i>Trogoderma granarium</i> Everts (Dermestidae)	Everts Khapra beetle	Dermestidae
<i>Attagenus unicolor</i> (Brahm)	Black carpet beetle	Dermestidae
<i>Lasioderma serricornis</i> (F.)	Cigarette beetle	Anobiidae
<i>Stegobium paniceum</i> (L.)	Drugstore beetle	Anobiidae
<i>Latheticus oryzae</i> (Tenebrionidae)	Waterhouse	Tenebrionidae
	Longheaded flour beetle	
<i>Gnatocerus cornutus</i> (F.)	Broadhorned flour beetle	Tenebrionidae
<i>Gnatocerus maxillosus</i> (F.)	Slenderhorned flour beetle	Tenebrionidae
<i>Tribolium castaneum</i> (Herbst)	Red flour beetle	Tenebrionidae
<i>T. confusum</i> Jacquelin du Val	Confused flour beetle	Tenebrionidae
<i>Palorus subdepressus</i> (Wollaston)	Depressed flour beetle	Tenebrionidae
<i>P. ratzeburgi</i> (Wissman)	Smalleyed flour beetle	Tenebrionidae
<i>Alphitobius diaperinus</i> (Panzer)	Lesser meal worm	Tenebrionidae
<i>Tenebrio molitor</i> (L.)	Yellow meal worm	Tenebrionidae
<i>Cynaues angustus</i> (Le Conte)	Larger black flour beetle	Tenebrionidae
<i>Carttodere constricta</i> (Gyllenhal)	Plaster beetle	Lathridiidae
<i>Corticaria pubescens</i> (Gyllenhal)	A minute brown scavenger beetle ^a	Lathridiidae
<i>Pseudeurostus hilleri</i> (Reitter)	A spider beetle ^a	Ptinidae
<i>Ptinus fur</i> (L.)	White marked spider beetle	Ptinidae
<i>Ptinus</i> spp	Spider beetles	Ptinidae

^a Common name not approved by the Entomological Society of America



Figure 2.2: The physical appearance of *Sitophilus zeamais* (Van Tonder and Prinsloo, 2000).

2.2.2 *Rhizoperta dominica* – lesser grain borer

The lesser grain borer is a member of the Bostrichidae family (Harney, 1993). It is 2-3 mm in length and shiny reddish-brown to black in colour. Its body is divided into two parts (Figure 2.3). The first part is a segment that consists of the head and bears the anterior legs but no wings. This segment also consists of rows of rounded teeth (Harney, 1993). Close to the head there are a number of projections (Cotton and Wilbur, 1982). The antennae have 10 segments with a large loosely attached 3 segmented club. The legs are short with distinct teeth on the outer margins (Harney, 1993).

The lesser grain borer is classified as a primary pest. It is known to attack a variety of plant materials including nuts, dried fruits and stored grain (Harney, 1993). A female can lay up to 500 eggs (Cotton and Wilbur, 1982). The eggs are deposited outside the kernels mostly on the loose grain, and the larvae hatch in a few days (Harney, 1993). The larvae feed on the flour produced by the boring actions of adults or directly boring into kernels. The larvae complete their growth phase inside the kernel before transformation into pupae. Once the insect escapes the cuticle of the pupa, it leaves the grain kernel. Oviposition differs from that of the maize weevil in that it is done outside the kernel. The developmental life cycle lasts around 30 days at the optimum temperature of 34°C.

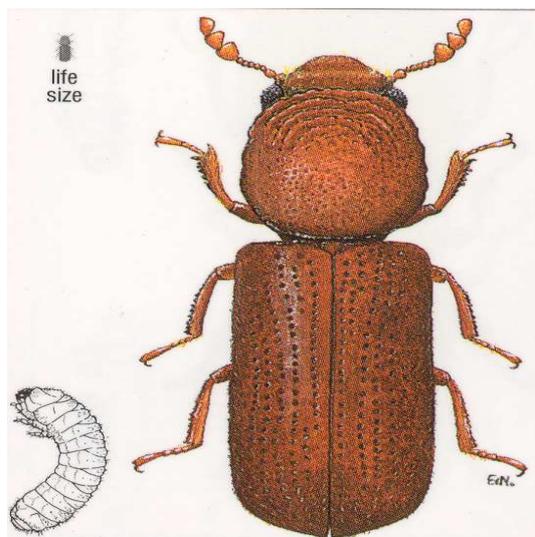


Figure 2.3: The physical structure of *Rhizopertha dominica* (Van Tonder and Prinsloo, 2000).

2.2.3 *Cryptolestes ferrugineus* – rusty grain beetle

The rust-red grain beetle is classified under the Laemophoeidae family (Harney, 1993). Three species of the same genus *Cryptolestes*, namely the flat grain, rust red grain and the flour mill beetles are all known as flat grain beetles because of their similarities in appearance and behaviour (Cotton and Wilbur, 1982).

The rust-red grain beetle is a secondary pest. Eggs are laid on broken kernels (Harney, 1993). The larvae are known to have preference for the wheat germ, where they feed themselves until they pupate. The larvae can also feed on dead insects. The life cycle of the rust red grain beetle is between 5-9 weeks depending on temperature.

According to Harney (1993) the rust red grain beetle is 1.8-2.2 mm in length and yellowish to reddish brown in colour. The head has sub-lateral lines forming slightly raised ridges (Figure 2.4, for visual reference). The antennae are half the length of the whole body and this is observed in both female and male sexes. They have 11 segments with the last three segments a little bigger than the rest. The wings are marked with parallel, fine and longitudinal lines, with four rows of equally spaced hair

like projections. The tarsal formula, which is the set up of the legs, fore-, mid-, and hind is 5-5-5 in females and 5-5-4 in males.

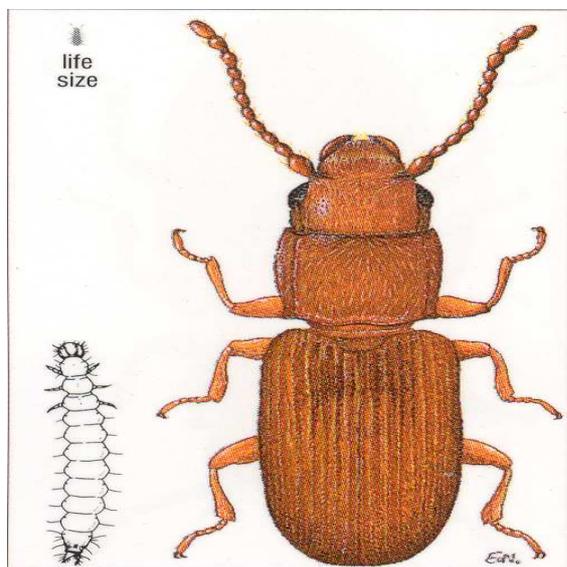


Figure 2.4: The physical appearance of *Cryptolestes ferrugineus* (Van Tonder and Prinsloo, 2000).

2.2.4 *Tribolium confusum* – confused flour beetle

Confused flour beetles are the members of the Tenebrionidae family (Harney, 1993). Cotton and Wilbur (1982) state that this pest can survive under bark, feeding off dead and living plants and animals. But now they are known to feed mostly on flour, dried fruits, nuts and other stored products.

The beetle is 2.6 to 4.4 mm in length and reddish to brown in colour (Harney, 1993). The head has of compound eyes on each side and is divided by a lobe (Smit, 1964). The antennae are divided into 11 segments, with the last three larger than the rest of the segments (Figure 2.5). The wings are marked with parallel, longitudinal, fine lines (Harney, 1993).

According to Smit (1964) and Cotton and Wilbur (1982) the optimum temperature for the life cycle development is 30 to 35°C. Under favourable conditions, a female can deposit 400 to 500 eggs at the rate of 6 to 12 eggs daily over a period of several months. The eggs are normally covered with a sticky substance that helps the eggs

to adhere to the sides of sacks. These eggs get covered by flour and this makes it difficult for millers to detect.

Under favourable conditions, eggs hatch within 7 days, but temperatures below 15°C and above 40°C do not favour hatching (Harney, 1993). Larvae develop for about a month to become pupae and two weeks afterwards adults emerge from the pupa cases. The life cycle from egg to adult is 6 weeks. Flour infested with large population of the species turns pink due to contamination by the insects' secretions (Cotton and Wilbur, 1982; Harney, 1993). The flour also acquires a sour, strong smell and during baking the dough does not rise as normal.

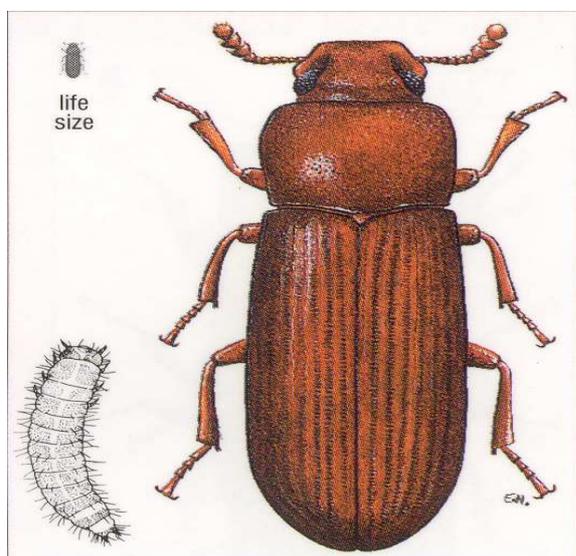


Figure 2.5: The physical structure of *Tribolium confusum* (Van Tonder and Prinsloo, 2000).

2.2.5 *Ephestia cautella* – tropical warehouse moth

This moth has a wingspan that is 6 to 23 mm long and has a simple eye, occurring in small groups (Harney, 1993). The colour of the forewings varies from yellow to grey, while the hind wings are greyish to white. Figure 2.6 illustrates the physical structure of the moth and larvae.

The moth lays its eggs on food material and the larvae emerge after 3 to 4 days. Under the right conditions the fully-grown larvae will then pupate. The complete life cycle is 8 to 9 weeks.

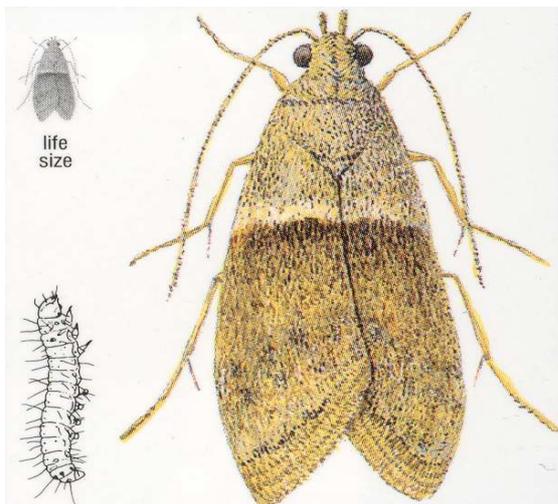


Figure 2.6: The physical appearance of *Ephestia cautella* (Van Tonder and Prinsloo, 2000).

2.3 ENVIRONMENTAL FACTORS THAT AFFECT THE DEVELOPMENT OF INSECTS

Most insects, including beetles and moths, go through four development stages, namely egg, larva, pupa and adult (Mason and Storey, 2003). The eggs exist for a few days up to several weeks under favourable conditions before they hatch into larvae. The larval stage is considered an important development stage. In terms of growth, an increase in weight is observed during this phase. When the larva consumes food, it consumes more than its own weight, therefore expanding in size. As it expands, it sheds off its skin to allow new growth in a process called moulting. Once the larva develops into a pupa, transformation into an adult follows shortly.

In order for insects to multiply, conditions need to be favourable. The most two important environmental factors that influence the growth of insects are temperature and moisture (Mason and Storey, 2003).

2.3.1 Temperature

There are three temperature zones for all insects: the optimum, the zone at which the highest rate of development can be achieved; the sub-optimum, a zone below or above the optimum zone in which insects can still complete their life cycles; and thirdly the lethal zone, temperatures above or below sub-optimum zone where insects are killed over time (Table 2.2) (Fields, 1992).

According to White (1995), insects in stored grain can survive at temperatures between 8 and 41°C. The optimum temperatures for growth and reproduction are between 13 and 25°C, and 33 and 35°C, respectively (Fields, 1992). The maximum temperature an insect species can survive is less than 5°C above its optimum temperature (Howe, 1965). The relative humidity needs to be between 50 and 70% (White, 1995). Temperatures below 13°C and above 35°C generally become unfavourable for insects to reproduce (Fields, 1992; Mason and Storey, 2003).

2.3.2 Moisture

Storage insects obtain their water for survival from grain. Mason and Storey (2003) observed that high insect populations were present in maize with a moisture content of only 10 to 12%. Some insect species prefer higher moisture conditions, above 13% (Storey et al., 1983). Increasing the grain moisture content favours insect development, but moisture content above 15% also favours the development of bacteria and fungi (Mason and Storey, 2003).

Table 2.2: The response of stored product insects to temperature* (Fields, 1992)

Temperature (°C)	Zone	Effect
50 to 60	Lethal	Death in minutes
45		Death in hours
35	Sub-optimum	Development stops
33 to 35		Development slows
25 to 33	Optimum	Maximum rate of development
13 to 25	Sub-optimum	Development slows
13 to 20		Development stops
5	Lethal	Death in days (unacclimated), movement stops
-10 to -5		Death in weeks to months
-25 to -15		Death in minutes, insects freeze

* Species, development stage and moisture content of food will influence the response to temperature

When the grain moisture content is low, insects obtain water by breaking down the grain or utilize their energy reserves in their fatty tissues (Cotton and Wilbur, 1982; Mason and Storey, 2003). Maize, rice and granary weevil adult population cannot grow inside the grain at moisture contents less than 9% (Cotton and Wilbur, 1982). Low moisture content results in the death of the insects (inside grain).

2.4 METHODS USED FOR INSECT CONTROL

Grain should be stored under conditions that are not favourable for insect infestation. Sound measures should be implemented to protect grain from insects. Physical methods and chemical pesticides have been widely used for control of pests. Physical methods include manipulation of temperature, atmospheric composition, desiccation using inert dusts, microwave energy and ionizing irradiation (Banks and Fields, 1995). These methods can be applied separately or in combination. According to Harein and Davis (1992) chemical pesticides are the most effective insect management tool. Non-residual pesticides include methyl bromide, phosphine, hydrogen cyanide and ozone.

2.4.1 Physical methods

2.4.1.1 Low temperature

Application of low temperatures to control insect infestation has two basic effects. It reduces the insect development rate and feeding, and it decreases their survival rate (Banks and Fields, 1995). These authors indicate how important it is that the temperature at which the insect population does not increase anymore defines the target temperature at which the grain mass needs to be maintained to prevent further infestation increase.

Stored grain insects have a broad range of tolerances to cold. According to Banks and Fields (1995) *T. castaneum*, *T. confusum* and *Oryzaephilus Mercator* (Fauvel) are some of the most cold susceptible insect species. Whereas *S. granaries*, *E. elutella*, *E. kuehniella*, *Trogoderma granarium* (Everts) and *Plodia interpunctella* (Hübner) are the most cold tolerant insect species. Most insects that infest stored products become inactive at temperatures below 10°C (Maier et al., 1996). Eggs are the most cold susceptible development stage (Fields, 1992).

Acclimation of insects to low temperatures is one of the important factors responsible for insect cold survival. By pre-exposing insects to temperatures of 20 to 10°C increases their survival at lower temperatures by 2 to 10 times (Fields, 1992). For example David et al. (1977) studied the effect of low temperature on *S. oryzae*, *R. dominica* and *S. granarius* acclimation at 21°C before placing them at 4.4°C. The author concluded that acclimation increased the survival chances of the insects.

2.4.1.2 High temperature

The same principle that applies to cold temperatures also applies for hot temperatures. The survival of insects depends on species, stage of development, duration of exposure, temperature, acclimation and relative humidity (Fields, 1992). Research on the application of high temperature on insects has been done using microwaves, fluidised-bed heating (air), high temperature/short time (HTST), high frequency waves and infrared.

A 5°C increase in temperature above the optimum temperature, stops the growth of an insect population (Banks and Fields, 1995; Fields, 1992). Mourier and Poulsen (2000) studied the effect of HTST techniques on wheat and maize infested with mites and insects. Wheat was infested with grain mites (Acari) and *S. granarius* (granary weevils), and maize was infested with *Prostephanus truncatus* (Horn) (larger grain borer). Grains were exposed to hot air in a microline toaster with a drum. More than 99% mortality was obtained for all stages of *S. granarius* and grain mites with an inlet temperature of 300°C to 350°C/ 40 sec exposure time. For the control of *P. truncates* in maize, an inlet temperature of 700°C to 750°C with an exposure time of 19 sec resulted in 100% mortality. The authors state that the HTST technique did not have any effect on the quality of grain except that germination was reduced.

Mechanisms suggested for the causes of death at high temperatures include changes in lipids, imbalances in the rate of biochemical reactions, perturbation of ionic activities, and desiccation (Fields, 1992). The phospholipid membranes become more fluid at high temperatures. Since the nervous system is dependent on membrane integrity, at elevated temperatures the nervous system loses its functionality. The structure of proteins is also affected at high temperatures. All the enzymes that are involved in the survival of the insects will be greatly affected, as they will start to denature (Fields, 1992). One disadvantage of high temperature treatment of maize is that it can negatively affect maize quality (Shivhare, 1992).

2.4.1.2.1 Microwaves

Electromagnetic energy is used for different applications in the food industry, such as sterilisation of food products (Banik et al., 2003); grain drying (Gunasekaran, 1990); physicochemical modification of starch (Lewandowicz et al., 2000; Szepes et al., 2005) and control of insect pests in agricultural products (Nelson and Kantrack, 1966; Nelson and Stetson, 1974; Nelson, 1985). Microwaves have a frequency band from 300-300 000 MHz in the electromagnetic spectrum (Ohlsson and Bengtsson, 2001).

Frequencies of 915 and 2450 MHz are used for industrial, commercial and domestic applications (Lambert, 1980). In a microwave oven, the waves are generated by a

device known as a magnetron and the microwaves are channelled via the waveguide to the oven cavity where the treated material is being held (Ohlsson and Bengtsson, 2001). Materials can be classified into three categories based on their interaction with microwaves, namely absorbing, reflecting and transparent (Ohlsson and Bengtsson, 2001). Absorbing materials consist of polar constituents e.g. water. In reflecting materials e.g. metals, only small amounts of microwaves can penetrate the materials and the rest of the microwaves are reflected. Reflecting materials are normally used to make waveguides for ovens. Transparent materials do not absorb or reflect microwave energy, e.g. glass, thus can be used as microwaving containers.

The difference between conventional and microwave heating is that in the former heat is applied on the outside of the food and is transferred to the middle by means of conduction (Harrison, 1980). Thus, it takes a long time for the centre of the food to reach the target temperature. In a microwave oven, heating is achieved mainly due to the movement of dipole water molecules at high speed as they absorb microwaves (Ohlsson and Bengtsson, 2001). As the water molecules continuously re-orientate themselves in the electromagnetic field, heat is generated throughout the food material, although penetration depth is dependent on the frequency of microwave used.

Microwave heating can be described in terms of the dielectric properties of food materials. Dielectric properties together with thermal and physical properties of food determine the absorption of microwave energy and heating behaviour of microwaved treated food materials (Dibben et al., 2001). The two most important factors are the dielectric constant (ϵ') and the dielectric loss factor (ϵ'') (Ohlsson and Bengtsson, 2001). These factors show the ability of materials to store energy and disperse energy, respectively. Factors that influence dielectric properties of materials include temperature, moisture content, density and structure of the material, molecular composition and the frequency of the applied electric field (Dibben et al., 2001).

According to Nelson (1972) a study on the use of microwave energy as a possible insect control measure was carried out in the late 1920's. Nelson, (1972) indicated that the use of microwave technology to control insect infestation has potential, but

the only obstacle is the capital costs required. According to Wang et al. (2003) the advantages of using microwave treatment include no chemical residues on products after treatment, and minimal impact on the environment.

During microwave heating of infested material, selective heating based on the relative moisture contents of the insects and food material can be achieved (Nelson, 1972). Due to the higher moisture content of insects, they will absorb microwave energy at a higher rate as compared to the food material therefore reaching a lethal temperature without damaging the food material. Rashkovan et al. (2003) measured the dielectric properties of wheat and granary weevils in the frequency band of 2 to 150 MHz. They found that the relative dielectric constant and loss factor of insects were greater than those of the host grain (Table 2.3). Nelson et al. (1997) presented data that confirmed that the dielectric factors of insects are higher than those of cereal grains. The moisture content of rice weevils was 49% as compared to the 10-12% moisture content of stored grains.

Table 2.3: The dielectric constant (ϵ') and the dielectric loss factor (ϵ'') of weevils and grain in relation to frequency (Rashkovan et al., 2003)

Frequency MHz	ϵ' Weevil	ϵ' Wheat	ϵ'' Weevil	ϵ'' Wheat
20	3.60	2.00	0.65	0.08
40	3.75	2.05	0.70	0.08
60	3.90	2.05	0.80	0.10
80	4.00	2.15	0.90	0.11
100	4.10	2.20	0.95	0.11
130	4.18	2.25	1.05	0.13
150	4.75	2.30	1.28	0.14

Halverson et al. (1996) studied the mortality of *S. zeamais* and *T. castaneum* in wheat when microwaved at a frequency of 10600 MHz at a power level of 9-20 kW. Mortalities of $\geq 93\%$ and $\geq 94\%$ were obtained for *S. zeamais* and *T. castaneum*, respectively. Nelson and Kantack (1966) obtained complete mortality of *S. granaries* when exposed to Radio frequency (RF) heating at a frequency of 39 MHz, and

temperature was raised to 41°C. Baker et al. (1956) obtained 100% mortality of *S. granaries* when treated at a frequency of 2450 MHz. Kirkpatrick and Roberts (1971) studied the mortality of *R. dominica*, *S. cerealella* and *S. oryzae* in wheat when microwave treated at a frequency of 2450 MHz at 2 kW. Total mortalities were obtained for *S. cerealella* and *S. oryzae* at an exposure time of 15 sec. Total mortality for *R. dominica* was only achieved at 25 sec exposure time. Hamid and Boulanger (1970) studied the mortality of *T. confusum* in wheat when microwave treated at 1.2 kW (frequency 2450 MHz). Total mortality of *T. confusum* was obtained when the temperature of wheat reached 65°C. Watters (1976) also studied microwave radiation of *T. confusum* in wheat with different moisture contents, at a microwave power of 30 W, and a frequency of 8500 MHz. With wheat at high moisture content (15.6%), total insect mortality in shorter exposure times was achieved as compared to wheat at 12.5 and 8.5% moisture contents.

Shayesteh and Barthakur (1996) studied microwave radiation of *T. confusum* in wheat flour and *P. interpunctella* (Hübner) in red wheat at a frequency of 2450 MHz, at different power dosages and microwave modes (continuous and pulsed). Total mortality for *T. confusum* and *P. interpunctella* was obtained at 150 W, at 20 and 10 min, respectively. Pulsed mode was more effective than the continuous mode in killing insects. Mishenko et al. (2000) studied the influence of high frequency non-ionising radiation on *S. granaries* (L.), *S. oryzae* (L.), *Tenebrio molitor* (L.), and *Alphitobius diaperinus* (Pz) at frequencies of 10, 47.5, 900 and 2450 MHz. Exposure times ranged from 5 to 120 sec. Complete mortality was obtained in the continuous mode at both 900 and 2450 MHz frequencies.

2.4.1.3 Ionizing irradiation

Two types of ionizing irradiation have been considered for insect control in grain; γ -radiation produced from ^{60}Co or ^{137}Cs sources and accelerated electrons (Banks and Fields, 1995). Gamma irradiation of *S. granaries* in wheat at doses of 0.05 to 10 kGy was found to cause significant effects on soluble protein content and kernel hardness (Warchalewski et al., 2000). Doses of 3 to 5 kGy that cause death of stored-products insect pests within 24 hours, can cause significant damage to processed quality of wheat and other grains (Banks and Fields, 1995). According to Hasan & Khan (1998)

high dosages of ionising irradiation has a risk of vitamin loss of treated products. Irradiation can reduce levels vitamins A, C, E, B and K (Banks and Fields, 1995).

2.4.1.4 Desiccation (Inert dusts)

Inert dusts have traditionally been used in the grain industry for protection against insect pests (Ebeling, 1971). It is only in the last 60 years that these materials have been commercialised for use in grain protection technology (Golob, 1997). The most commonly used type of inert dust is Diatomaceous Earth (DE). DE is the fossil remains of diatoms (Golob, 1997). The sources of DE can either be marine or freshwater. The main constituent of DE is silica which makes up 90%, although other minerals like iron oxide, aluminium, magnesium and sodium are present (Banks and Fields, 1995).

Prior to processing of DE, it normally has a moisture content of 50% (Korunic, 1998). During processing the moisture content is reduced to between 2 and 6%. The final product is a talc-like fine powder, which is believed to be non-toxic to humans (Korunic, 1998). However, Korunic et al. (1996) raised concerns about lung damage (silicosis) development due to inhalation of DE dusts. The mode of action on insects of inert dusts is by desiccation (loss of water by insects). DE is known to absorb liquids two to three times its weight (Korunic, 1998). When insects crawl on DE material, the dust particles get trapped on the insect's protective wax coat on the cuticle. This results in approximately 30% loss of insect body weight, or 60% of their water, leading to death (Ebeling, 1971).

Aldryhim (1990) studied the response of *T. confusum*, *S. granarius*, and *R. dominica* when treated with a DE product (Dryacide) at 20 and 30°C and 40 and 60% relative humidity. *S. granarius* was found to be more susceptible to Dryacide than *T. confusum*, especially at 30°C. Under the same temperature conditions, Dryacide was more effective at 40% than 60% relative humidity. The study revealed that the effectiveness of Dryacide to *R. dominica* was also more pronounced at 30°C than 20°C. At high temperatures, the metabolic activity rate of insects is increased. Therefore more DE particles get trapped on the cuticle. Low relative humidity (40%) reduced progeny of *S. granaries* by 100% at both 20°C and 30°C. The progeny of *R.*

dominica was also greatly reduced at 40% relative humidity at both 20°C and 30°C. Some of the main problems with the use of inert dusts are that they affect the handling properties of grain mass, causing flowability reduction, a reduction in test weight, looseness, friction and dusty appearance of the grain mass (Korunic, 1998).

2.4.1.5 Controlled Atmosphere

Controlled atmosphere (CA), also known as modified atmosphere, is a disinfection technology that involves altering the natural storage gases to render the atmosphere in the store unfavourable to insect pests (Banks and Fields, 1995). The most commonly used gases are carbon dioxide (CO₂), oxygen (O₂), and nitrogen (N₂). CA has two modes of action which induce physiological and biochemical stress in pests: hypercarbia (increased CO₂ content) and hypoxia or anoxia (reduction of O₂ level) (Ofuya and Reichmuth, 2002). According to Bell and Armitage (1992) the raised level of CO₂ also promotes spiracular opening in the insects and this additionally increases water loss.

Annis and Morton (1997) studied the effect of 15 to 100% CO₂ on the development stages of *S. oryzae*. The development stages were incubated in wheat at 25°C and 60% relative humidity. They found that pupae were the most tolerant stage for all CO₂ concentrations and that eggs were the only stage with 100% mortality at 20% CO₂ for less than 30 days. Gunasekaran and Rajendran (2005) found the pupal stage of both *Stegobium paniceum* and *Lasioderma serricornis* to be the most tolerant stage when exposed to CO₂. The authors studied three different combinations of CO₂ concentrations: constant, increasing (30, 40 and 60%) and decreasing levels (60, 40, and 30%), at 27°C and 70% relative humidity. They concluded that mortality was more pronounced under both increasing and decreasing concentrations of CO₂ but not at constant concentrations.

2.4.2 Chemical pesticides

Non-residual chemical pesticides are often applied as fumigants. Fumigation is a method where pesticides in gaseous form at ambient temperatures and pressures,

and are applied to stored products at sufficient concentrations toxic to insects pests (Taylor, 1994).

2.4.2.1 Ozone

Ozone (O₃) is a strong antimicrobial oxidising agent. It has a number of applications in the food industry (Kim et al., 1999) including reducing the microbial load on surfaces. Ozone is naturally formed at small amounts by the action of solar ultraviolet (UV) irradiation on oxygen (Kim et al., 1999). When generated on site prior to use, it can be produced from oxygen in the air by radiation at 185 nm wavelength, emitted by high transmission UV lamps. Ozone is an attractive insect control measure as its degradation product is oxygen, therefore leaving no undesirable residues on products (Mendez et al., 2003).

Kells et al. (2001) investigated the efficacy of ozone as a non-residual chemical treatment to control insects in stored maize. Treatment of maize with 50 ppm ozone for 3 days was effective against *T. castaneum*, *S. zeamais*, and *P. interpunctella*. Mortality was 92-100%. The authors explain that ozone treatment happens in two phases. Phase 1 involves the slow movement of ozone through the maize with quick degradation. In Phase 2, the ozone moves freely with little degradation because molecular sites that are accountable for degradation are saturated. The rate of saturation is dependent on the velocity of ozone through the grain mass. For maize, it was found that 0.03 m/s was optimum. Based on this theory Mendez et al. (2003) examined ozone flow characteristics through stored wheat, which is less porous than maize. It was concluded that for wheat a higher velocity of 0.04 m/s was required.

2.4.2.2 Methyl bromide

Methyl bromide has been used around the world as a quarantine treatment for food processing plants, insects control in buildings and commodities, as well as for soil to control nematodes, weeds and pathogens (Fields and White, 2002). This fumigant is widely used because of its speed in killing the targeted organism. When compared to other fumigants, only methyl bromide can provide successful results in less than 24 hours (Taylor, 1994).

Methyl bromide has a boiling point of 3.6°C , and is odourless and colourless (Fields and White, 2002). The use of methyl bromide may not be ideal as a fumigant of bulk grain because Taylor (1994) observed that the use of methyl bromide to eradicate insects in silos was ineffective. The author found the penetrating properties of the gas though the grain inadequate.

Methyl bromide has been formally classified as an ozone depleting substance by the Parties to the Montreal Protocol (Taylor, 1994). It was decided that the use of the fumigant will be phased out by year 2005 in developed countries and 2015 in developing countries. Bromine is an efficient ozone depleter as it causes ozone to lose an oxygen atom (Fields and White, 2002). This causes the ozone layer to become thinner and allows additional ultraviolet B-radiation to reach the earth, thereby increasing the risk of skin cancer and cataracts in humans.

2.4.2.3 Phosphine

Phosphine was first used as a bulk grain fumigant in Germany around 1937 (Hairen and Davis, 1992). The fumigant is generated by the action of moisture in ambient air on a metal phosphide (Price, 1985). The metal phosphide can be either aluminium or magnesium, formulated with other ingredients in the form of tablets, plates, pellets and sachets. Phosphine has a characteristic odour, described as garlic. This odour seems to be due to the presence of other compounds produced along with phosphine and they may be preferentially absorbed during fumigation treatments (FAO, 1984). The effectiveness of phosphine as a disinfectant of grain and products requires 4 to 5 days (Taylor, 1999). Other disadvantages of phosphine are that it is more suited to use at higher temperatures (preferably $>15^{\circ}\text{C}$). It has a corrosion action on some metals including copper. Advantages of phosphine are that it is not strongly sorbed by most commodities, it diffuses and penetrates well, and leaves little or no chemical residues (Taylor, 1999).

2.4.2.4 Hydrogen cyanide

Hydrogen cyanide has been widely used for the control of insects in dry products such as cereals, milled cereal products, nuts, dried fruits and tobacco (Hooper et al., 2003). It has also been used for the disinfestation of rodents in places such as mills and ships. The fumigant has been overshadowed by other fumigants which are considered more efficient like methyl bromide (Food and Agriculture Organization, 2005). The disadvantages include its high dermal toxicity which makes it hazardous to applicators and also residues are a concern (Fields and White, 2002).

2.4.2.5 Insect resistance

Insect resistance to fumigants is a huge problem. According to Zettler and Cuperus (1990), the cause of insect resistance is inefficient fumigation practices and the misuse or over use of pesticides. During fumigation, susceptible insects will be killed while the resistant insects will not. The latter will reproduce and transfer the resistance genes to their offspring (Harein and Davis, 1992). Zettler and Cuperus (1990) found strains of *T. castaneum* and *R. dominica* resistant to phosphine. The ineffective application of phosphine to eradicate insects should therefore be ceased and correct methods utilized.

2.5 MICROWAVE MEASUREMENTS AND DIFFERENCES BETWEEN PULSED AND CONTINUOUS MICROWAVE HEATING

Microwave generators can be operated either in continuous or pulsed (intermittent) modes (Figure 2.7). In the former mode, energy is supplied continuously at constant power level whereas in the latter, energy is pulsed in an on-off manner (Mijović and Wijaya, 1990). In the pulsed mode, high intensity microwaves are produced for a period of few microseconds or milliseconds, with the power supply recharging in-between the pulses (personal communication with Thys Rossouw, Design Engineer, Delphius Technologies, Pretoria). According to Mankowski (2000) the fundamental purpose of all pulsed power systems is to convert a low-power, long-time input into a high-power, short-time output.

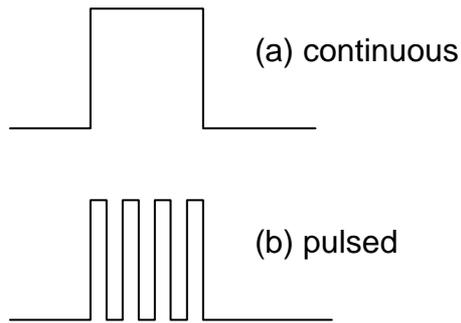


Figure 2.7: Different microwave modes (a) continuous output, (b) pulse output.

2.5.1 Microwave power measurements

Commonly used instruments to measure microwave power are of the thermal type; measuring the input power in terms of the heat produced in a water load (Lane, 1972). There are three categories of instruments; thermocouples, bolometers and calorimeters (Laverghetta, 1988).

2.5.1.1 Calorimeters

This instrument works on the dual load principle, where one load functions as power absorber while the second load acts as a temperature reference. A temperature sensor registers the difference in temperature of the two loads (Fantom, 1990). The calorimeter is used for power measurements from 10 W and higher (Laverghetta, 1988). They are known to be not accurate on one hand and convenience factors such as adequate sensitivity and rapid response on one hand (Fantom, 1990).

2.5.1.2 Bolometers

The bolometer measures power by using a temperature-sensitive resistor, known as the bolometer element. A temperature rise caused by microwave absorption produces a change in resistance. The instrument is direct heated, meaning the element functions both as power absorber and temperature sensor (Fantom, 1990). The instruments are sensitive power detectors, and are capable of measuring as little as a few microwatts of power (Laverghetta, 1988).

2.5.1.3 Thermocouples

A thermocouple is formed when two wires of different metals have one of their junctions at a higher temperature than the other (Laverghetta, 1988). The instrument measures the temperature rise of microwave treated water, and this temperature change is used as an indication of power. The primary applications of thermocouples are in the range of 0,1 W to 10 W (Laverghetta, 1988). According to Datta et al. (2001) thermocouples are not suitable to measure temperature in microwave oven, but can be used to measure temperature of food immediately after microwave treatment.

According to Fantom (1990), there are a few aspects that led to the possible elimination of thermocouples as power measurement instruments. Firstly, a thermocouple wire cannot be made as thin as the bolometer, due to the material used for thermocouples. Secondly, a thermocouple senses temperature only at a single point of the wire, whereas with the bolometer temperature is sensed throughout the length of the wire. This is advantageous due to its low cost (Childs et al., 2000)

2.5.2 Temperature measurements

Metallic thermocouples are unsatisfactory for measuring temperature in microwave ovens, due to the fact that they absorb energy and cause electromagnetic field disturbances during heating, which may result in variability in heating patterns. Thermocouples which are shielded with aluminium material can be used to measure temperature during microwaving (Datta et al., 2001).

Fiberoptic thermometers can also be used to measure temperature. The material used for fibres and sensors are non-metal, and electrically non-conducting (Datta et al., 2001). There are two types of fiberoptic thermometers. One type measures temperature at points along the length of the fiberoptic cables, and the other type measures temperature at one point normally at the end of the cable (Datta et al., 2001). Ideally a temperature measurement instrument should be used in the microwave oven and it should sense temperature change throughout the wire, thus those with continuous measure are favoured.

2.5.3 Pulsed and continuous microwave processing

Gunasekaran (1990) studied the use of continuous and pulsed microwave modes for the drying of maize. The power setting for both modes was 250 W. Drying was more rapid in the continuous mode than the pulsed mode, but the continuous mode required higher total power input. In the pulsed mode, an increase in power-off times assisted in reducing the total power required for drying.

Yang and Gunasekaran (2001) studied the temperature distribution in a 2% agar gel during continuous and pulsed microwave heating. A hot spot at the centre of the agar was observed during heating. The spot was severe in the continuous mode, but less significant in pulsed microwave mode especially when longer pulsed power-off times were employed. The power-off times allowed distribution of temperature, which minimised the development of hot spots.

Yang and Gunasekaran (2004) proposed models for predicting the interior temperature distribution during pulsed and continuous microwave heating based on Lambert's law and Maxwell's equations. The authors reported that better temperature uniformity in agar samples was observed during pulsed microwave heating.

Gunasekaran and Yang (2007) optimised a pulsed microwave heating process for precooked mashed potato cylinders. The power-on and power-off temperature constraints were found to be very critical in the optimisation of pulsed microwave heating. The authors concluded that power-on temperature constraints produce a suitable temperature gradient while the power-off constraint allows temperature equalization within the sample to occur.

Even though the application of continuous mode may result in quicker drying of grain products, it requires more power input (Gunasekaran, 1990). The application of pulsed microwave heating results in consistent temperature distribution in the heated product and power-off periods provide time for moisture redistribution throughout the product (Gunasekaran and Yang, 2007).

2.6 EFFECT OF MICROWAVE ENERGY ON MAIZE QUALITY

Heating of food can induce physical changes or chemical reactions, e.g. protein denaturation, browning and starch gelatinisation, which can affect sensory characteristics of food products (Lewis and Heppell, 2000). The primary advantages of microwave heating over conventional heating are that it generates heat faster and saves energy (Thostenson and Chou, 1999). However, microwave heating has shortcomings such as uneven heat distribution (Oliveira and Franca, 2002) during processing of certain foods.

The effect of microwave energy on the drying of maize (Shivhare, 1992; Velu et al., 2006), and wheat (Walde et al., 2002) has been investigated. Shivhare (1992) studied varying power levels at 0.25 W/g and 0.75 W/g. Negative effects on germination, test weight and physical changes were observed when power level of 0.75 W/g. Velu et al. (2006) studied dry milling characteristics of microwaved dried maize. The proximate composition of grains and ground products showed no change in protein and starch contents. However, viscosity was found to decrease with an increase in microwave drying times. The authors concluded that the viscosity changes indicated that microwave drying had an effect on the structure of protein and starch. Walde et al. (2002) found that microwave drying had an effect on the structural and functional characteristics of wheat gluten content.

Kirkpatrick and Roberts (1971) studied the use of microwave energy to control insects in wheat. Exposure of insect infested wheat to microwaves at 2 kW for 10 sec did not result in total insect mortality, but a reduction of 8% in germination was observed in the treated wheat. Lewandowicz et al. (2000) studied the effect of microwave irradiation on the physicochemical properties and structure of wheat and maize starches. Starch samples at 30% moisture content were treated at 0.5 W/g microwave power for 60 min. Microwave radiation caused a shift in the gelatinisation range to higher temperatures, and a drop in solubility and crystallinity was observed. Szepes et al. (2005) investigated the effect of microwave irradiation on maize (6.8% moisture content) and potato (9.7% moisture content) starches when treated for 15 minutes in a domestic oven. The crystallinity of maize starch decreased while it increased for potato starch. Hamid and Boulanger (1970) studied the baking qualities

of microwave treated wheat, at temperatures of 55, 65 and 80°C. The treatments had no effect on the milling quality or the protein content of the wheat. However, the breadmaking quality was affected deleteriously and progressively more as the treatment temperature increased. Hoffman and Zabik (1985) studied the effects of microwave cooking on nutrients in food systems. The authors observed that equal or better retention of thiamine, pyridoxine, folacin, riboflavin and ascorbic acid was achieved during microwave heating as compared to convectional heating.

2.7 MEASUREMENT METHODS OF MAIZE QUALITY (PHYSICAL PROPERTIES)

2.6.1 Moisture content

The moisture content of grain is an important factor as it influences subsequent processing required after harvesting as well as the pricing in the market (Johnson and Lamp, 1966).

Moisture content measurement methods are divided into two classes, namely direct and indirect methods. The direct method uses heat to remove moisture from the sample, and moisture content percentage result is based on the weight lost or removed water (Johnson and Lamp, 1966). The indirect method involves the measurement of a property of material that depends upon moisture content (Johnson and Lamp, 1966).

Direct methods are accurate but time consuming, while indirect methods are common for their rapidness and simplicity but their accuracy may be poor (Paulsen et al., 2003). One of the common standardised direct method is the oven method (Johnson and Lamp, 1966). Electromagnetic fields have been used to measure moisture content of maize (Kraszewski and Nelson, 1994). The method provided reliable results, and was found to be fast and non-destructive. The other common method for moisture content analysis is electrical meters. The meter introduces grain into an electrical circuit and measures the resistance/ dielectric constant of the grain (Pande, 1975). The advantages are that these meters can be calibrated for a variety of grains and can be used anywhere and anytime. One of the disadvantages is that

the meters only measure free water and cannot measure bound water even though a constant value is used (Pande, 1975). It is assumed that the amount of bound water is constant, which may not be case.

2.6.2 Hardness tests

Hardness of maize, often referred to as the amount of vitreous (hard) endosperm in the kernel relative to the amount of floury (soft) endosperm, is of great importance to producers and processors in the grain industry as it affects the grinding power requirements (Paulsen et al., 2003). Vitreous and floury endosperm character is hereditary, but can also be influenced by the environment (Paulsen et al., 2003).

There are a several measurement methods used for the determination of hardness (vitreousness) in cereal grains. Vitreous/opaque ratio of kernels can be estimated by hand dissection (Yuan and Flores, 1996), and Felker and Paulis (1993) combined the use of hand dissection and image analysis. The most common method of determining grain hardness, the Stenvert hardness test, was studied by Pomeranz et al. (1984, 1985, 1986a); Kirleis and Stroshine, (1990); Li et al. (1996). It measures maize grain resistance to grinding, height of column of ground maize (as index of packing and fluffiness) and ratio of coarse to fine particles (determined by weight of sieved fractions). The particle size index was studied by Abdelrahman and Hosney (1984), which measures the weight of particles retained on screens (150 μm openings) after grinding, which indicates the harder endosperm. The near infrared reflectance (NIR) was studied by Pomeranz et al. (1986b). The endosperm hardness determination estimates are conducted at a wavelength of 1 680 nm. Recently, the use of image analysis to measure vitreousness in maize has gained widespread use due to its rapidness and its non-destructive approach (Nielsen, 2003; Erasmus and Taylor, 2004).

2.6.3 Kernel weight

Kernel weight gives an indication of kernel size (Paulsen et al., 2003). It involves the weighing of one hundred representative whole maize kernels without visual cracks or any other mechanical damage. Another indicator of physical size is the percent thins

test (Paulsen et al., 2003). The test involves sieving of 250 g of maize over a 7.94 mm sieve. The amount passing through the sieve, expressed in a weight percentage is known as the percent thins.

2.6.4 Test weight

Test weight is a measure of bulk density which is obtained by weighing a specific volume of grain (Paulsen et al., 2003). The mass of grain is a combination of the grain and the voids between the grains (Hoseney and Faubion, 1992). Measurement of test weight is important in storing and transporting maize because it determines the volume required for a given lot of maize (Paulsen et al., 2003). A sample is placed in a funnel holder while the hopper valve is closed. Once the hopper valve is opened, the sample will drop in a kettle/ container. The maize is weighed and calculated in, for example, kilograms per hectolitre. Moisture content affects test weight. Maize at high moisture content has low test weight (Paulsen et al., 2003). The drying of maize at high temperatures was found to affect test weight negatively (Brown et al., 1979; Peplinski et al., 1994). Test weight is one of the factors that affect the yield of dry milling products. Paulsen and Hill (1985) found that to obtain high yield of large flaking grits, maize should have high test weight and low breakage susceptibility.

2.6.5 Stress Cracks

Stress cracks are internal fissures in maize kernels that extend from the vitreous endosperm towards the pericarp (Paulsen et al., 2003). The causes of stress cracks are induced moisture gradients or stresses caused during drying at high temperatures with subsequent rapid cooling (Gunasekaran and Muthukumarappan, 1993).

Kirleis and Stroshine (1990) inspected maize kernels for stress cracks by placing kernels on a light box with the germ side placed downwards toward the light source. The authors developed a stress crack index (SCI) that indicates the severity of the stress cracks. The equation of $SCI = (\% \text{ single stress cracks} \times 1) + (\% \text{ double stress cracks} \times 3) + (\% \text{ multiple stress cracks} \times 5)$. The detection of stress cracks in maize

kernels can be detected by image analysis methods (Gunasekaran et al., 1987) such as frequency domain (Hang et al., 1996) and magnetic resonance imaging (Song and Litchfield, 1994).

2.8 CONCLUSIONS

The use of chemical pesticides as an insect control measure has shortcomings. For example, methyl bromide, the most commonly used pesticide for insect infestation, is being phased out by 2015 as it is ozone depleting. The most commonly used pesticide, phosphine, requires a few days for it to be effective. Secondly, the possibility of insects building up resistance to pesticides caused by inefficient practises cannot be ignored.

A cost effective, alternative method other than chemical pesticides is needed to eradicate insect storage pests in maize. The ideal method should have little or no effect on maize quality. Microwave energy has been investigated as a tool to control insect infestation in cereal grains. There is evidence that the use of continuous microwave energy is effective in the killing of insect pests in cereal grains but continuous microwaves can have a negative effect on grain quality. Thus, this research will investigate the use of pulsed microwave mode versus continuous mode on the control of 5 common insect species that infest maize grain. The effect of microwave energy on the physicochemical properties of maize will be investigated. Parameters to be studied include microwave mode, power level and exposure time.

3. OBJECTIVES AND HYPOTHESES

3.1 OBJECTIVES

- To determine the optimal microwave conditions for the eradication of insect storage pests in maize kernels.
- To determine the effect of microwave energy on maize quality, subsequent to optimisation of the process for the eradication of insects pests.

3.2 HYPOTHESIS

- Microwave energy will result in the scalding of insect storage pests due to selective heating, related to the different dielectric properties between insects and maize kernels.
- The optimised microwave process will not cause significant thermal damage of maize kernels because of the low microwave energy absorption and the short exposure time.

4. RESEARCH

The two research chapters were written according to the format required by the journal *Cereal Chemistry*.

Experimental design

The experimental design that was followed to determine the optimum microwave conditions to control insect infestation of maize kernels, and the effect of microwave energy on the physicochemical properties of maize kernel quality is explained in Figure 4.1.1.

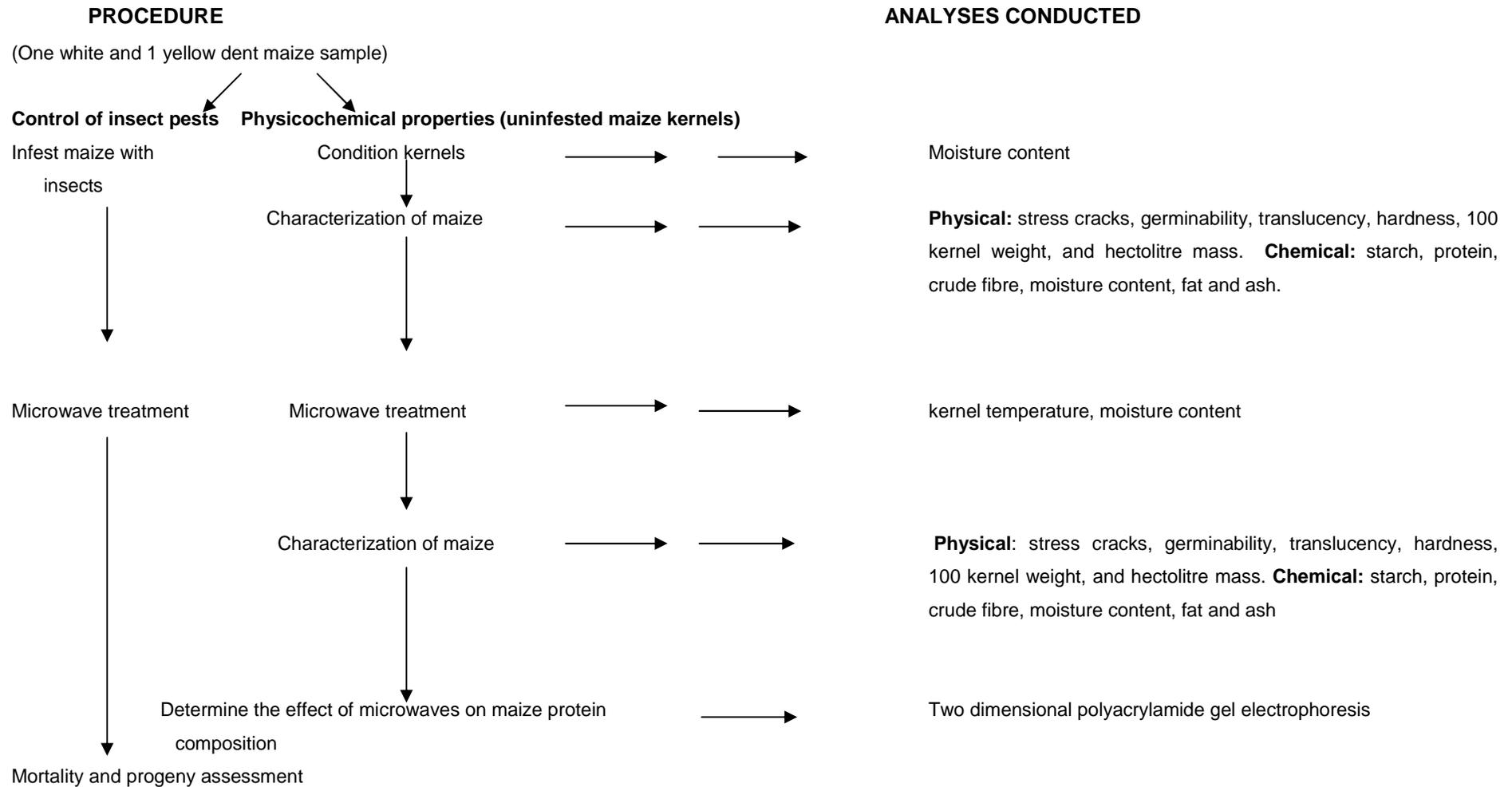


Figure 4.1.1: Experimental design followed to determine the optimum microwave conditions to control insect infestation in maize kernels, and to investigate the effect of microwave energy on the physicochemical properties on maize kernel quality.

4.1 MICROWAVE ERADICATION OF MAIZE KERNEL INSECT STORAGE PESTS

ABSTRACT

Forty six microwave treatment conditions were investigated to identify and optimize a condition that was able to eradicate five common insect storage pests in maize in South Africa, namely *Sitophilus zeamais* (Motschulsky), *Rhizopertha dominica* (Fabricius) *Ephestia cautella* (Walker), *Cryptolestes ferrugineus* (Stephens) and *Tribolium confusum* (Jacquelin du Val) Two microwave processing modes, namely continuous and pulsed modes and other parameters such as length of microwave cavity, power dosage, and exposure time were investigated. Pulsed mode microwave was found more effective than continuous mode. Exposure time was a significant parameter. Too short exposure times did not eradicate insects, while too long exposure times (use of 728 mm microwave cavity) resulted in kernel swelling, localised popping and discolouration. The microwave treatment identified to eradicate all insect species and their developmental stages without visible kernel damage was pulsed mode at 2450 MHz frequency, using a 483 mm long microwave cavity, at a power level of 1.5 kW, with an exposure time of 9 sec.

Keywords: insect storage pests, pulsed microwave energy, maize quality

4.1.1 INTRODUCTION

Protection of stored grain from insect infestation and damage is a worldwide problem (Rashkovan et al 2003). Not only does insect infestation cause losses of stored grain (Warchalewski et al 2000; Rashkovan et al 2003) it can also adversely affect grain quality (Jood et al 1992a; 1992b; Jood and Kapoor 1992). Chemical pesticides are generally used to control insect infestations (White, 1995). However, according to Kent and Evers (1994) the disadvantages on the use of chemical pesticides include the insects' ability to build up resistance to pesticides, and if contact pesticide is used, chemical residue can be left on the surface of the product after treatment. These problems have led to research on the possible use of microwave energy as an

alternative insect control measure (Nelson 1996). Baker et al (1956), Hamid and Boulanger (1970), Kirkpatrick and Roberts (1971), Watters (1976), Halverson et al (1996), Shayesteh and Barthakur (1996) and Mishenko et al (2000) studied the potential use of microwave radiation as a control measure of insect infestation in cereal and cereal products. The different dielectric properties between insects and grains appear to make it possible to selectively heat insects without damaging the cereal grains (Nelson 1972). This is achievable as insects are known to have moisture contents 3-5 times higher than those of grains (Mishenko et al 2000).

Watters (1976) investigated microwave radiation for the control of *Tribolium confusum* (Jacquelin du Val) adults in wheat with different moisture contents. A microwave power output of 30 W, operated in the pulsed mode at a frequency of 8500 MHz was used. With wheat at high moisture content (15.6%), 100% insect mortalities in shorter exposure times were achieved as compared to wheat at 12.5 and 8.5% moisture contents. The authors concluded that microwave treatment time was directly proportional to moisture content.

Shayesteh and Barthakur (1996) studied microwave radiation of *T. confusum* in whole wheat flour and *Plodia interpunctella* (Hübner) in red wheat at a frequency of 2450 MHz. Both continuous and pulsed modes were investigated at different power outputs of 75, 100, and 150 W at exposure times of 5, 10, 20 and 40 min. The authors concluded that the pulsed mode was more effective than the continuous mode in killing insects. Mishenko et al (2000) studied the influence of high frequency non-ionising radiation on *Sitophilus granaries* (L.), *S. oryzae* (L.), *Tenebrio molitor* (L.), and *Alphitobius diaperinus* (Pz) at frequencies of 10, 47.5, 900 and 2450 MHz. Exposure times ranged from 5 to 120 sec. Complete mortality was obtained in the continuous mode at both 900 and 2450 MHz frequencies.

There has been considerable research on the use of microwave heating to control insect infestation in wheat but little or no research done on maize. It is known that the status of any particular insect pest may vary between different grain commodities, different varieties of the same commodity, different climatic regions and agro-industrial systems and between different socio-economic groups (FAO, 2007). The hot climate in the African continent plays a major role in the high survival or pesticide

resistance of storage pests (FAO, 2007). The primary objective of this study was to determine a microwave treatment condition that would eradicate five insect species and all its life stages, namely *Sitophilus zeamais* (Motschulsky), *Rhizopertha dominica* (F.), *Ephestia cautella* (Walker), *Cryptolestes ferrugineus* (Stephens), and *T. confusum* which are commonly found in maize and maize products (FAO 2007), without the treatment affecting maize quality

4.1.2 MATERIALS AND METHODS

4.1.2.1 Insect species

Five insect species were used in the study: *S. zeamais* (maize weevil) adults; *R. dominica* (lesser grain borer) adults; *E. cautella* (tropical warehouse moth) eggs; *T. confusum* (confused flour beetle) adults; and *C. ferrugineus* (rust grain beetle) adults. The insects were obtained from the Agricultural Research Council, Plant Protection Research Institute, Roodeplaat, South Africa.

4.1.2.2 Propagation of insects

Adults of *R. dominica*, *S. zeamais*, *C. ferrugineus*, *T. confusum* and eggs of *E. cautella* were propagated in 2 L jars containers on different substrates, and under different temperature and relative humidity conditions. *R. dominica* and *S. zeamais* were propagated on whole-wheat kernels 30-34°C and 70% relative humidity (RH). *T. confusum* was propagated on milled wheat 31-34°C at 50-55% RH, *C. ferrugineus* on broken or milled sunflower seeds and oats, at 31-34°C and 50-55% RH, and *E. cautella* on a mixture of milled wheat, milk powder, honey and yeast at 31-34°C and 50-55 RH. For infestation of maize kernels, 500 g each of white and yellow maize whole kernels (13-15%) were infested with 250 adult insects (of each beetle species). The maize samples (white and yellow) were incubated for two weeks prior to microwave treatment. During this stage the adults laid eggs which developed through different stages (larvae, and adults). For *E. cautella*, 200 g each of white and yellow maize kernels were infested with 0.3 g of 24 hour old eggs. Maize kernels were then infested with eggs 24 hours prior to microwave treatment. For the microwave

treatment, the infested maize kernels were divided into masses of 10 g per glass test tube (Figure 4.1.2a). Controls (infested maize) were propagated under the same conditions as mentioned above for each insect species were not microwaved. The number of eggs/larvae/adults per test tube was not measured before treatment. Vials containing adults only with no maize kernels were microwave treated as well (Figure 4.1.2b). Each vial contained 30 adult beetles.

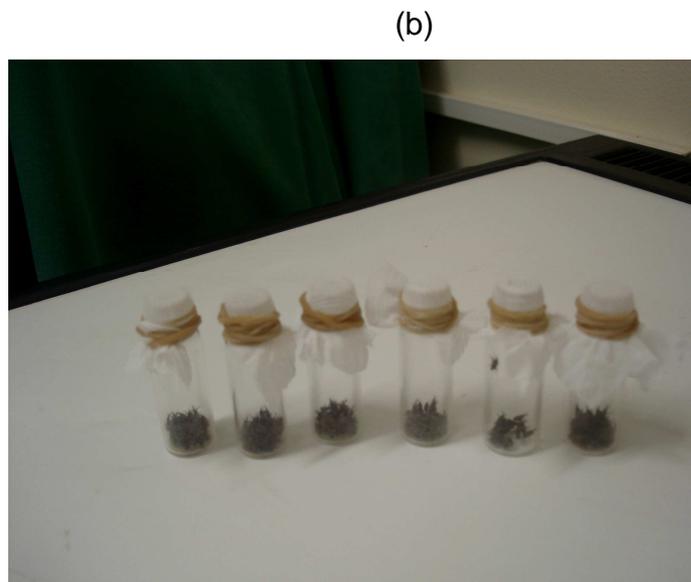
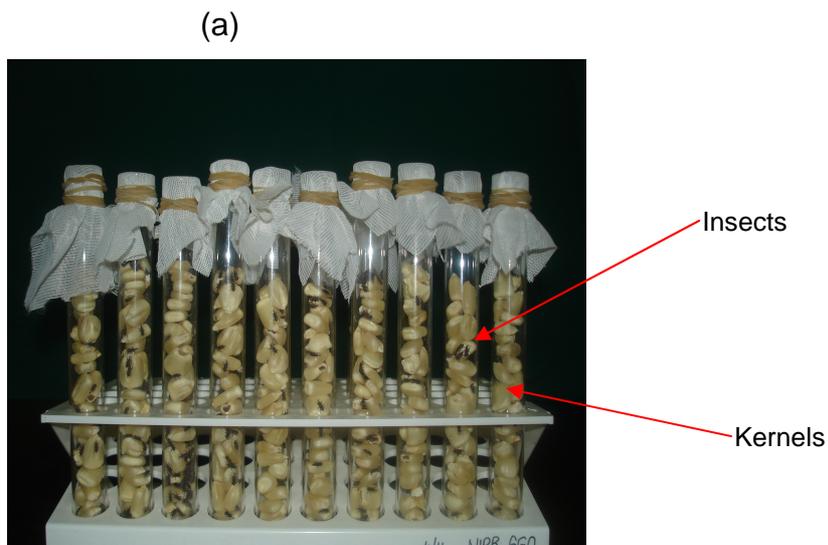


Figure 4.1.2: Presentation of maize kernels infested with insect pests for microwave eradication studies. (a) Insect infested white maize in glass test tubes. (b) Insects in small vials, no maize kernels.

4.1.2.3 Microwave unit

A microwave unit capable of being operated either on continuous or pulsed mode was used. A 2 kW source Power Generator PM740, (Alter Power Systems, Italy) operated at a frequency of 2450 MHz was used to generate the microwave field. In continuous mode, a fraction of this total 2 kW power could be selected and was applied with 100% duty. In pulsed mode, a duty cycle of 25% was used, so that a maximum power of four times that which was applied in the continuous mode was delivered during the pulses, while the average power applied remained the same as that used in the continuous mode. Figure 4.1.3 illustrates the microwaving treatment unit.

4.1.2.4 Microwave treatment

Maize kernels were microwave treated using two different systems: free fall and pulley systems. The former was evaluated first. This system allowed vials (Figure 4.1.2b) containing insects to be exposed to microwaves by falling (gravity) through a quartz tube within a microwave cavity (728 mm long, Figure 4.1.3). The latter system involved the use of an extra internal quartz tube, which could move freely within the stationary quartz tube. The vials containing insects only without kernels (Figure 4.1.2b) were placed in the internal quartz tube. The contents of a test tube (insect infested maize kernels) were emptied into the internal tube prior to microwave treatment and returned back into test tube after treatment. The insects were exposed to microwaves for different treatment times.

For the free fall system, the microwave cavity had a length of 728 mm, placed at a lengthwise angle of 45° to the horizontal. A vial containing 30 insects was inserted at the top of the quartz tube (22 mm internal diameter (ID), 1010 mm length) (Figure 4.1.3, D) and allowed to fall. The exposure time of the insect vials in the microwave cavity ranged between 2 and 3 sec. In order to obtain high insect mortality, insect vials were exposed to microwaves more than once (multiple passes).

In the pulley system, insect infested maize kernels were emptied into a quartz tube [19 mm (ID), 498 mm length] (Figure 4.1.3, B). The tube was plugged at the bottom with glass wool. A nylon string, attached to the top end of the tube, and the other end

of the string attached to a spindle driven by an electric motor (Figure 4.1.3, A) allowed the insect infested maize to be pulled at a constant rate through the external quartz tube. Infested maize kernels were exposed to microwaves only once (single pass).

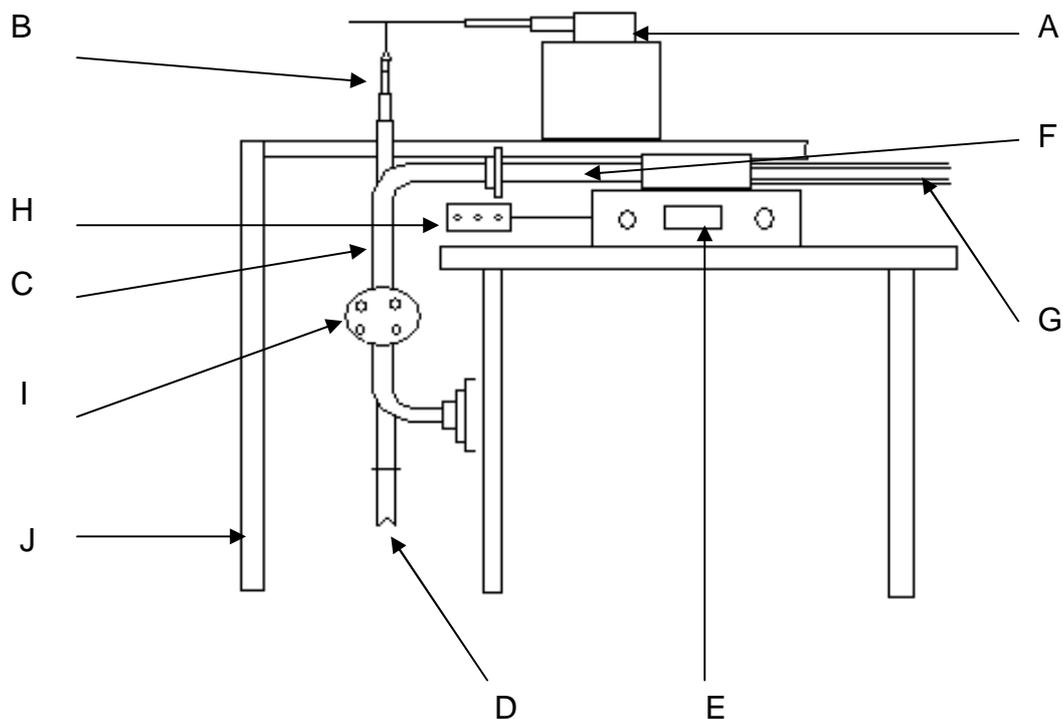


Figure 4.1.3: Combined Pulsed/ Continuous microwave unit used for treatment of maize kernels (front view). (A) Electric motor turning spindle; (B) inner quartz tube; (C) wave guide; (D) outer quartz tube; (E) Power generator (F) microwave power guide and (G) cooling water in/out; (H) remote switch; (I) flange; (J) table. Angle of microwave cavity (quartz tube) to the horizontal not shown in this figure.

The motor speed could be adjusted to give different exposure times for insects in the microwave field. The length of the microwave cavity was also varied, from 728 mm to 483 mm, by removing the middle section of the cavity. The microwave cavity was at an angle of 45° to the horizontal.

4.1.2.5 Insect mortality assessment

Insect mortality was assessed in two ways. For vials containing adult beetles only, the percentage mortality was calculated by counting the number of dead insects after treatment, and expressing as a percentage of the total number of insects. For insect infested maize kernels, microwave treated maize kernels and controls were incubated for six weeks at 28°C and 55-70% relative humidity to determine progeny development of adult insects. If no live insects were observed after incubation, then the microwave treatment was considered effective in eradication of the insects and their development stages.

4.1.2.6 Preliminary microwave studies

In preliminary work only *R. dominica* and *S. zeamais* species were investigated. Varying microwave conditions were investigated to eradicate the two insect species. Successful microwave treatment conditions were used as baselines to treat three additional insect species namely, *E. cautella* (eggs), *T. confusum* and *C. ferrugineus*.

For the determination of the most effective treatment conditions for eradication of insect species, several parameters: microwave mode and length of cavity, insect species, power level, and exposure times were studied. In the free fall system *R. dominica* and *S. zeamais* species were microwave treated at two different power levels of 1.8 and 2.0 kW in pulsed mode only. In the pulley system insect species were treated at different power levels ranging between 0.5 and 2 kW, using the 728 and 483 mm long microwave cavities, under pulsed and continuous microwave modes at exposure times varying between 0 and 18 sec.

4.1.2.7 Statistical analysis

Analysis of data performed using Statgraphics Centurion, Version XV (Statpoint Incorporated, Herndon, Virginia, USA). Multifactor analysis of variance (ANOVA) was applied using the LSD test. All data was considered significant at $p < 0.01$.

4.1.2 RESULTS

Table 4.1.1: Effect of microwave power and number of exposure times on the mortality of *S. zeamais* and *R. dominica* using a free falling microwave system with a cavity length of 728 mm, operated in pulsed mode at 2450 MHz frequency

Insect species	Power (kW)	Number of times insects exposed to microwave treatment *	Mortality #†
<i>S. zeamais</i>	1.8	5	100 ± 0.0
	1.8	5	100 ± 0.0
	1.8	6	100 ± 0.0
	1.8	8	100 ± 0.0
	2.0	3	100 ± 0.0
	2.0	3	100 ± 0.0
<i>R. dominica</i>	1.8	9 ^	98.7 ^{bc} ± 2.3
	1.8	11	94.0 ^a ± 2.0
	1.8	18	95.3 ^{ab} ± 3.1
	1.8	20	96.7 ^{abc} ± 1.2
	2.0	8	100.0 ^c ± 0.0
	2.0	10	99.7 ^{bc} ± 0.6

Number of dead insects after treatment divided by the number of alive insects (mean and standard deviation).

* All treatments were done in triplicate.

† Values with different superscripts within a block are statistically significantly different ($p < 0.01$)

^ Data for less than 8 times not shown

Complete mortality of *S. zeamais* at both 1.8 and 2 kW power levels, with 5 and 3 times microwave exposure, respectively, was achieved. Complete mortality of *R. dominica* was only obtained at the higher power level of 2 kW, and the adult insects had to be repeatedly exposed to microwaves at least 8 times.

Table 4.1.2: Effect of microwave power level and exposure time on the mortalities of *S. zeamais* and *R. dominica* species and their progeny, and physical damage to white maize kernels when infested maize was microwave treated once (single exposure time) in the pulsed mode using the pulley system, with a 728 mm long microwave cavity, at 2450 MHz frequency

Insect species	Pulsed mode		100% mortality Yes/No	Visible damage to maize kernels after treatment	Number of insect progeny #
	Power level (kW)	Exposure time * (sec)			
<i>S. zeamais</i> – control	0	0	No	None	19
<i>R. dominica</i> – control	0	0	No	None	10
<i>S. zeamais</i>	0.5	3	No	None	nd
	0.5	6	No	None	nd
	0.5	9	No	None	nd
	0.5	12	No	None	nd
<i>R. dominica</i>	0.5	3	No	None	nd
	0.5	6	No	None	nd
	0.5	9	No	None	nd
	0.5	12	No	None	nd
<i>S. zeamais</i>	1.0	3	No	None	nd
	1.0	6	No	None	nd
	1.0	9	No	None	nd
	1.0	12	Yes	None	0
<i>R. dominica</i>	1.0	3	No	None	nd
	1.0	6	Yes	None	0
	1.0	9	Yes	None	0
	1.0	12	Yes	None	0

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Table 4.1.2 continued

<i>S. zeamais</i>	1.5	3	No	None	nd
	1.5	6	No	None	nd
	1.5	9	Yes	None	0
	1.5	12	Yes	Popping, swelling, and discolouration	0
<i>R. dominica</i>	1.5	3	No	None	nd
	1.5	6	Yes	None	0
	1.5	9	Yes	None	0
	1.5	12	Yes	Popping, swelling, and discolouration	0
<i>S. zeamais</i>	2.0	3	No	None	nd
	2.0	6	No	None	nd
	2.0	9	Yes	Popping, swelling, and discolouration	0
	2.0	12	Yes	Popping, swelling, and discolouration	0
<i>R. dominica</i>	2.0	3	No	None	nd
	2.0	6	No	None	nd
	2.0	9	Yes	Popping, swelling, and discolouration	0
	2.0	12	Yes	Popping, swelling, and discolouration	0

nd = Progeny not determined in samples which had live insects after treatment, except controls, as the objective was to determine whether eggs would hatch after treatment.

* All treatments were done in triplicate. Exposure times were once-off, no repetitions were performed.

Progeny: microwave treated maize kernels were incubated for 6 weeks to determine if eggs in kernels hatched. Average values for the number of progeny are given.

In the pulsed mode using the pulley system with a 728 mm long microwave cavity, the two lowest dosage microwave treatments that resulted in complete mortality of both *S. zeamais* and *R. dominica* species, with no progeny and no visible kernel damage were at 1 and 1.5 kW power levels for 12 and 9 sec exposure times, respectively. Other treatments giving complete mortality (1.5 kW for 12 sec; and 2.0 kW for 9 and 12 sec) resulted in visible kernel damage in terms of swelling, popping, and discolouration.

Table 4.1.3: Effect of microwave power level and exposure time on the mortalities of *S. zeamais* and *R. dominica* species and their progeny, and physical damage to white maize kernels when insects and maize were microwave treated once (single exposure time) in the continuous mode using the pulley system, with a 728 mm long microwave cavity, at 2450 MHz frequency

Insect species	Continuous mode		Exposure time * (sec)	100% mortality Yes/No	Visible damage to maize kernels after treatment	Number of insect progeny #
	Power level (kW)					
<i>S. zeamais</i> – control	0		0	No	None	19
<i>R. dominica</i> – control	0		0	No	None	10
<i>S. zeamais</i>	0.5		3	No	None	nd
	0.5		6	No	None	nd
	0.5		9	No	None	nd
	0.5		12	No	None	nd
<i>R. dominica</i>	0.5		3	No	None	nd
	0.5		6	No	None	nd
	0.5		9	No	None	nd
	0.5		12	No	None	nd
<i>S. zeamais</i>	1.0		3	No	None	nd
	1.0		6	No	None	nd
	1.0		9	No	None	nd
	1.0		12	Yes	None	0
<i>R. dominica</i>	1.0		3	No	None	nd
	1.0		6	Yes	None	0
	1.0		9	Yes	None	0
	1.0		12	Yes	None	0

PTO

Table 4.1.3 continued

<i>S. zeamais</i>	1.5	3	No	None	nd
	1.5	6	Yes	None	0
	1.5	9	Yes	Discolouration	0
	1.5	12	Yes	Popping, swelling, and discolouration	0
<i>R. dominica</i>	1.5	3	No	None	nd
	1.5	6	Yes	None	0
	1.5	9	Yes	Discolouration	0
	1.5	12	Yes	Popping, swelling, and discolouration	0
<i>S. zeamais</i>	2.0	3	No	None	nd
	2.0	6	Yes	None	0
	2.0	9	Yes	Popping, swelling, and discolouration	0
	2.0	12	Yes	Popping, swelling, and discolouration	0
<i>R. dominica</i>	2.0	3	No	None	nd
	2.0	6	Yes	None	0
	2.0	9	Yes	Popping, swelling, and discolouration	0
	2.0	12	Yes	Popping, swelling, and discolouration	0

nd = Progeny not determined in samples which had live insects after treatment, except controls, as the objective was to determine whether eggs would hatch after treatment.

* All treatments were done in triplicate. Exposure times were once-off, no repetitions were performed.

Progeny: microwave treated maize kernels were incubated for 6 weeks to determine if eggs in kernels hatched. Average values for the number of insect progeny are given.

In the continuous mode using the pulley system with a 728 mm long microwave cavity, the three lowest dosage microwave treatments that resulted in complete mortality of both *S. zeamais* and *R. dominica* species, with no progeny and no visible kernel damage were at 1, 1.5 and 2 kW power levels for 12, 6 and 6 sec exposure times, respectively. Other treatments giving complete mortality (1.5 and 2 kW both for 9 and 12 sec) resulted in visible kernel damage in terms of swelling, popping, and discolouration.

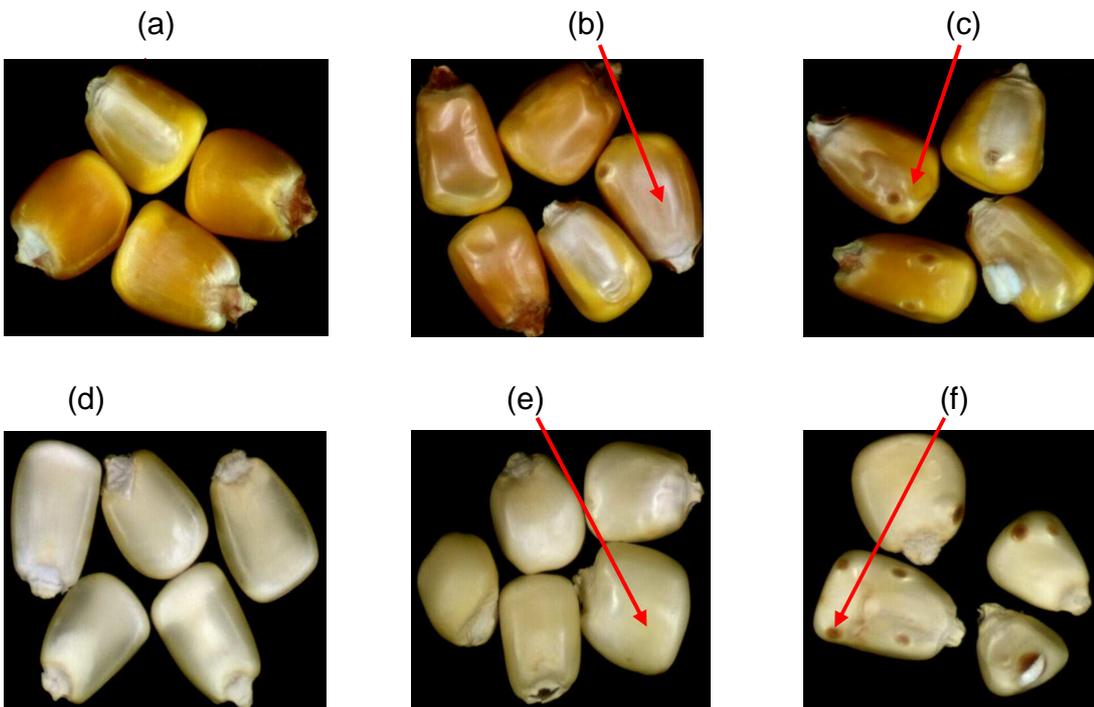


Figure 4.1.4: Physical damage to white and yellow maize kernels by high power levels with long exposure times. (a) yellow maize controls; (b) swelling; (c) discolouration; (d) white maize controls; (e) swelling; (f) discolouration.

The microwave treatments that resulted in visible physical damage of maize kernels were at pulsed mode 1.5 and 2.0 kW power levels for 12 sec, and both 9 and 12 sec exposure times, respectively. For the continuous mode, they were 1.5 and 2.0 kW power levels both for 9 and 12 sec exposure times.

Table 4.1.4: Effect of microwave power level and exposure time on the mortalities of *S. zeamais* and *R. dominica* species and their progeny, and physical damage to white and yellow maize kernels when insects and maize were microwave treated once (single exposure time) in the continuous and pulsed modes using the pulley system, with a modified 483 mm long microwave cavity, at 2450 MHz frequency

Maize type	Microwave Mode	Power level (kW)	Insect species	Exposure time * (sec)	Visible damage on kernels	100% mortality Yes/ No	Progeny Development # Yes/ No
White	Controls	None	<i>S. zeamais</i> <i>R. dominica</i>	None None	None None	No No	Yes Yes
	Pulsed	1.0	<i>S. zeamais</i> <i>R. dominica</i>	9 9	None None	Yes Yes	No Yes
	Pulsed	1.5	<i>S. zeamais</i> <i>R. dominica</i>	7 7	None None	Yes Yes	No No
	Continuous	1.5	<i>S. zeamais</i> <i>R. dominica</i>	4 4	None None	Yes Yes	No Yes
	Continuous	2.0	<i>S. zeamais</i> <i>R. dominica</i>	4 4	None None	Yes Yes	No No
Yellow	Controls	None	<i>S. zeamais</i> <i>R. dominica</i>	None None	None None	No No	Yes Yes
	Pulsed	1.0	<i>S. zeamais</i> <i>R. dominica</i>	9 9	None None	Yes Yes	No Yes
	Pulsed	1.5	<i>S. zeamais</i> <i>R. dominica</i>	7 7	None None	Yes Yes	No No
	Continuous	1.5	<i>S. zeamais</i> <i>R. dominica</i>	4 4	None None	Yes Yes	No No
	Continuous	2.0	<i>S. zeamais</i> <i>R. dominica</i>	4 4	None None	Yes Yes	No No

* All treatments were done in triplicate. Exposure times were once-off, no repetitions were performed.

Progeny: microwave treated maize kernels were incubated for 6 weeks to determine if eggs in kernels hatched.

The exposure times of the successful treatments (100% mortality, no progeny and no visible kernel damage) in Tables 4.1.2 and 4.1.3 (unmodified, 728 mm long cavity used), were reduced with the modified, 483 mm short cavity. The 6 sec exposure time was reduced to 4 sec, the 9 sec exposure time to 7 sec, and the 12 sec exposure time to 9 sec. Both pulsed and continuous microwave modes were effective in killing the insect species. The two microwave treatments that resulted in complete mortality of both *S. zeamais* and *R. dominica* species, with no progeny and no visible kernel damage were pulsed mode at 1.5 kW power level for 7 sec exposure time, and continuous mode at 2 kW power level for 4 sec exposure time. There were no differences between white and yellow maize types in terms of insect mortalities.

Table 4.1.5: Effect of microwave power level and exposure time on the mortalities of *C. ferrugineus* and *T. confusum* adults, when insects (no maize kernels) were microwave treated once (single exposure time) in the continuous and pulsed modes using the pulley system, with a modified 483 mm long microwave cavity, at 2450 MHz frequency

Insect species	Microwave mode & power level	Exposure time (sec)	Mortality #*† (%)
<i>C. ferrugineus</i>	Control	0s	0.0 ± 0.0
	Pulsed, 1.0 kW	12s	100.0 ^b ± 0.0
	Pulsed, 1.5 kW	9s	100.0 ^b ± 0.0
	Pulsed, 1.5 kW	12s	100.0 ^b ± 0.0
	Pulsed, 2.0 kW	4s	89.0 ^a ± 8.5
	Pulsed, 2.0 kW	6s	97.7 ^{ab} ± 4.0
	Cont., 1.0 kW	9s	98.0 ^a ± 1.7
	Cont., 1.0 kW	12s	96.7 ^a ± 5.8
	Cont., 1.5 kW	7s	95.7 ^a ± 5.1
	Cont., 1.5 kW	9s	100.0 ^a ± 0.0
	Cont., 2.0 kW	6s	100.0 ^a ± 0.0
<i>T. confusum</i>	Control	0s	0.0 ± 0.0
	Pulsed, 1.0 kW	12s	100.0 ^a ± 0.0
	Pulsed, 1.5 kW	9s	100.0 ^a ± 0.0
	Pulsed, 1.5 kW	12s	100.0 ^a ± 0.0
	Pulsed, 2.0 kW	4s	97.7 ^a ± 4.0
	Pulsed, 2.0 kW	6s	100.0 ^a ± 0.0
	Cont., 1.0 kW	9s	100.0 ± 0.0
	Cont., 1.0 kW	12s	100.0 ± 0.0
	Cont., 1.5 kW	7s	100.0 ± 0.0
	Cont., 1.5 kW	9s	100.0 ± 0.0
	Cont., 2.0 kW	6s	100.0 ± 0.0

All treatments were done in triplicate. Average values of mortality were used.

Number of dead insects after treatment divided by the number of alive insects (mean and standard deviation).

† Values with different superscripts within a block are statistically significantly different ($p < 0.01$).

All power levels, exposure times in both pulsed and continuous modes gave 100% mortalities of *C. ferrugineus* and *T. confusum* species. The two lowest dosage microwave treatments that resulted in 100% mortality of both species were pulsed mode at 1 and 1.5 kW power levels, for 12 and 9 exposure times, respectively.

Table 4.1.6: Effect of microwave power level and exposure time on the mortalities of different insect species and their progeny, when insects in white maize kernels were microwave treated once (single exposure time) in the pulsed mode using the pulley system, with a modified 483 mm long microwave cavity, at 2450 MHz frequency

Insect Species	Microwave mode & power	Exposure time * (sec)	100% mortality Yes/ No	Progeny development # Yes/ No
<i>S. zeamais</i> - (controls)	None	None	No	Yes
<i>R. dominica</i> - (control)	None	None	No	Yes
<i>E. cautella</i> - (control)	None	None	No	Yes
<i>T. confusum</i> - (control)	None	None	No	Yes
<i>C. ferrugineus</i> – (control)	None	None	No	Yes
<i>S. zeamais</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>R. dominica</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>E. cautella</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>T. confusum</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>C. ferrugineus</i>	Pulsed, 1.0 kW	12s	Yes	Yes
<i>S. zeamais</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>R. dominica</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>E. cautella</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>T. confusum</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>C. ferrugineus</i>	Pulsed, 1.5 kW	9s	Yes	No

* All treatments were done in triplicate. Exposure times were once-off, no repetitions were performed.

Progeny: microwave treated maize kernels were incubated for 6 weeks to determine if eggs in kernels hatched into insects.

In the pulsed mode using the pulley system with a modified 483 mm long microwave cavity, the lowest dosage that resulted in complete mortality of *S. zeamais*, *R. dominica*, *E.cautella*, *T. confusum* and *C. ferrugineus* species infested in white maize, with no progeny was 1.5 kW power level, 9 sec exposure time.

Table 4.1.7: Effect of microwave power level and exposure time on the mortalities of different insect species and their progeny, when insects in yellow maize kernels were microwave treated once (single exposure time) in the pulsed mode using the pulley system, with a modified 483 mm long microwave cavity, at 2450 MHz frequency

Insect Species	Microwave mode & power	Exposure time* (sec)	100% mortality Yes/ No	Progeny development # Yes/ No
<i>S. zeamais</i> - (controls)	None	None	No	Yes
<i>R. dominica</i> - (control)	None	None	No	Yes
<i>E. cautella</i> - (control)	None	None	No	Yes
<i>T. confusum</i> - (control)	None	None	No	Yes
<i>C. ferrugineus</i> – (control)	None	None	No	Yes
<i>S. zeamais</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>R. dominica</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>E. cautella</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>T. confusum</i>	Pulsed, 1.0 kW	12s	Yes	No
<i>C. ferrugineus</i>	Pulsed, 1.0 kW	12s	No	nd
<i>S. zeamais</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>R. dominica</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>E. cautella</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>T. confusum</i>	Pulsed, 1.5 kW	9s	Yes	No
<i>C. ferrugineus</i>	Pulsed, 1.5kW	9s	Yes	No

* All treatments were done in triplicate. Exposure times were once-off, no repetitions were performed.

Progeny: microwave treated maize kernels were incubated for 6 weeks to determine if eggs in kernels hatched into insects.

nd = Progeny not determined for samples which had live insects after treatment, except controls, as the objective was to determine whether insect eggs would hatch into insects after treatment.

In the pulsed mode using the pulley system with a modified 483 mm long microwave cavity, the lowest dosage that resulted in complete mortality of *S. zeamais*, *R. dominica*, *E.cautella*, *T. confusum* and *C. ferrugineus* species infested in yellow maize, with no progeny was 1.5 kW power level, 9 sec exposure time.

4.1.3 DISCUSSION

In the initial stage of the project, the idea was to use a free falling system in order to have a simple system that would be practical and easily implemented at pilot or production scales. However, the use of the free fall system required insects to be passed through the microwaves multiple times to obtain mortality. This was attributed to the short exposure times which were ineffective in the killing of insects. No literature could be found that used this type of system. It was impossible to increase the exposure time on this system. Therefore the system was not used further in the research study. The pulley system was then investigated where longer exposure times (single passes) achieved 100% insect mortality. This finding was in agreement with that of Kirkpatrick and Roberts (1971) who achieved 100% mortality of *R. dominica* only with long exposure times of 20 sec. As the pulley system was found to be appropriate it was implemented for the remainder of the research study.

Even though complete mortality of insects could be achieved, kernel damage (Tables 4.1.2 and 4.1.3) was also observed with the use of pulley system with the 728 mm microwave cavity. Kernel damage occurred as popping, swelling and discolouration (Figure 4.1.4). The damage was attributed to long exposure times at high power levels, causing local overheating. Various authors (Webber et al 1946; Nelson 1972; Mishenko et al 2000) observed that with microwave heating at higher power intensities and longer exposure times, an electric arc occurred between kernels, thus charring the grain. To minimize kernel damage, the exposure times were reduced. This was achieved by redesigning the microwave cavity to make it shorter by removing the middle section of the wave guide.

The successful treatments with 100% mortality, no progeny and no kernel damage in Tables 4.1.2 and 4.1.3 (unmodified, 728 mm long cavity) were used as the baseline for the 483 mm long cavity studies. The four treatments were repeated with the shorter, modified cavity, except that the exposure times were reduced. With the modified cavity, out of the four evaluated treatments, only two treatments were effective in the

eradication of *S. zeamais* and *R. dominica*. These treatments were then evaluated with three additional species *E. cautella*, *T. confusum* and *C. ferrugineus*. The two treatments (pulsed 1.5 kW for 7 sec, and continuous 2 kW for 4) were found to be ineffective in killing of *T. confusum* and *C. ferrugineus* species but effective on *E. cautella* (data not shown). As *R. dominica* is known to be one of the heat resistant species (Fields 1992; Beckett and Morton 2003), it was expected that *T. confusum* and *C. ferrugineus* would be completely eradicated by the two treatments. Based on these results, more microwave treatments with longer exposure times were studied (Table 4.1.5). Only two microwave treatments (pulsed mode) resulted in 100% mortality of *T. confusum* and *C. ferrugineus* species. When looking at all the treatments investigated, *T. confusum* adults had higher mortalities as compared to *C. ferrugineus* adults. This was attributed to the larger size of *T. confusum* (2.6-4.4 mm) compared to *C. ferrugineus* (1.8-2.2 mm). The larger size of *T. confusum* presumably allowed better absorption of microwaves and also promoted better heat transfer from kernels to insects. Shayesteh and Barthakur (1996) observed that insect size played a role in the killing of *T. confusum* and *Plodia interpunctella*. The former which was larger in size than the latter, had higher mortalities.

Pulsed microwave mode was found to be more effective than continuous mode in the treatment of insects (Table 4.1.5). The two microwave treatments that resulted in 100% mortality of both *C. ferrugineus* and *T. confusum* species were pulsed mode at 1 and 1.5 kW power levels, with 12 and 9 sec exposure times, respectively. Presumably as a result of the higher power delivered during the pulses, higher mortalities were achieved in the pulsed mode. In continuous mode 100% duty cycle was applied, while in the pulsed mode only 25% duty cycle. The 25% duty cycle allowed maximum power delivered during the pulses to be four times higher than that of continuous, while the average power applied remained the same as that used in the continuous mode. The findings of this work agree with other published work where it was found that pulsed mode was also more effective than the continuous mode for the drying of grain (Gunasekaran 1990; Shivhare et al 1992; Chau et al 2003).

The two successful microwave treatments (pulsed mode) were then evaluated on all five insect species (Tables 4.1.6 and 4.1.7). The observed survival of *C. ferrugineus* at 1 kW power dosage at 12 sec exposure time can be attributed to its small size even though the exposure time was long although the power level was low. The treatment at 1.5 kW power level for 9 sec exposure time gave complete mortality of *C. ferrugineus* and all other tested insect species.

4.1.4 CONCLUSIONS

Microwave treatment can achieve 100% mortality of five insect species that infest maize. Pulsed mode, 1.5 kW power level at 9 sec exposure time using a 483 mm long cavity (pulley system) can be used. The treatment is effective because insect mortalities are achieved with a single exposure with no visible kernel damage. Thus the use of microwave has potential to be used as an insect control measure for stored maize.

4.1.5 REFERENCES

Baker, V. H., Wiant, D. E. and Taboada, O. 1956. Some effects of microwaves on certain insects which infest wheat and flour. *J. Econ. Entomol.* 49:33-37.

Beckett, S. J., and Morton, R. 2003. Mortality of *Rhyzopertha dominica* (F.) (Coleoptera:Bostrychidae) at grain temperatures ranging from 50°C to 60°C obtained at different rates of heating in a spouted bed. *J. Stored Prod. Res.* 39:313–332.

Chau, K. J., Mujumdar, A. S., and Chou, S. K. 2003. Intermittent drying of bioproducts – an overview. *Bioresour. Technol.* 90:285-295.

Fields, P. G., 1992. The control of stored product insects and mites with extreme temperatures. *J. Stored Prod. Res.* 28:89-118.

Food and Agriculture Organization, 2007. Insect pests of stored grains in hot climates. <http://www.fao.org/docrep/T1838E/T1838E1f.htm>. (website accessed_October 2007).

Gunasekaran, S. 1990. Grain drying using continuous and pulsed microwave energy. *Dry. Technol.* 8:1039-1047.

Halverson, S. L., Burkholder, W. E., Bigelow, T. S., Nordheim, E. V., and Misenheimer, M. E. 1996. High - power microwave radiation as an alternative insect control method for stored products. *Stored Prod. Entomol.* 89:1638-1648.

Hamid, M. A. K., and Boulanger R. J. 1970. Control of moisture and insect infestation by microwave power. *Milling* 152:25-32.

Jood, S., and Kapoor, A. C. 1992. Effect of storage and insect infestation on protein and starch digestibility of cereal grains. *Food Chem.* 44:209-212.

Jood, S, Kapoor, A. C. and Singh, R. 1992a. Biological evaluation of protein quality of maize as affected by insect infestation. *J. Agric. Food Chem.* 40:2439–2442.

Jood, S., Kapoor, A. C. and Singh, R. 1992b. Mineral content of cereal grains as affected by storage and insect infestation. *J. Stored Prod. Res.* 28:147-152.

Kent, N. L., and Evers, A. D. 1994. Cereal crops: economics, statistics and uses. Pages 1-28 in: *Kent's Technology of Cereals*, 4th ed. Elsevier Science: Oxford.

Kirkpatrick, R. L., and Roberts, J. R. Jr. 1971. Insect control in wheat by use of microwave energy. *J. Econ. Entomol.* 64:950-951.

Mishenko, A. A., Malinin, O. A., Rashkovan, V. M., Basteev, V. A., Bazyma, L. A., Mazalov, Yu P. and Kutovoy, V. A. 2000. Complex high - frequency technology for protection of grain against pests. *J. Microwave Power Electromagn. Energy* 35:179-184.

Nelson, S. O. 1972. Insect – control studies with microwaves and other radiofrequency energy. *Bull. Entomol. Soc. Am.* 19:157-163.

Nelson, S.O. 1996. Review and assessment of radio-frequency and microwave energy for stored-grain insect control. *Trans. ASAE.* 39:1475-1484.

Rashkovan, V. M., Khizhnyak, N. A., Basteev, A. V., Bazyma, L. A., Niño de Rivera, L. and Ponomaryova, I. A. 2003. Interaction of electromagnetic waves with granular agricultural product and insects. *J. Microwave Power Electromagn. Energy* 38:225-234.

Shayesteh, N., and Barthakur, N. N. 1996. Mortality and behavior of two stored-product insect species during microwave irradiation. *J. Stored Prod. Res.* 32:239-246.

Shivhare, U. K. 1992. Drying characteristics of corn in a microwave field in a microwave field with a surface-wave applicator (Ph.D. Thesis summary). *Dry. Technol.* 10:1323-1326.

Warchalewski, J. R., Pra dzynska, A., Gralik, J. and Nawrot, J. 2000. The effect of gamma and microwave irradiation of wheat grain on development parameters of some stored grain pests. *Nahrung* 44:411-414.

Watters F. L. 1976. Microwave radiation for control of *Tribolium confusum* in wheat and flour. *J. Stored Prod. Res.* 12:19-25.

Webber, H. H., Wagner, R. P. and Pearson, A. G. 1946. High frequency electrical field as lethal agents for insects. *J. Econ Entomol.* 39:487-498.

White, N. D. G. 1995. Insects, mites, and insecticides in stored-grain ecosystem. Pages 123-167 in: *Stored-Grain Ecosystems*. D. S Jayas, N. D. G. White and W. E. Muir, eds. Marcel Dekker: New York.

4.2 EFFECT OF MICROWAVE ENERGY ON MAIZE KERNEL PHYSICOCHEMICAL PROPERTIES

ABSTRACT

Two microwave treatments, one at lower power dosage and shorter exposure times (normal) and the other at higher power dosage and longer exposure times (harsh) were used to determine the effect of microwave treatment for insect disinfestation on maize kernel physicochemical properties. White and yellow maize kernels were used for the study. The physical properties investigated included test weight, moisture content, 100 kernel weight, germinability, hardness, and stress cracks. Normal microwave treatment (pulsed mode, 1.5 kW at 9 sec exposure time) only had negative effects on moisture content and 100 kernel weight but no effect on other physical properties. The harsh microwave treatment (pulsed mode, 2.0 kW at 18 sec exposure time) had severe negative effects on all the above mentioned parameters. In addition, it (harsh treatment) affected the extractability of maize proteins and is not recommended as a grain treatment.

Keywords: maize quality, physicochemical properties, microwave energy, insect disinfestation

4.2.1 INTRODUCTION

Maize is the third most important cereal grain after wheat and rice (FAO 2002). It is one of South Africa's most important agricultural products, being used as a staple food by millions of people. Maize contributes approximately 42% to the gross value of field crops (South African Department of Agriculture 2004).

Maize grain quality is one of the major factors that has an influence on the quality of the final milled products. According to Pomeranz et al (1986b) the kernel physical properties that affect the yield and quality of final products are test weight, kernel weight, kernel hardness, breakage susceptibility and water absorptivity. Dry millers and snack food processors seek hard endosperm maize for large flaking grits (samp) (Stroshine et al 1986; Paulsen et al 2003), while wet millers require maize not to be dried at elevated temperatures, because it negatively affects starch recovery (Haros et al 2003; Paulsen et al 2003).

Drying of maize kernels at elevated temperatures (Brekke et al 1973; Brown et al 1979; Peplinski et al 1994; Haros et al 2003), as well as insect infestation (Jood et al 1992a, 1992b; Jood and Kapoor 1992) of kernels negatively affect maize quality. For example, drying of maize kernels at high temperatures results in low flaking grit yield during dry milling and separation of starch and proteins during wet milling becomes more difficult (Peplinski et al 1994).

Generally fumigation is used to control insect infestation of stored maize. But the disadvantages of the use of chemical pesticides which include the insects' ability to build up resistance (Kent and Evers 1994), and the phasing out of methyl bromide (Bell, 2000), have led to research on the use of microwave energy as a non-chemical, insect control measure (Nelson 1996).

Microwave energy may be effective in the killing of insects, as was shown in the previous research chapter but little is known on its effect on the physicochemical quality of maize. Velu et al (2006) studied dry milling characteristics of microwaved maize.

They found that microwave dried maize consumed less grinding energy during milling, but the drying had an effect on the structure of the protein and starch which resulted in lower paste viscosities.

The primary objective of this study was to determine the effect of microwave energy on maize kernel physicochemical properties.

4.2.2 MATERIALS AND METHODS

4.2.2.1 Maize samples

Yellow maize kernels supplied by Tongaat Hulett Starch, (Johannesburg, South Africa) and white maize kernels by Ruto Mills (Johannesburg, South Africa) were used in the study. Maize kernels at moisture content between 12-13% were stored at 4°C prior to the study.

4.2.2.2 Microwave unit

A microwave unit capable of being operated either on continuous or pulsed mode was utilized. As in the previous chapter, a 2 kW source Power Generator PM740, (Alter Power Systems, Italy) operated at a frequency of 2450 MHz was used to generate the microwave field. The unit had a 483 mm long cavity where an outer, stationary 22 mm ID, 1010 mm long quartz tube was positioned. In pulsed mode, a duty cycle of 25% was used, so that a maximum power was delivered four times during the pulses in order to obtain 100% power.

4.2.2.3 Microwave treatment

Microwave kernels were treated using the pulley system as described in the previous chapter. Four kilograms each of yellow and white maize kernels were microwave treated. Prior to the microwave treatment, maize kernels were equilibrated to a moisture content of 14%. The maize kernels were placed into the internal quartz tube, [19 mm

(ID), 498 mm length]. The tube was plugged at the bottom with glass wool. A nylon string, attached to the top end of the tube, and the other end of the string attached to a spindle driven by an electric motor allowed the maize kernels to be pulled at a constant rate through the external quartz tube. Maize kernels were exposed to microwaves only once (single pass).

Two microwave treatment conditions were investigated. The normal treatment (pulsed mode, 1.5 kW power level, 9 sec exposure time) caused no visible physical damage to maize kernels. The harsh treatment (pulsed mode, 2.0 kW power level, 18 sec exposure time) resulted in visible physical damage of kernels. Controls were equilibrated to 14% moisture content and not microwaved.

4.2.2.4 Temperature measurements

Temperature measurements were made by placing a thermocouple at different positions in the inner tube (2, 20 and 40 cm from the bottom of the tube) during microwave treatments.

4.2.2.5 Chemical analyses

Proximate analyses were performed by the Southern African Grain Laboratory (SAGL), Pretoria, South Africa. The analyses were, for crude fat (AACC 30-25), crude fibre (AACC 32-10), moisture content two-stage oven method (AACC 44-15A), ash (AACC 08-02), crude protein (AACC 46-30) (AACC International 2000), and total starch (ICC Std No 123/1). Analyses were done in duplicate except moisture content which was in triplicate.

4.2.2.6 Physical analyses

Maize kernels (100 g) were examined for single, double and multiple stress cracks over a light box. A stress crack index was calculated following a method described by Kirleis and Stroshine (1990). Test weight (hectolitre mass) was determined following the AACC method 55-10 (AACC 2000). Kernel weight was determined for 100 whole kernels with

no visual cracks or any other mechanical damages. Translucency was measured using a non-destructive image analysis technique described by Erasmus and Taylor (2004). Stenvert hardness measurements were determined as described by Pomeranz et al (1985). The physical tests were replicated three or more times for each sample.

4.2.2.7 Germinability

This was determined by a method developed by CSIR and Monsanto Seed Company. Two sheets of brown paper (31 X 55cm), (Agricol, Cape Town, South Africa) were placed at the bottom and 3 layers of cellulose wadding paper (31X 55 cm), (Agricol) on top of the sheets. A third sheet of brown paper was placed on top of wadding paper. The papers were moistened with water to generate suitable conditions for germination. Maize kernels (100) were placed on top of the brown paper with the kernels' germs facing upwards. A fourth wet sheet of brown paper was used to cover the kernels. All the layers of paper were rolled up into a roll and the ends were fastened with elastic bands. The rolls were placed in a plastic bag to retain moisture and incubated at 27°C for 4-5 days. Germination tests were done in triplicate.

4.2.2.8 2-D Gel Electrophoresis

Only protein samples of white maize control and harsh treated maize were separated on gels. White maize kernels (3-4) were ground into fine powder in liquid nitrogen with mortar and pestle. Proteins were extracted by direct resolubilization of the fine powder (30 mg) in 1000 µl lysis buffer (9M urea; 4% (w/v)

3-[(3-Cholamidopropyl)dimethylammonio]-2-hydroxy-1-propanesulfonate CHAPS; 1% (w/v) dithiothreitol (DTT); 0.8% (w/v) pharmalyte (pH 3-10) and 0.002% (w/v) bromophenol blue). The sample was sonicated at room temperature for 30 min, then centrifuged at 15 000 x g at 10°C for 15 min. The supernatant was removed and stored at 4°C. Protein concentration was determined by the method of Bradford (1976).

Isoelectric focusing (IEF) of soluble proteins was performed in the first dimension using 24 cm Immobiline Drystrip (IPG strip) (Amersham Biosciences, United Kingdom) with a

linear pH gradient of 3-10. The optimized final protein concentration used for electrophoresis was 60 μg . The protein sample was suspended in a rehydration buffer (8M urea, 0.5 % (w/v) CHAPS, 0.28 % (w/v) DTT, 0.5 % (v/v) IPG buffer pH 3-10 and 0.002% Bromophenol blue), and the total volume of sample and buffer was 450 μl which was pipetted onto an strip holder. The IPG strip (24 cm in length; pH 3-10) was placed in the protein solution, gel facing downwards. 2-3 ml of the IPG drystrip cover fluid (Amersham Biosciences) was overlaid to cover the strip to minimize evaporation of protein solution before the transparent lid was placed. The IEF was performed using an Ettan IPGphor II system (Amersham Biosciences) with the surface of the IPGphor maintained at 20°C. The first step of IEF process was the active rehydration of IPG strips which was carried out for 15 hrs at 0 V, after which the initial IEF of proteins was started using the step and hold gradients of 500 V for 1hr, 1000 V for 1 hr and finally 8000 V for 8 hr for 20 min. The focused strips were run immediately on a 2D gel electrophoresis following the method described by Natarajan et al (2005). The electrophoresis was carried out at room temperature on the Ettan DALT*twelve* system (Amersham Biosciences, United Kingdom). Image acquisition of the gels was done using the GS-800 densitometer (Biorad, United States) using PDQuest software. The resulting two gels protein spots were matched manually.

4.2.2.9 Statistical analysis

Analysis of data was performed using Statgraphics Centurion, Version XV (Statpoint Incorporated, Herndon, Virginia, USA). Multifactor analysis of variance (ANOVA) was applied using the Least significant difference test. All data were considered significant at $p < 0.01$.

4.2.3 RESULTS

Microwave heating was found not to be uniform throughout the quartz tube, as the temperature readings along different points of the tube varied for both the normal and harsh treatments. The temperature reached by the maize kernels was 4 to 7°C higher at the bottom of the tube. Also, the temperature in the harsh treatment was found to be some 30% higher than in the normal treatment.

Table 4.2.1: Maximum measured temperatures of maize kernels at three different points along the quartz tube during the normal and harsh microwave treatments

Position of probe (cm) ^	Temperature (°C) #	
	Normal treatment * (white & yellow maize)	Harsh treatment ** (white & yellow maize)
2	41.5 ^a ± 0.5	57.0 ^a ± 2.5
20	43.7 ^a ± 1.6	61.4 ^a ± 2.6
40	48.0 ^b ± 1.7	68.4 ^b ± 2.2

Measurements were done in triplicate

values with different superscripts in the same cells are statistically significantly different (p<0.01)

^ distance of the probe from the bottom of the quartz tube

* normal treatment = (pulsed mode, at 1.5 kW, 9 sec exposure time)

** harsh treatment = (pulsed mode, at 2 kW, 18 sec exposure time)

Table 4.2.2: Effect of normal and harsh microwave treatments on the physical properties of yellow and white maize kernels

Physical/ Chemical property	Microwave treatment	Yellow # maize	White # maize
Moisture content (% wb)	control	14.1 ^c ± 0.3	14.3 ^c ± 0.5
	normal *	12.7 ^b ± 0.2	12.2 ^b ± 0.6
	harsh**	12.2 ^a ± 0.2	11.8 ^a ± 0.3
100 kernel weight (g)	control	49.4 ^b ± 0.7	41.7 ^c ± 0.7
	normal	46.7 ^a ± 0.2	40.6 ^b ± 0.3
	harsh	47.6 ^a ± 0.4	39.0 ^a ± 0.4
Test weight (kg/hl)	control	77.5 ^b ± 0.4	82.9 ^b ± 0.7
	normal	77.5 ^b ± 0.3	82.0 ^b ± 0.5
	harsh	75.6 ^a ± 0.3	80.8 ^a ± 0.1
SCI (Stress Cracks Index)	control	32.0 ^a ± 4.4	14.8 ^a ± 7.5
	normal	27.3 ^a ± 3.3	12.8 ^a ± 2.5
	harsh	42.7 ^b ± 2.3	28.3 ^b ± 3.9
Germination (%)	control	95.5 ^b ± 3.5	100.0 ^b ± 0.0
	normal	97.2 ^b ± 1.0	100.0 ^b ± 0.0
	harsh	3.4 ^a ± 2.3	20.7 ^a ± 0.7
Translucency (%) ^ (whole kernel)	control	18.7 ^a ± 3.1	37.4 ^a ± 0.5
	normal	15.1 ^a ± 1.3	33.8 ^a ± 2.0
	harsh	5.7 ^b ± 0.8	24.2 ^b ± 3.4
Translucency (%) ^^ (endosperm)	control	29.0 ^a ± 5.7	53.4 ^a ± 1.9
	normal	23.6 ^a ± 1.6	46.5 ^a ± 2.4
	harsh	8.9 ^b ± 1.2	35.0 ^b ± 4.4

All treatments were done in triplicate.

values with different superscripts in the same cells are statistically significantly different (p<0.01)

* normal treatment = (pulsed mode, at 1.5 kW, 9 sec exposure time)

** harsh treatment = (pulsed mode, at 2 kW, 18 sec exposure time)

^ translucency = translucency calculated on whole maize kernel area

^^ translucency = translucency calculated on maize kernel endosperm area only

Both normal and harsh microwave treatments decreased the moisture content and 100 kernel weight of the white and yellow maize kernels. The harsh treatment decreased test weight, severely decreased germinability, and translucency. The harsh treatment also increased the amount of stress cracks in the maize kernels.

Table 4.2.3: Effect of normal and harsh microwave treatments on the chemical properties of yellow and white maize kernels

Chemical Property	Microwave treatment	Yellow # maize	White # maize
Starch content (% db) †	control	75.6 ^a ± 0.1	76.4 ^a ± 0.1
	normal*	78.7 ^b ± 0.0	78.8 ^c ± 0.0
	harsh**	75.5 ^a ± 0.1	77.9 ^b ± 0.2
Fat (% db)	control	4.2 ^a ± 0.0	3.7 ^b ± 0.0
	normal	4.3 ^b ± 0.0	3.5 ^a ± 0.0
	harsh	4.3 ^b ± 0.0	3.5 ^a ± 0.1
Protein (% db)	control	8.6 ^a ± 0.1	8.6 ^c ± 0.0
	normal	8.8 ^a ± 0.0	8.0 ^a ± 0.0
	harsh	8.8 ^a ± 0.1	8.2 ^b ± 0.0
Crude fibre (% db)	control	1.2 ^a ± 0.0	1.1 ^a ± 0.0
	normal	1.5 ^b ± 0.0	1.4 ^b ± 0.0
	harsh	2.1 ^c ± 0.1	1.8 ^c ± 0.1
Ash (% db)	control	1.1 ^a ± 0.0	1.1 ^a ± 0.0
	normal	1.2 ^b ± 0.0	1.1 ^a ± 0.0
	harsh	1.2 ^b ± 0.0	1.2 ^b ± 0.0

All treatments were done in triplicate.

* values with different superscripts in the same cells are statistically significantly different (p<0.01)

† db = dry basis

* normal treatment = (pulsed mode, at 1.5 kW, 9 sec exposure time)

** harsh treatment = (pulsed mode, at 2 kW, 18 sec exposure time)

The microwave treatments apparently gave significant increases in starch, crude fibre and ash, increases and decreases in fat (depending on kernel type), and a decrease in protein.

Table 4.2.4: Effect of normal and harsh microwave treatments on the hardness of yellow and white maize kernels as determined by the Stenvert Hardness Tester

Parameter	Sample	Yellow maize [#]	White maize [#]
Resistance time (s)	control	96.3 ^a ± 6.7	103.0 ^a ± 1.0
	normal [*]	108.0 ^b ± 2.0	127.0 ^b ± 4.8
	harsh ^{**}	109.0 ^{ab} ± 7.8	129.5 ^b ± 5.7
C/F ratio [†]	control	1.8 ^a ± 0.2	1.4 ^a ± 0.1
	normal	1.4 ^a ± 0.0	1.3 ^a ± 0.0
	harsh	1.5 ^a ± 0.1	1.4 ^a ± 0.2
Column height (cm) [^]	control	4.3 ^a ± 0.1	4.2 ^a ± 0.1
	normal	4.5 ^a ± 0.1	4.5 ^b ± 0.1
	harsh	4.5 ^a ± 0.1	4.4 ^{ab} ± 0.1

All treatments were done in triplicate.

[#] values with different superscripts in the same cells are statistically significantly different (p<0.01)

[†] Coarse/ fine ratio by weight

^{*} normal treatment = (pulsed mode, at 1.5 kW, 9 sec exposure time)

^{**} harsh treatment = (pulsed mode, at 2 kW, 18 sec exposure time)

[^] the total column height of freshly ground maize (before sieving into fine and coarse particles)

Both microwave treatments caused a significant (p<0.01) increase in the hardness (resistance time) of white maize kernels. Yellow maize showed an indication of an increase in hardness.

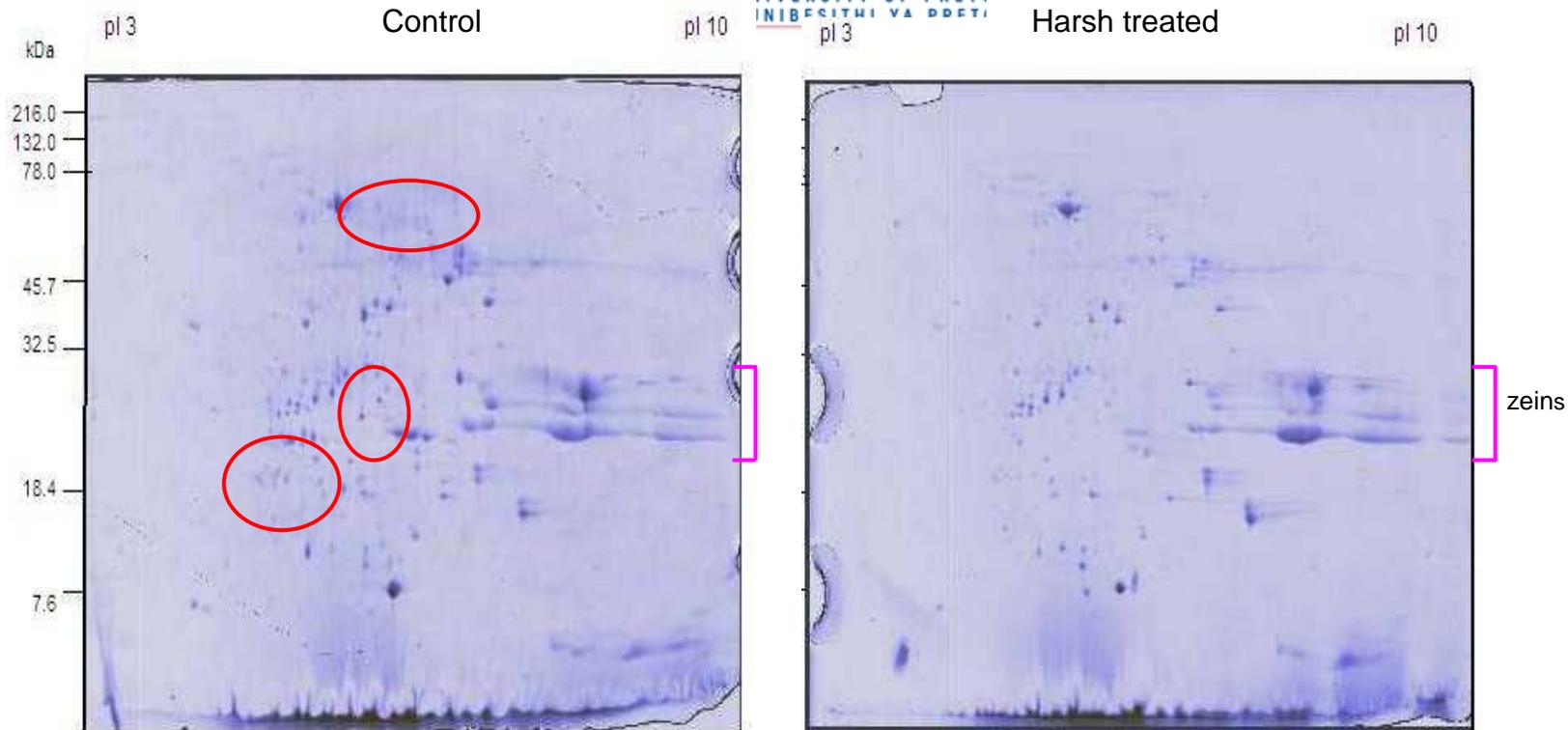


Figure 4.2.1: Effect of harsh microwave treatment on the extractability of total proteins of white maize kernels separated by 2D-PAGE. The circles indicate regions of proteins spots that were present in control maize kernels but not present in harsh treated maize kernels.

Harsh microwave treatment had a negative effect on the extractability of proteins in the maize kernel (Figures 4.2.1 and 4.2.2). The zein proteins were, however, not affected.

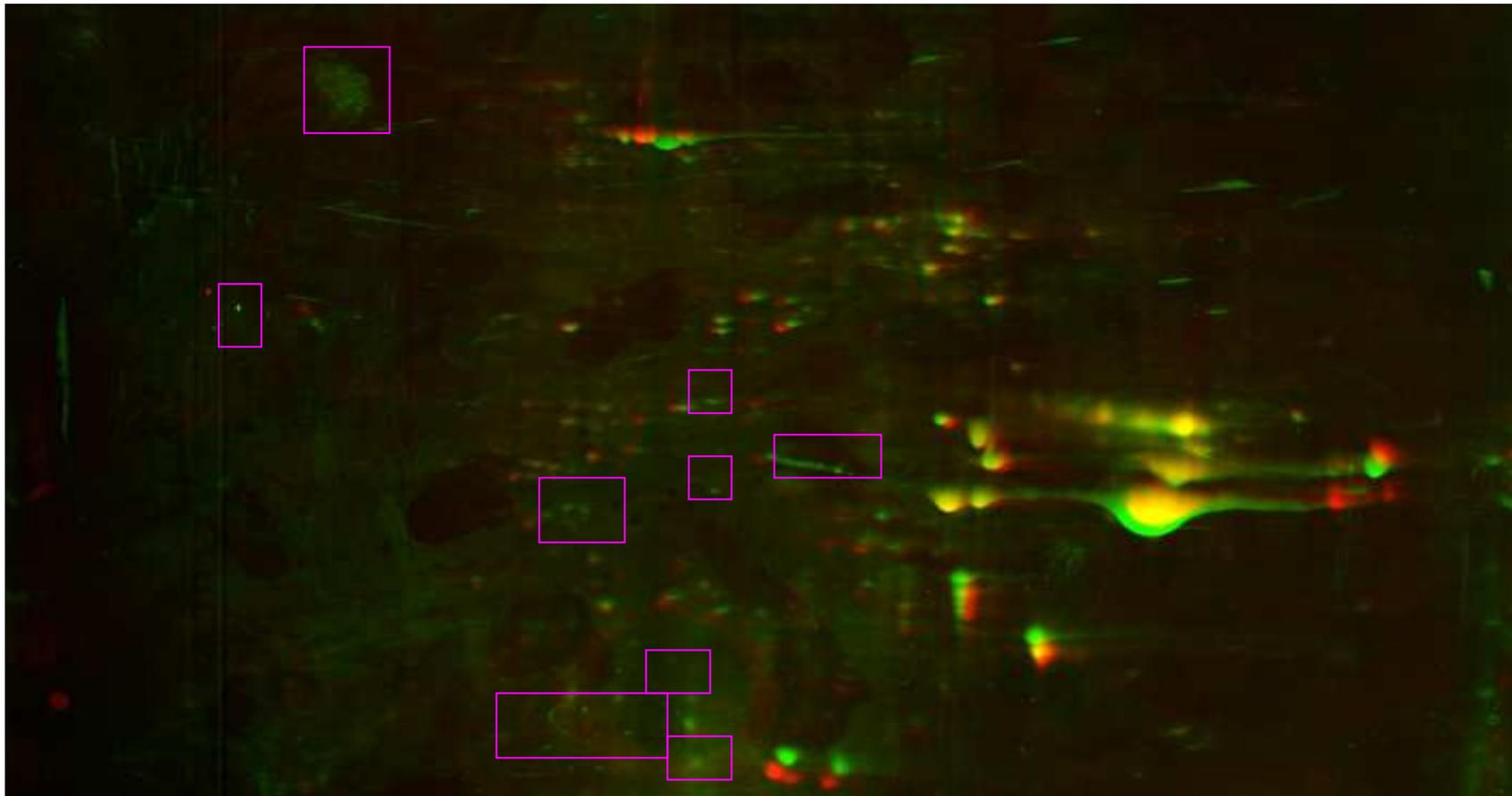


Figure 4.2.2: A superimposed image of control and harsh treated gels separated by 2D-PAGE. The control maize protein spots are represented by the green colour and harsh treatment maize protein spots by red. The yellow colour results from the accurate superimposed red (treated) and green (control) spots. The rectangles indicate regions of protein spots that were present in the extract from control maize kernels but not present in the extract from harsh treated maize kernels.

4.2.4 DISCUSSION

The higher temperature in the maize kernels during the harsh microwave treatment resulted in a change in maize kernel physicochemical properties. The adverse effect of high temperature agrees with the work of Peplinski et al (1994) who found that kernel physical characteristics decreased in quality when maize was dried at temperatures of 55°C and above.

The loss of water vapour during microwave heating was the reason for the decrease in moisture content, 100 kernel weight and test weight (Table 4.2.2). These results agree with those reported by Brown et al (1979) even though these authors used a crossflow drier not a microwave. The authors studied the effect of three drying methods (high-temperature batch drying, dryeration and low-temperature in-bin drying) on the quality of maize harvested at different moisture contents. They observed that the test weight of maize dried with both batch and dryeration methods decreased as the drying temperatures were increased from 45 to 60°C and higher (80 and 100°C).

The reason why there was an increase in stress cracks in the harsh treatment was because of the moisture gradients created in maize kernel during microwave heating. According to Paulsen et al (2003) and Kirleis and Stroshine (1990), when maize kernels are rapidly dried and cooled, stress cracks are formed due to the moisture gradients. What probably happened during the harsh microwave treatment was that, as water molecules were heated inside the kernel, the rate of moisture movement from the inside of kernel to the surface of the kernel was faster than the rate at which the water was being evaporated from the surface of the kernel. This caused moisture gradients within the kernel.

The reduction in maize kernel germinability was because of denaturation of the proteins responsible for seed viability. This is supported by the less extractable proteins observed by 2D PAGE (Figures 4.2.1 and 4.2.2). According to Pomeranz (1992), Roberts (1972) found that loss of seed viability can be caused by denaturation of essential metabolites. When Kirkpatrick and Roberts (1971)

microwave treated wheat at 2 kW for 10 sec exposure time, germination was reduced by 8%. The harsh microwave treatment reduced germination by 79% and

96% for white and yellow maize, respectively. The vast difference in germination between the current study (harsh treatment) and that of Kirkpatrick and Roberts (1971) can be attributed to the longer exposure times to microwaves which presumably affected the maize enzymes involved in germination.

The reason for the decrease in translucency was because of air pockets created between the starch granules and protein matrix. According to Duvick (1961), during drying, the thin protein matrix in the floury endosperm ruptures and causes minute air pockets around the starch granules. Due to light refraction by the air pockets, there is an opaque appearance.

Most of the increases and decreases in chemical contents (Table 4.2.3) can be attributed to random experimental variation. A probable reason for the large increase in crude fibre was because of cross linking of proteins with other substances such as starch and fibre or aggregation of proteins. This was supported by the observation that harsh microwave treated maize proteins were found to be less extractable (Figure 1 and 2). Sulphydryl groups in proteins or peptides can react with substances such as carbohydrates, when grain is dried at elevated temperatures (Wall et al 1975). The authors state that insolubility of proteins may also be caused by heat denaturation of proteins to form random structures that permit hydrogen bonding or hydrophobic functional groups to interact non-covalently and produce molecular aggregates. Drying of maize kernels at elevated temperatures can greatly reduce extractable water and saline soluble proteins (albumins and globulins). McGuire and Earle (1958) showed a decrease in water-extractable nitrogen at temperatures from 48.9 to 93.3°C, Wall et al. (1975) observed the changes from 60-143°C, Wight (1981) saw the changes at from 80 to 100°C, and Peplinski et al (1994) only observed the changes from 70 to 100°C. In this study the extractability of the zein proteins (maize prolamins) was not affected by microwave heating. This agrees with the findings of Peplinski et al (1994) and Wight (1979). These authors could not find notable changes on the extractability of zeins from maize dried at elevated temperatures. This can be attributed to the fact that zeins only have low levels of tryptophan and lysine (Lawton and Wilson 2003), which presumably make the proteins less susceptible to Maillard reaction.

Both microwave treatments caused an increase in the hardness (resistance time) of the maize kernels (Table 4.2.4). Considering the sharp increase in the amount of stress cracks on maize kernels for the harsh treatment, it was expected that the resistance time for the harsh treated maize would actually be shorter and not longer, as the kernels should break more easily during milling. However, Kirleis and Stroshine (1990) did not observe a change in hardness of maize dried at temperatures up to 60°C. A slight decrease in resistance time was only seen with drying at higher temperatures. Interestingly, when Pomeranz et al (1986a) increased the moisture content of maize kernels from 12 to 16% and dried them between 82-93°C, they observed an increase in the resistance time. The unexpected effect on resistance time has been linked to the type and amount of proteins, particularly γ -zeins in maize endosperm, which influence maize hardness (Mestres and Matencio 1996; Chandrashekar and Mazhar 1999).

4.2.5 CONCLUSIONS

The potential use of microwave energy as an insect infestation control measure for stored maize depends on the microwaving conditions. Both normal (pulsed mode at 1.5 kW power dosage, for 9 sec exposure time) and harsh (pulsed mode, 2 kW power dosage, for 18 sec exposure time) treatment conditions can eradicate adult insects and their eggs, but only the former maintains maize quality.

4.2.6 REFERENCES

AACC International, 2000. Approved Methods of the AACC, 10th Ed. AACC Method 08-02, Method 30-25, Method 32-10, Method 44-15A, Method 46-30, and Method 55-10. American Association of Cereal Chemists: St Paul, MN.

Bell, C. H. 2000. Fumigation in the 21st century. *Crop Prot.* 19:563-569.

Bradford, M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal. Biochem.* 72:248-254.

Brekke, O. L., Griffin, Jr., and Shove, G. C. 1973. Dry milling of corn artificially dried at various temperatures. *Trans. ASAE* 16:761-765.

Brown, R. B., Fulford, G. N., Daynard, T. B., Meiering A. G., and Otten, L. 1979. Effect of drying method on grain quality. *Cereal Chem.* 56:529-532.

Chandrashekar, A., and Mazhar, H. 1999. The biochemical basis and implications of grain strength in sorghum and maize. *J. Cereal Sci.* 30:193-207.

Duvick, D. N. 1961. Protein granules in maize endosperm cells. *Cereal Chem.* 38:374-385.

Erasmus, C., and Taylor, J. R. N. 2004. Optimising the determination of maize endosperm vitreousness by rapid non-destructive image analysis technique. *J. Sci. Food Agric.* 84:920-930.

Food and Agriculture Organization, 2002. Assessment of the world food security situation. Committee on world food security, 28th session, Rome. http://www.fao.org/docrep/meeting/004/y6441e/y6441e00.htm#P292_17440. (website accessed May 2007).

Haros, M., Tolaba M. P. and Suárez, C. 2003. Influence of corn drying on its quality for the wet milling process. *J. Food Eng.* 60:177-184.

ICC, 1994. ICC standard no 123/1. International Association for Cereal Science and Technology: Vienna.

Jood, S., and Kapoor, A. C. 1992. Effect of storage and insect infestation on protein and starch digestibility of cereal grains. *Food Chem.* 44:209-212.

Jood, S, Kapoor, A. C. and Singh, R. 1992a. Biological evaluation of protein quality of maize as affected by insect infestation. *J. Agric. Food Chem.* 40:2439-2442.

Jood, S., Kapoor, A. C. and Singh, R. 1992b. Mineral content of cereal grains as affected by storage and insect infestation. *J. Stored Prod. Res.* 28:147-152.

Kent, N. L., and Evers, A. D. 1994. Cereal crops: economics, statistics and uses. Pages 1-28 in: *Kent's Technology of Cereals*, 4th ed. Elsevier Science: Oxford.

Kirkpatrick, R. L., and Roberts, J. R. Jr. 1971. Insect control in wheat by use of microwave energy. *J. Econ. Entomol.* 64:950-951.

Kirleis, A. W., and Stroshine, R. L. 1990. Effects of hardness and drying air temperature on breakage susceptibility and dry milling characteristics of yellow dent corn. *Cereal Chem.* 67:523-528.

Lawton, J. W., and Wilson, C. M. 2003. Protein of the kernel. Pages 314-354 in: *Corn Chemistry and Technology*. P. J. White and L. A. Johnson, eds. American Association of Cereal Chemists: St Paul, MN.

McGuire, T. A., and Earle, F. R. 1958. Changes in the solubility of corn protein resulting from the artificial drying of high-moisture corn. *Cereal Chem.* 35:179-188.

Mestres, C., and Matencio, F. 1996. Biochemical basis of kernel milling characteristics and endosperm vitreousness of maize. *J. Cereal Sci.* 24:283-290.

Natarajan, S., Xu, C., Caperna, T. J. and Garrett, W. M. 2005. Comparison of protein solubilization methods suitable for proteomic analysis of soybean seed proteins. *Anal Biochem.* 342:214-220.

Nelson, S.O. 1996. Review and assessment of radio-frequency and microwave energy for stored-grain insect control. *Trans. ASAE.* 39:1475–1484.

Paulsen, M. R., Watson, S. A. and Singh, M. 2003. Measurement and maintenance of corn quality. Pages 1159-1219 in: *Corn: Chemistry and Technology.* P. J. White and L. A. Johnson, eds. American Association of Cereal Chemists: St. Paul.

Peplinski, A. J., Paulis, J. W, Bietz, J. A. and Pratt, R. C. 1994. Drying of high moisture corn: Changes in properties and physical quality. *Cereal Chem.* 7:129-133.

Pomeranz, Y. 1992. Biochemical, functional, and nutritive changes during storage. Pages 55-141 in: *Storage of cereal grains and their products.* D. B. Sauer, ed. American Association of Cereal Chemists: St Paul, MN.

Pomeranz, Y., Czuchajowska, Z. and Lai, F. S. 1986a. Comparison of methods for determination of hardness and breakage susceptibility of commercially dried corn. *Cereal Chem.* 63:39-43.

Pomeranz, Y., Hall, G. E., Czuchajowska, Z., Martin, C. R. and Lai, F.S. 1986b. Test weight, hardness, and breakage susceptibility of yellow dent corn hybrids. *Cereal Chem.* 63:349-351.

Pomeranz, Y., Czuchajowska, Z., Martin, C. R. and Lai, F. S. 1985. Determination of corn hardness by the Stenvert Hardness Tester. *Cereal Chem.* 62:108-112.

Roberts, E. H. 1972. Dormancy: A factor affecting seed survival in soil. Pages 321-359 in: *Viability of Seed.* E. H. Roberts, ed. Syracuse University Press: New York.

South African Department of Agriculture, 2004. Trends in the agricultural sector 2004. <http://www.nda.agric.za/docs/trends2004.pdf>. (website accessed February 2006).

Stroshine, R. L., Kirlies, A. W., Tuite, J. F., Bauman, L. F. and Emam, A. 1986. Differences in the grain quality among selected corn hybrids. *Cereal Foods World* 31:311-316.

Velu, V., Nagender, A., Rao, P. P. G. and Rao, D. G. 2006. Dry milling characteristics of microwave dried maize grains (*Zea mays L.*). *J. Food Eng.* 74:30-36.

Wall, J. S., James, C. and Donaldson, G. L. 1975. Corn proteins: Chemical and physical changes during drying of grain. *Cereal Chem.* 52:779-790.

Wight, A. W. 1979. Detection of heat damage in artificially fried maize. *South African Food Review* 6:63-70.

Wight, A. W. 1981. Changes in properties of some maize cultivars associated with artificial drying at elevated temperatures. *Starch* 33:165-168.

5. GENERAL DISCUSSION

The main objective of the study was to determine whether it was possible to eradicate storage insect pests in maize using microwave energy without any adverse effects on grain quality. The major finding of research chapter 1 was that microwave energy can eradicate adult insects and their eggs. To obtain 100% insect mortality, the relatively short time of 9 sec at 1.5 kW (pulsed mode) power dosage was required. Chapter 2 indicated that some of the microwave conditions that can eradicate insect pests did not adversely affect maize quality.

5.1 Experimental design

In the initial stages of the study, a free falling system was investigated as a system that would have been ideal for implementation at production scale. However, as the system was used, it became obvious that it would not be effective in the killing of the insects due to the fact that the exposure time was too short and insects had to be repeatedly treated in order to kill them. To increase the exposure time, one would need a longer microwave cavity which would require more working space in a maize mill. To estimate the required length of the microwave cavity if the free falling system was to be used, the distance fallen after a time of t seconds is given by the formula;

$$d = 0.5 * g * t^2$$

where g = the acceleration of force of gravity (9.8 m/s/s on Earth).

d = length of microwave cavity in metres

At $t = 9$ sec (exposure time)

$$d = (0.5) * (9.8 \text{ m/s}^2) * (9 \text{ s})^2 = 397 \text{ m}$$

Thus, the microwave cavity would require too much space. Therefore a free falling system would not be feasible at the current power levels.

One thing that was not executed well during the insect killing experiments, which would have yielded valuable data, was temperature measurements. The data would

have indicated thermal death temperatures of the individual insect species. In the initial stage of the project appropriate temperature probes for use with microwaves were investigated. The two commonly used probes to measure microwave temperature are infrared and thermocouple probes (Datta et al 2001). The use of an infrared probe would have required the microwave cavity to be modified, which meant extra costs. The cavity as well as the outside quartz tube would have been modified by creating openings to mount the probe. The use of the infrared probe would have been a challenge in that it would have required measuring the temperature of maize kernels, and not the air in the tube. Infrared probes are commonly used to measure the temperature of surfaces (Goedeken et al 1991). So the thermocouple was the ideal probe to use in the study as no cavity modifications were required and due to its low cost (Childs et al 2000). However, the project was on a tight time frame, and by the time it was decided on the thermocouple probe, the study on the killing of the insects was complete. Therefore, temperature was only measured during the maize quality study.

It would probably have been better to evaluate the killing of all 5 insect species at once instead of using two species (*S. zeamais* and *R. dominica*) as the baseline. Or investigate all species at ones but with less number of insects used per species. Though *R. dominica* is one of the most heat tolerant species (Fields 1992; Beckett and Morton 2003), *S. zeamais* is not the second most heat tolerant insect species among the five studied. Fields (1992) refers to Oostuizen (1935) that *R. dominica* and *T. confusum* are the most heat tolerant among the species tested. The selection of the *R. dominica* and *S. zeamais* to use in the study was made as a result of a recommendation by the ARC entomologists. Parallel experiments to kill all 5 species would have saved time and minimised the chances that two of the new species were not completely eradicated by the two treatments that were effective on *S. zeamais* and *R. dominica* (Table 4.1.4). To save time and considering the different biology of pests (internal vs external habitats) it would have been better if one each primary and secondary insect species were used for preliminary studies. However, there was no control of this because at the initial stages of the project, there was funding for only two insect species (*R. dominica* and *S. zeamais*). Funding to study the other three insect species was only awarded at a later stage, from a different funding body.

In this study, total mortality of five adult insect species was achieved within a very short exposure time (9 sec). This can be attributed to the high microwave power dosage (1.5 kW) and grain high moisture content. Baker et al (1956) microwave treated *T. confusum* adults in wheat at 0.94 kW at 2450 MHz. Total mortality was observed 1 week after treatment with an exposure time of 21 sec. Watters (1976) achieved 100% mortality of *T. confusum* in wheat at 0.03 kW at 105 sec exposure time at 8500 MHz frequency, only in wheat of high moisture content of 15.6%. Surprisingly, Shayesteh and Barthakur (1996) observed a decrease in *T. confusum* mortality when moisture content of wheat was increased from 6 or 9% to 12%. The authors proposed that the decrease in insect mortality was probably due to the protection offered by the higher amount of water. Higher moisture content of grain should actually work in favour of high insect mortalities not against, because at high moisture levels the heating rate is higher and insects get exposed to high temperatures (Nelson and Kantack 1966). This could be explained because *T. confusum* is a secondary insect. Because *T. confusum* is found outside not inside the kernel, the increase of kernel moisture content will not necessarily increase mortality of *T. confusum*.

5.2 Analytical Methodologies

The determination of maize kernel stress cracks using the light box method of Kirleis and Stroshine (1990) was found to be time-consuming and not very accurate. In the current study the standard deviations varied from 2.3 to 7.5 and the method was found to be subjected to operator error. It is recommended to use image analysis to detect kernel stress cracks in future. Gunasekaran et al (1987) and Yie et al (1993) used image analysis machines to detect stress cracks. The former authors developed an image processing algorithm and could detect 90% of the stress cracks, although kernels had to be carefully positioned to obtain usable images. The latter authors developed a high speed, machine image algorithm for on-line detection of stress cracks. The accuracies ranged from 83-98% and the processing time for each kernel was 2 seconds, even though the standard deviations ranged between 2.3 to 4.5.

It would also have been valuable if the proteins separated on 2D electrophoresis could have been identified. Two dimensional electrophoresis is one of the most important proteomics tools (Lauber et al 2001), and in order to quantify and identify the separated protein spots, one requires instrumentation such as the Matrix-Assisted Laser Desorption Ionization Time-of-Flight Mass Spectrometer (MALDI-TOF MS) (Pandey and Mann 2000; Mann et al 2001). Even though the MALDI-TOF MS was available for use during the course of the study, the running costs, proteomics course costs (background purposes) were not planned on the project, and this made it impractical to use the equipment to quantify the extracted maize proteins.

5.3 Way forward

If the microwave grain storage insect pest eradication process is going to be implemented at production scale, it will be advisable that maize kernels are microwave treated prior to silo entry as well as prior to maize product packaging. The treatment prior to silo storage will eradicate the insects, and therefore reduce the amount of damaged kernels caused by insect infestation, assuming no insects will enter silo after maize treatment. To microwave process maize kernels prior to silo storage will mean many tons more maize kernels per day will have to be processed as compared to if only maize products were microwave processed before packaging. The treatment of maize products prior to packaging will eradicate insect eggs that survive the milling process, or contaminate the product during the milling process. This will minimise the return of products from the retail stores to the mills.

To avoid re-infestation of maize products, it was indicated by Mr. Mike van Deventer (Production Manager, Godrich Flour Mill, 2007 - personal communication) retail stores also need to be disinfested on a regular base. There is no point if insect free products are stored on shelves of retail stores which are infested by insects from other cereal products. To avoid the re-infestation, products may need to be vacuum sealed in plastic that is insect resistant as most of these insects can penetrate the products' packaging material. However, this will increase the cost of packaging which will in turn increase the retail price of the products to consumers.

Ruto Mills (Pretoria, South Africa) is the 8th biggest wheat and maize mill in the world. According to Mr. Klaas Dumas (Mill Manager) and Mr. Jaco Venter (Silo Manager) it costs R0,76/ton (\$0.11, exchange rate of 7R/\$) when phosphine pellets are used to fumigate maize kernels in silos for grains that would be stored for 2-3 months.

To estimate the cost to microwave process maize products at production scale, one can extrapolate from laboratory scale data of 82 g of sample per 9 sec at 1.5 kW. If a mill processes 24 tons of maize kernels per hour, of which 72.5% equivalent to 17.4 tons is converted into different maize products for human consumption while the remaining 27.5% is for animal feed. Therefore to handle a 17.4 tons/hour stream of maize products would require a 796 kW microwave. Running costs would be about R204.70/hr at Eskom's (electricity utility) 2007/08 Miniflex tariffs (Eskom 2007), which translates to a treatment cost of R11.76/ton (approximately US\$ 1.66/ ton). The capital cost required to build a microwave unit also needs to be taken into consideration, as it will constitute the bulk of microwave treatment costs. Mr. Thys Rossouw of Delphius Technologies (personal communication) estimated a ballpark figure of \$1.35 million for a complete microwave unit (generator, cooling system and applicator). Even under optimistic conditions (20 year lifetime, zero maintenance cost, exchange rate of 7R/\$) this capital cost will add an additional \$0.44/ton. An estimated microwave treatment cost would be \$ 2.1/ton as compared to \$ 0.11/ton when chemical pesticides are applied. This calculation indicates that microwave treatment will cost at least 20 times as much as the current fumigation process.

Other authors e.g. Nelson (1996) have also found the cost of microwave or radiofrequency energy application for insect control purposes to be several times greater than that of chemical pesticides. Nelson and Whitney (1960) estimated the cost of radiofrequency treatment of wheat to control stored grain insects to be \$1.43/ton when wheat was heated from 27 to 66°C, using a 200 kW radio-frequency generator with a capacity of 9800 tons/hr for 2000 hr/year. Interestingly, Hamid and Boulanger (1970) estimated the cost to microwave wheat to control stored grain insects to be less than the cost of fumigation. The estimated cost was \$17.96 per ton when wheat was heated from 22 to 65°C, at a power of 1.27 kW. Before it can be decided if microwave technology should be used or not as an insect infestation control measure, the cost of pesticide use, the time it requires for the pesticide to be

effective, the number of times pesticide are applied in a year, the losses due to insect infestation should be taken into consideration.

Currently, pneumatic tube conveyors, which use low-pressure air are used to move agricultural products in mills. To design the conveyors, the properties of grains are studied by considering either bulk or individual units of the material (Güner 2007). It is important to have an accurate estimate of shape, size, volume, density, surface area and other characteristics which may be considered as engineering parameters for that product (Güner 2007). The pneumatic system is the ideal system to be used to move maize product streams for microwave treatment because the set-up and the construction of the tube conveyors allows little or no chance of maize product contamination. Microwave treatment of maize products should take place in quartz tube of a certain length, where the microwave unit will be positioned. After treatment, the maize products should be moved to storage bins using pneumatic conveyors. The material used to make the tube conveyors should be resistant to high temperatures as product temperature may be high after treatment. Probably cool air should be blown to lower the temperature of the microwave treated maize products.

Despite the fact that the economic aspects of applying microwave and radio-frequency energy for insect control purposes are discouraging, they are potentially viable due to the environmental and health hazards associated with chemical residual pesticides. To minimise the treatment costs, it is thus suggested that only a unit for the treatment of the maize products should be evaluated (Figure 5.1). The maize milling process which comprise of the cleaning and conditioning of maize kernels, sieving and milling of maize fractions, will not be changed. Prior to packaging or bin storage, maize meal should be moved using pneumatic conveyors to be microwave treated. The reason being, that it is better to treat maize products as they have higher monetary returns that maize kernels.

The use of microwave technology for insect pest eradication is a field that can be explored further. Other research opportunities/ gaps that should be explored include:

- Effects of pulsed microwave disinfection on the maize products' nutritional quality (starch and protein digestibility, mineral and vitamins bioavailability).
- Heat treatment can have a negative effect on food nutrients. According to Slavin et al (2000) heat processing may cause the development of resistant starch which can be resistant to digestion and absorption in the small intestine. Ideally the use of microwave technology should have little or no effect on the maize products' nutritional properties.

- Effects of pulsed microwave processing on heat transfer phenomena in maize products. If energy transfer phenomena can be understood better, then it will be easier to improve the system to be energy efficient. This might lower the running costs in terms of lower power usage and also reduce the treatment exposure times.

5.4 Challenges

Some of the challenges that are going to be encountered will be the design of the microwave unit itself. The cavity design, in terms of dimensions should be able to achieve the required field intensity and exposure times. The microwave field inside the cavity should be uniform to ensure the insects receive microwave exposure and do not escape during treatment. Another challenge is to optimise energy transfer to the grain product.

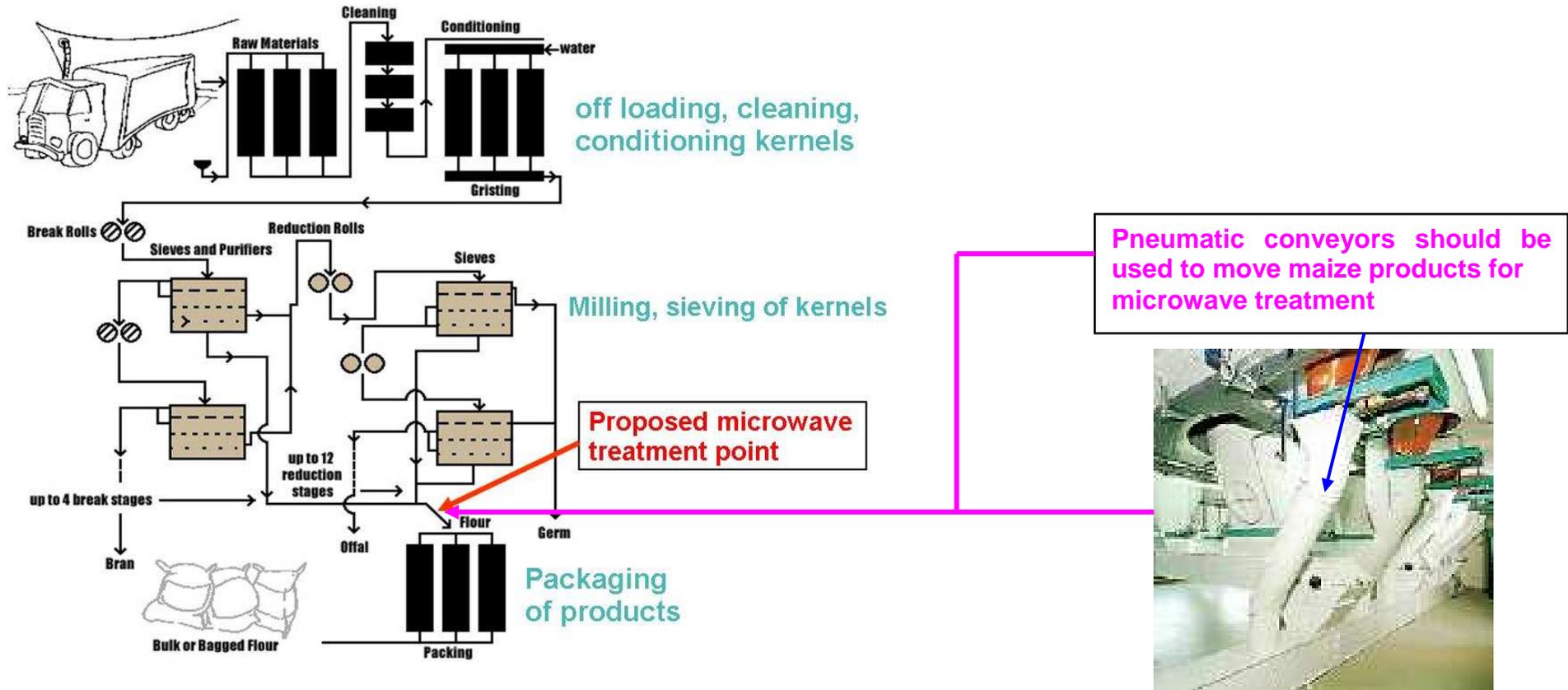


Figure 5.1: Proposed system for microwave treatment of maize products to control storage insect pests during the maize milling process.

6. CONCLUSIONS

The objective of the research was to determine if microwave energy can eradicate storage insect pests without damaging maize quality. The findings, of the first phase of the study supports the hypothesis that microwave energy can kill insects in maize by scalding due to selective heating. This is related to the higher dielectric properties of insects compared to maize kernels, because of their higher moisture content. Exposure time and length of the microwave cavity are very important parameters in the microwave treatment as they can influence kernel damage (swelling, discolouration and popping) as a result of heating. Consequently, to achieve 100% mortality of all insect species, the pulsed mode 1.5 kW power level at 9 sec exposure time using the 483 mm long cavity should be used.

The second phase of the study also supports the hypothesis that the microwave treatment can be optimized so that it will not cause significant thermal damage to the maize kernels. While both normal (power dosage of 1.5 kW, for 9 sec exposure time) and harsh (2 kW, 18 sec exposure time) treatment conditions can eradicate adult insects and their eggs, only the former maintains maize quality. The normal microwave treatment causes significant moisture and kernel weight losses.

The use of microwave technology has potential to be used as an insect control measure in maize as an alternative to chemical pesticides. Although the cost of microwave technology as an insect infestation control measure may seem high, its safety (no residual left on products) and its environmental friendliness (no ozone depletion) should be taken into consideration.

Future research work should focus on the effect of microwave processing on maize products' nutritional quality (starch and protein digestibility, mineral and vitamins bioavailability). Research can be conducted on how the ideal microwave cavity design (size and dimensions), should be to improve the efficiency of the process in a maize mill.

7. REFERENCES

AACC International, 2000. Approved Methods of the AACC, 10th Ed. AACC Method 08-02, Method 30-25, Method 32-10, Method 44-15A, Method 46-30, and Method 55-10. American Association of Cereal Chemists: St Paul, MN.

Abdelrahman, A. A., and Hosney, R.C. 1984. Basis of hardness in pearl millet, grain sorghum, and corn. *Cereal Chem.* 61:232-235.

Aldryhim, Y. N. 1990. Efficiency of the amorphous silica dust, dryacide, against *Tribolium confusum* Duv. and *Sitophilus granarius* (L.) (Coleoptera: Tenebrionidae and Curculionidae). *J. Stored Prod. Res.* 26:207-210.

Annis, P. C., and Morton, R. 1997. The acute mortality effects of carbon dioxide on various life stages of *Sitophilus oryzae*. *J. Stored Prod. Res.* 33:115-124.

Baker, V. H., Wiant, D. E. and Taboada, O. 1956. Some effects of microwaves on certain insects which infest wheat and flour. *J. Econ. Entomol.* 49:33-37.

Banik, S., Bandyopadhyay, S., and Ganguly, S. 2003. Bioeffects of microwave - a brief review. *Bioresour. Technol.* 87:155-159.

Banks, J., and Fields, P. 1995. Physical methods for insect control in stored grain ecosystem. Pages 353-409 in: *Stored-Grain Ecosystems*. D. S. Jayas, N. D. G. White and W. E. Muir, eds. Marcel Dekker: New York.

Beckett, S. J., and Morton, R. 2003. Mortality of *Rhyzopertha dominica* (F.) (Coleoptera:Bostrychidae) at grain temperatures ranging from 50°C to 60°C obtained at different rates of heating in a spouted bed. *J. Stored Prod. Res.* 39:313-332.

Bell, C. H. 2000. Fumigation in the 21st century. *Crop Prot.* 19:563-569.

Bell, C. H., and Armitage, D.M. 1992. Alternative storage practices. Pages 249-311 in: Storage of Cereal Grains and their Products. D. B. Sauer, ed. American Association of Cereal Chemists: St Paul, MN.

Bradford, M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal. Biochem.* 72:248-254.

Brekke, O. L., Griffin, Jr., and Shove, G. C. 1973. Dry milling of corn artificially dried at various temperatures. *Trans. ASAE* 16:761-765.

Brown, R. B., Fulford, G. N., Daynard, T. B., Meiering A. G., and Otten, L. 1979. Effect of drying method on grain quality. *Cereal Chem.* 56:529-532.

Chandrashekar, A., and Mazhar, H. 1999. The biochemical basis and implications of grain strength in sorghum and maize. *J. Cereal Sci.* 30:193-207.

Chau, K. J., Mujumdar, A. S., and Chou, S. K. 2003. Intermittent drying of bioproducts – an overview. *Bioresour. Technol.* 90:285-295.

Childs P. R. N., Greenwood, J. R. and Long, C. A. 2000. Review of temperature measurement. *Rev. Sci. Instrum.* 71:2959-2978

Cotton, R. T., and Wilbur, D. A. 1982. Insects. Pages 281-318 in: Storage of Cereal Grains and their Products. M. C. Christensen, ed. American Association of Cereal Chemists: St Paul, MN.

David, M. D., Mills, R. B., and White, G. D. 1977. Effects of low temperature acclimation on developmental stages of stored-product insects. *Environ. Entomol.* 6:181-184.

Datta, A. K., Berek, H., Little, A., and Ramaswamy, H. S. 2001. Measurement and instrumentation. Pages 279-296 in: Handbook of Microwave Technology for Food applications A. K. Datta and R. C. Anantheswaran, eds. Marcel Dekker: New York.

- Dibben, D Datta, A. K., and Anantheswaran, R. C. 2001. Electromagnetics: Fundamental aspects and numerical modelling. Pages 1-28 in: Handbook of Microwave Technology for Food Applications. A. K. Datta and R. C. Anantheswaran, eds. Marcel Dekker: New York.
- Duvick, D. N. 1961. Protein granules in maize endosperm cells. *Cereal Chem.* 38:374-385.
- Ebeling, W. 1971. Sorptive dusts for pest control. *Annu. Rev. Entomol.* 16:123-158.
- Eckhoff, S. R. 1995. Maize. Pages 2825-2831 in: *Encyclopaedia of Food Science, Food Technology and Nutrition*. R. Macrae, R. K. Robinson and M. J. Sadler, eds. Academic Press: London.
- Erasmus, C., and Taylor, J. R. N. 2004. Optimising the determination of maize endosperm vitreousness by rapid non-destructive image analysis technique. *J. Sci. Food Agric.* 84:920-930.
- Eskom, 2007. Tariffs and Charges from 01 April 2007 to 31 March 2008. www.eskom.co.za. (website accessed September 2007).
- Fantom, A. 1990. Calorimeters. Pages 16-113 in: *Radiofrequency & Microwave Measurement*. Peter Peregrinus: London.
- Felker, F. C., and Paulis, J. W. 1993. Quantitative estimation of corn endosperm vitreosity by video image analysis. *Cereal Chem.* 70:685-689.
- Fields, P. G., 1992. The control of stored product insects and mites with extreme temperatures. *J. Stored Prod. Res.* 28:89-118.
- Fields, P. G., and White, N. D. G. 2002. Alternatives to methyl bromide treatments for stored-product and quarantine insects. *Annu. Rev. Entomol.* 47:331-360.

Food and Agriculture Organization, 1984. Manual of fumigation for insect control. <http://www.fao.org/docrep/X5042E/x5042E0a.htm#phosphine> (website accessed May 2007).

Food and Agriculture Organization, 1992. Maize in human nutrition. <http://www.fao.org/docrep/T0395E/T0395E01.htm>. (website accessed June 2004).

Food and Agriculture Organization, 2002. Assessment of the world food security situation. Committee on world food security, 28th session, Rome. http://www.fao.org/docrep/meeting/004/y6441e/y6441e00.htm#P292_17440_ (website accessed May 2007).

Food and Agriculture Organization, 2005. Evaluation of the hazard to consumers resulting from the use of fumigants in the protection of food. Meeting report No. PL/1965/10/2. <http://www.inchem.org/documents/jmpr/jmpmono/v65apr09.htm> (website accessed July 2005).

Food and Agriculture Organization, 2007. Insect pests of stored grains in hot climates. <http://www.fao.org/docrep/T1838E/T1838E1f.htm>. (website accessed October 2007).

Goedeken, D. L., Tong, C. H. and Lentz, R. R. 1991. Design and calibration of a continuous temperature measurement system in a microwave cavity by infrared imaging. *J. Food Process. Preserv.* 15:331-337.

Golob, P. 1997. Current status and future perspectives for inert dusts for control of stored product insects. *J. Stored Prod. Res.* 33:69-79.

Gunasekaran, S. 1990. Grain drying using continuous and pulsed microwave energy. *Dry. Technol.* 8:1039-1047.

Gunasekaran, S., and Muthukumarappan, K. 1993. Breakage susceptibility of corn of different stress-cracks categories. *Trans. ASAE* 36:1445-1446.

Gunasekaran, N., and Rajendran, S. 2005. Toxicity of carbon dioxide to drugstore beetle *Stegobium panacenum* and cigarette beetle *Lasioderma serricorne*. J. Stored Prod. Res. 41:283-294.

Gunasekaran, S., and Yang, H.W. 2007. Optimization of pulsed microwave heating. J. Food Eng. 78:1457-1462.

Gunasekaran, S., Cooper, M., Berlage, A. G., and Krishnan, P. 1987. Image processing for stress cracks in corn kernels. Trans. ASAE 30:266-271.

Güner, M. 2007. Pneumatic conveying characteristics of some agricultural seeds. J. Food Eng. 80:904–913.

Halverson, S. L., Burkholder, W. E., Bigelow, T. S., Nordheim, E. V. and Misenheimer, M. E. 1996. High - power microwave radiation as an alternative insect control method for stored products. Stored Prod. Entomol. 89:1638-1648.

Hamid, M. A. K., and Boulanger R. J. 1970. Control of moisture and insect infestation by microwave power. Milling 152:25-32.

Hang, Y. J., Feng, Y., and Weller, C. L. 1996. Frequency domain image analysis for detecting stress cracks in corn kernels. Appl. Eng. Agric. 12:487-592.

Harein, P. K., and Davis, R. 1992. Control of stored grain insects. Pages 491-534 in: Storage of Cereal Grains and their Products. D. B. Sauer, ed. American Association of Cereal Chemists: St Paul, MN.

Harney, M. 1993. Pages 1-112 in: Plant Protection Research Institute Handbook No 1. Gutenberg Book Printers: Pretoria.

Haros, M., Tolaba M. P. and Suárez, C. 2003. Influence of corn drying on its quality for the wet milling process. J. Food Eng. 60:177-184.

Harrison, D. L. 1980. Microwave versus conventional cooking methods: Effects on food quality attributes. *J. Food Prot.* 43:633-637.

Hasan, M., and Khan, A. T. 1998. Control of stored-product pests by irradiation. *Integrated Pest Managem. Rev.* 3:15-29.

Hoffman, C. J., and Zabik, M. E. 1985. Effects of microwave cooking/ heating on nutrients and food systems: A review of recent studies. *J. Am. Diet. Assoc.* 85:922-926.

Hooper, J. L., Desmarchelier, J. M., Ren, Y. and Allen, S. E. 2003. Toxicity of cyanide to insects of stored grain. *Pest Manag Sci.* 59:353-357.

Hoseney, R. C. 1994. Pages 1-28 in: *Principles of cereal science and technology*, 2nd ed. American Association of Cereal Chemists: St Paul, MN.

Hoseney, R. C., and Faubion, J. M. 1992. Physical properties of cereal grains. Pages 1-38 in: *Storage of cereal grains and their products*. D. B. Sauer, D.B., ed. American Association of Cereal Chemists: St Paul, MN.

Howe, R. W. 1965. A summary of estimates of optimal and minimal conditions for population increase of some stored products insects. *J. Stored Prod. Res.* 1:177-184.

ICC, 1994. ICC standard no 123/1. International Association for Cereal Science and Technology: Vienna.

Inglett, G. E. 1970. Pages 123-138 in: *Corn: Culture, Processing, Products*. The AVI Publishing Company: London.

Johnson, W. H., and Lamp, B. J. 1966. Measurement of factors related to corn condition. Pages 343-367 in: *Principles, equipment and system for corn harvesting*. Agricultural Consulting Associates: Ohio.

Jood, S., and Kapoor, A. C. 1992. Effect of storage and insect infestation on protein and starch digestibility of cereal grains. *Food Chem.* 44:209-212.

Jood, S, Kapoor, A. C. and Singh, R. 1992a. Biological evaluation of protein quality of maize as affected by insect infestation. *J. Agric. Food Chem.* 40:2439–2442.

Jood, S., Kapoor, A. C. and Singh, R. 1992b. Mineral content of cereal grains as affected by storage and insect infestation. *J. Stored Prod. Res.* 28:147-152.

Jugenheimer, R. W. 1976. Pages 25-43 in: *Corn Improvement, Seed Production and Uses.* John Wiley & Sons: New York.

Kells, S. A., Mason, L. J., Maier, D. E. and Woloshuk, C. P. 2001. Efficacy and fumigation characteristics of ozone in stored maize. *J. Stored Prod. Res.* 37:371-382.

Kent, N. L., and Evers, A. D. 1994. Pages 1-28 in: *Kent's Technology of Cereals*, 4th ed. Elsevier Science: Oxford.

Kim, J. B., Yousef, A. E. and Dave, S. 1999. Application of ozone for enhancing the microbiological safety and quality of foods: A review. *J. Food Prot.* 62:1071-1087.

Kirkpatrick, R. L., and Roberts, J. R. Jr. 1971. Insect control in wheat by use of microwave energy. *J. Econ. Entomol.* 64:950-951.

Kirleis, A. W., and Stroshine, R. L. 1990. Effects of hardness and drying air temperature on breakage susceptibility and dry milling characteristics of yellow dent corn. *Cereal Chem.* 67:523-528.

Korunic, Z. 1998. Diatomaceous earth, a group of natural insecticides. *J. Stored Prod. Res.* 3:87-97.

Korunic, Z., Fields, P. G., Kovacs, M. I. P., Noll, J. S., Lukow, O. M., Demianyk, C. J. and Shibley, K. J. 1996. The effect of diatomaceous earth on grain quality. *Postharvest Biol. Technol.* 9:373-387.

Kraszewski, A. W., and Nelson, S. O. 1994. Determination of moisture content and bulk density of shelled corn by measurement of microwave parameters. *J. Agric. Eng. Res.* 58:37-46.

Lambert, J. P. 1980. Biological hazards of microwave radiation. *J. Food Prot.* 43:625-628.

Lane, J. A. 1972. Pages 3-15 in: *Microwave Power Measurement*. Peter Peregrinus: London.

Lauber, W. L., Carroll, J. A., Dufield, D. R., Kiesel, J. R., Radabaugh, M. R. and Malone, J. P. 2001. Mass spectrometry compatibility of two-dimensional gel protein stains. *Electrophoresis* 22:906-918

Laverghetta, T. S. 1988. Pages 333-418 in: *Modern Microwave Measurements and Techniques*. Artech House: Norwood, MA.

Lawton, J. W., and Wilson, C. M. 2003. Proteins of the kernel. Pages 314-354 in: *Corn: Chemistry and Technology*, 2nd Ed. P. J. White and L. A. Johnson, eds. American Association of Cereal Chemists: St Paul, MN.

Lewandowicz, G., Jankowski, T. and Formal, J. 2000. Effect of microwave radiation on physico-chemical properties and structure of cereal starches. *Carbohydr. Polym.* 42:193-199.

Lewis, M., and Heppell, N. 2000. Pages 1-31 in: *Continuous Thermal Processing of Foods – Pasteurization and UHT Sterilization*. Aspen Publishers: Gaithersburg, ML.

Li, P. X.-P., Handacre, A. K., Campanella, O. H. and Kirkpatrick, K. J. 1996. Determination of endosperm characteristics of 38 corn hybrids using the Stenvert Hardness Test. *Cereal Chem.* 73:466-471.

- Maier, D. E., Adams, W. H., Throne, J. E. and Mason, L. J. 1996. Temperature management of the maize weevil, *Sitophilus zeamais* Motsch. (Coleoptera: Curculionidae), in three locations in the United States. *J. Stored Prod. Res.* 32:255-273.
- Mankowski, J. 2000. A review of short pulse generator technology. *IEEE Trans. Plasma. Sci.* 28:102-108.
- Mann, M., Hendrickson, R. C. and Pandey, A. 2001. Analysis of proteins and proteomics by mass spectrometry. *Annu. Rev. Biochem.* 70:437-473.
- Mason, L., and Storey, C. L. 2003. Effect and control of insects affecting corn quality. Pages 221-239 in: *Corn Chemistry and Technology*, 2nd Ed. P. J. White L. A. Johnson, eds. American Association of Cereal Chemists: St Paul, MN.
- McGuire, T. A., and Earle, F. R. 1958. Changes in the solubility of corn protein resulting from the artificial drying of high-moisture corn. *Cereal Chem.* 35:179-188.
- Mendez, F., Maier, D. E., Mason, L. J. and Woloshuk, C. P. 2003. Penetration of ozone into columns of stored grains and effects on chemical composition and processing performance. *J. Stored Prod. Res.* 39:33-44.
- Mestres, C., and Matencio, F. 1996. Biochemical basis of kernel milling characteristics and endosperm vitreousness of maize. *J. Cereal Sci.* 24:283-290.
- Mijović, J., and Wijaya, J. 1990. Review of cure of polymers and composites by microwave energy. *Polym. Compos.* 11:183-191.
- Mishenko, A. A., Malinin, O. A., Rashkovan, V. M., Basteev, V. A., Bazyma, L. A., Mazalov, Yu P. and Kutovoy, V. A. 2000. Complex high - frequency technology for protection of grain against pests. *J. Microwave Power Electromagn. Energy* 35:179-184.

Mourier, H., and Poulsen, K. P. 2000. Control of insects and mites in grain using a high temperature/ short time (HTST) technique. *J. Stored Prod. Res.* 36:309-318.

Natarajan, S., Xu, C., Caperna, T. J. and Garrett, W. M. 2005. Comparison of protein solubilization methods suitable for proteomic analysis of soybean seed proteins. *Anal Biochem.* 342:214-220.

Nelson, S. O. 1972. Insect – control studies with microwaves and other radiofrequency energy. *Bull. Entomol. Soc. Am.* 19:157-163.

Nelson, S. O. 1985. RF and microwave energy for potential agricultural applications. *J. Microwave Power* 20:65-70.

Nelson, S.O. 1996. Review and assessment of radio-frequency and microwave energy for stored-grain insect control. *Trans. ASAE* 39:1475–1484.

Nelson, S.O., and Kantack, B. H. 1966. Stored-grain insect control studies with radio-frequency energy. *J. Econ. Entomol.* 59:588-594.

Nelson, S. O., and Stetson, L. E. 1974. Comparative effectiveness of 39- and 2,450-MHz electric fields for control of rice weevils in wheat. *J. Econ. Entomol.* 67:592-595.

Nelson, S. O., and Whitney W. K. 1960. Radio-frequency electric fields for stored-grain insect control. *Trans. ASAE* 3:133-137,144

Nelson, S. O., Bartley Jr., P. G. and Lawrence, K. C. 1997. Measuring RF and Microwave permittivities of adult rice weevils. *IEEE Trans. Instrum. Meas.* 46:941-946.

Nielsen, J. P. 2003. Evaluation of malting barley quality using exploratory data analysis. II. The use of kernel hardness and image analysis as screening methods. *J. Cereal Sci.* 38:247-255.

Ofuya, T. I., and Reichmuth, C. 2002. Effect of relative humidity on the susceptibility of *Callosobruchus maculatus* (Fabricius) (Coleoptera: Bruchidae) to two modified atmospheres. *J. Stored Prod. Res.* 38:139-146.

Oliveira, M.E.C., and Franca, A.S., 2002. Microwave heating of foodstuffs. *J. Food Eng.* 53:347-359.

Ohlsson, T., and Bengtsson, N. 2001. Microwave technology and foods. Pages 34:65-140 in: *Advances in Food and Nutrition Research*. S. L. Taylor, ed. Academic Press: San Diego, CA.

Pande, A., 1975. Pages 3:717-780 in: *Handbook of Moisture Determination and Control*. Marcel Dekker: New York.

Pandey, A., and Mann, M. 2000. Proteomics to study genes and genomes. *Nature* 405:837-846.

Paulsen, M. R., Hill, L. D. 1985. Corn quality factors affecting dry milling performance. *J. Agric. Eng. Res.* 31:255-263.

Paulsen, M. R., Watson, S. A. and Singh, M. 2003. Measurement and maintenance of corn quality. Pages 1159-1219 in: *Corn: Chemistry and Technology*, 2nd. Ed. P. J. White and L. A. Johnson, eds. American Association of Cereal Chemists: St. Paul.

Peplinski, A. J., Paulis, J. W, Bietz, J. A. and Pratt, R. C. 1994. Drying of high moisture corn: Changes in properties and physical quality. *Cereal Chem.* 7:129-133.

Pomeranz, Y. 1992. Biochemical, functional, and nutritive changes during storage. Pages 55-141 in: *Storage of Cereal Grains and their Products*. D. B. Sauer, ed. American Association of Cereal Chemists: St Paul, MN.

Pomeranz, Y., Czuchajowska, Z. and Lai, F. S. 1986a. Comparison of methods for determination of hardness and breakage susceptibility of commercially dried corn. *Cereal Chem.* 63:39-43.

Pomeranz, Y., Hall, G. E., Czuchajowska, Z., Martin, C. R. and Lai, F.S. 1986b. Test weight, hardness, and breakage susceptibility of yellow dent corn hybrids. *Cereal Chem.* 63:349-351.

Pomeranz, Y., Martin, C. R., Taylor, D. D. and Lai, F. S. 1984. Corn hardness determination. *Cereal Chem.* 61:147-150.

Pomeranz, Y., Czuchajowska, Z., Martin, C. R. and Lai, F. S. 1985. Determination of corn hardness by the Stenvert Hardness Tester. *Cereal Chem.* 62:108-112.

Price, N. R. 1985. The mode of action of fumigants. *J. Stored Prod. Res.* 21:157-164.

Rashkovan, V. M., Khizhnyak, N. A., Basteev, A. V., Bazyma, L. A., Niño de Rivera, L. and Ponomaryova, I. A. 2003. Interaction of electromagnetic waves with granular agricultural product and insects. *J. Microwave Power Electromagn. Energy* 38:225-234.

Roberts, E. H. 1972. Pages 321-359 in: *Viability of Seed*. Syracuse University Press: New York.

Shayesteh, N., and Barthakur, N. N. 1996. Mortality and behavior of two stored-product insect species during microwave irradiation. *J. Stored Prod. Res.* 32:239-246.

Shivhare, U. K. 1992. Drying characteristics of corn in a microwave field in a microwave field with a surface-wave applicator (Ph.D. Thesis summary). *Dry. Technol.* 10:1323-1326.

Shukla, R., and Cheryan, M. 2001. Zein: The industrial protein from corn. *Ind. Crops Prod.* 13:171-192.

Slavin, J. L., Jacobs, D. and Marquart, L. 2000. Grain Processing and Nutrition. *Crit. Rev. Food Sci. Nutr.* 40:309–326.

Smit, B. 1964. Pages 7-17 in: *Insects in Southern Africa: How to Control Them.* Oxford University Press: Cape Town.

Song, H. P., and Litchfield, J. B. 1994. Measurement of stress cracking in maize kernels by magnetic resonance imaging. *J. Agric. Res.* 57:109-118.

South African Department of Agriculture, 2004. Trends in the agricultural sector 2004. <http://www.nda.agric.za/docs/trends2004.pdf>. (website accessed February 2006).

Storey, C. L., and Sauer, D. B. and Walker, D. 1983. Insect populations in wheat, corn, and oats stored on the farm. *J. Econ. Entomol.* 76:1323-1330.

Stroshine, R. L., Kirlies, A. W., Tuite, J. F., Bauman, L. F. and Emam, A. 1986. Differences in the grain quality among selected corn hybrids. *Cereal Foods World* 31:311-316.

Szepes, A., Hasznos-nezdei, M., Kovács, J., Funke, Z., Ulrich, J. and Szabó-Révész, S. 2005. Microwave processing of natural biopolymers – studies on the properties of different starches. *Int. J. Pharm.* 302:166-171.

Taylor, R. W. D. 1994. Methyl bromide – is there any future for this noteworthy fumigant? *J. Stored Prod. Res.* 30:253-260.

Taylor, R. W. 1999. Alternatives and potential alternatives for methyl bromide for disinfecting durable commodities. Pages 61-72 in: *Proceedings of the Workshop on Methyl Bromide Alternatives for North African and Southern European Countries.* G. Tierney, C. Gilfillan, S. Kilkwe and J. Forrest, eds. Rome, Italy, 1998.

Thostenson, E. T., and Chou, T. W. 1999. Microwave processing: fundamentals and applications. *Compos. Pt. A-Appl. Sci. Manuf.* 30:1055-1071.

Van Tonder, H. J., and Prinsloo, G. L. 2000. Plant Protection Research Institute Handbook No.11. ARC-Plant Protection Research Institute, Pretoria.

Velu, V., Nagender, A., Rao, P. P. G. and Rao, D. G. 2006. Dry milling characteristics of microwave dried maize grains (*Zea mays L.*). *J. Food Eng.* 74:30-36.

Walde, S. G., Balaswamy, K., Velu, V. and Rao, D.G. 2002. Microwave drying and grinding characteristics of wheat (*Triticum aestivum*). *J. Food Eng.* 55:271-276.

Wall, J. S., James, C. and Donaldson, G. L. 1975. Corn proteins: Chemical and physical changes during drying of grain. *Cereal Chem.* 52:779-790.

Wang, S., Tang, J., Johnson, J. A., Mitcham, E., Hansen, J. D., Hallmann, G., Drake, S. R. and Wang, Y. 2003. Dielectric properties of fruits and insects pests as related to radio frequency and microwave treatments. *Biosyst. Eng.* 85:201-212.

Warchalewski, J. R., Pra dzynska, A., Gralik, J. and Nawrot, J. 2000. The effect of gamma and microwave irradiation of wheat grain on development parameters of some stored grain pests. *Nahrung* 44:411-414.

Watson, S. A. 2003. Description, development, structure, and composition of the corn kernel. Pages 69-106 in: *Corn: Chemistry and Technology*. P. J. White, and L. A. Johnson, eds. American Association of Cereal Chemists: St Paul, MN.

Watters F. L. 1976. Microwave radiation for control of *Tribolium confusum* in wheat and flour. *J. Stored Prod. Res.* 12:19-25.

Webber, H. H., Wagner, R. P. and Pearson, A. G. 1946. High frequency electrical field as lethal agents for insects. *J. Econ. Entomol.* 39:487-498.

White, N. D. G. 1995. Insects, mites, and insecticides in stored-grain ecosystem. Pages 123-167 in: *Stored-Grain Ecosystems*. D. S. Jayas, N. D. G. White and W. E. Muir, eds. Marcel Dekker: New York.

Wight, A. W. 1979. Detection of heat damage in artificially fried maize. *South African Food Review* 6:63-70.

Wight, A. W. 1981. Changes in properties of some maize cultivars associated with artificial drying at elevated temperatures. *Starch/ Stärke* 33:165-168.

Yang, H. W., and Gunasekaran, S. 2001. Temperature profiles in a cylindrical model food during pulsed microwave heating. *J. Food Sci.* 66:998-1004.

Yang, H. W., and Gunasekaran, S. 2004. Comparison of temperature distribution in model food cylinders based on Maxwell's equations and Lambert's law during pulsed microwave heating. *J. Food Eng.* 64:445-453.

Yie, T. J., Liao, K., Paulsen, M. R., Reid, J. F. and Maghirang, E. B. 1993. Corn kernel stress crack detection by machine vision. ASAE meeting Presentation. Paper No. 93-3526. Chicago, Illinois.

Yuan, J., and Flores, R. A. 1996. Laboratory dry-milling performance of white corn: effect of physical and chemical corn characteristics. *Cereal Chem.* 73:574-578.

Zettler, J. L., and Cuperus, G. W. 1990. Pesticide resistance in *Tribolium castaneum* (Coleoptera: Tenebrionidae) and *Rhyzopertha dominica* (Coleoptera: Bostrichidae) in wheat. *J. Econ. Entomol.* 83:1677-1681.