INTRODUCTION

CHAPTER 1: INTRODUCTION

1.1 STATEMENT OF THE PROBLEM

Brown bread is an important staple food in many countries (Gan, Ellis, Vaughan & Galliard, 1989; Cauvain, 1998a) and in some, e.g. South Africa, it accounts for more than 46% of bread produced (Randall, Wessels & Traut, 1995). Annually, 3-3.5 billion loaves of brown bread are sold in South Africa. This equates to 4-5 slices of bread per capita per day (Mr. P.J. Cownie, Executive Director, South African Chamber of Baking -Personal communication). The high production of brown bread can be attributed to an increase in demand for food products containing elevated levels of dietary fibre (Galliard, 1986a), and also in the case of South Africa the lower selling price of brown bread. Epidemiological observations that several diseases of "civilisation", such as coronary heart disease, diabetes, and some colon diseases are most prevalent in western countries, have heightened interest in the inclusion of more fibre in our diets (Pomeranz, Shogren, Finney & Bechtel, 1977; Johnson & Southgate, 1994).

Brown bread is bread made from part or all of the pericarp of wheat, in addition to the endosperm. The non-endosperm components (germ, bran and epicarp hairs) of the wheat caryopsis are known to be responsible for producing the low specific volume and dense crumb structure of traditional wholemeal bread (Gan, Galliard, Ellis, Angold & Vaughan, 1992). Their effect cannot be explained solely in terms of the dilution effect on the gluten-forming proteins and the precise reason for the decrease in loaf volume, caused by their addition, is not known (Bloksma & Bushuk, 1988).

The chemical effects of bran on baking quality have been studied extensively (Sullivan, Howe & Schmalz, 1936; Hullet & Stern, 1941; Warwick, Farrington & Shearer, 1979; Bell, Daniels & Fisher, 1979; Barnes & Lowy, 1986; Galliard, 1986a; Galliard 1986b; Tait & Galliard, 1988; Stear, 1990; Schofield & Chen, 1995; Nelles, Randall & Taylor, 1998). From these studies it can be concluded that the reduction in baking performance may be due mainly to the presence of lipid metabolising enzymes (present in bran and germ fractions), as well as reducing substances such as glutathione.

The physical effects of bran on baking quality have also been studied (Galliard & Gallagher, 1988; Gan et al, 1989; Gan et al, 1992;), but in less detail. It appears that the non-endosperm components, notably the epicarp hairs, play a prominent role in disrupting the gluten protein matrix (Gan et al, 1989).

The situation with brown bread in South Africa is unusual in three ways. Firstly, not all South African wheats have excellent breadmaking qualities (Cheetham, 1988). Secondly, the consumption of brown bread is much higher than in other countries. This makes the production of bread with a good loaf volume and crumb structure all the more important. Thirdly, and perhaps the most important difference is the method of brown bread production. In South Africa brown bread is produced by adding bran back to the white base flour (replacing up to 15% of the flour), in contrast with other countries where brown flour is generally produced during the milling process itself. The manufacture of brown bread by the addition of bran to white flour presents the baker with an opportunity to optimise baking performance and loaf volume, if it could be established in what condition the bran should be added, for example what size or shape the bran should be, or how it should be treated before addition.

At present, the absence of a complete understanding of the effects of bran makes the manufacture of whole wheat bread with the same loaf volume as white bread expensive, since extra flour must be used to compensate for the bran effect (Lai, Davis & Hoseney, and 1989a). It has recently been suggested that further research is needed to elucidate the interplay of what appears to be numerous chemical and physical effects (Gan, Ellis & Schofield, 1995).

INTRODUCTION

1.2 **OBJECTIVES**

The main objective of this project was:

To investigate the chemical and physical effects of different brans on brown bread

This incorporated investigation of the following:

- The influence of bran origin on loaf volume \bullet
- The effect of bran type and bran particle size on loaf volume \bullet
- The effect of heat treatment of the bran on loaf volume \bullet
- The effect of bran on bubble formation in the dough \bullet
- The influence of bran on sensory quality of brown bread \bullet

CHAPTER 2: LITERATURE REVIEW

2.1 THE WHEAT KERNEL

2.1.1 Structure

The structure of the wheat kernel has been reviewed by many authors including Evers & Bechtel (1988) and Hoseney (1994). Members of the grass family (Poaceae), which include wheat, produce dry one-seeded fruits. This type of fruit is a caryopsis but is commonly called a kernel or grain. A longitudinal and cross section of a caryopsis or kernel of wheat is shown diagrammatically in Figure 1.

Fig. 1: Longitudinal and cross sections of a wheat kernel (Hoseney, 1994).

The kernels average about 8 mm in length and weigh about 35 mg. Wheat kernels are rounded on the dorsal side and have a longitudinal crease the length of the ventral side (opposite the germ). Wheat grains can be separated into three main parts: the bran, the germ or embryo, and the endosperm. The relationship between grain components and major mill fractions are shown in Table 1.

Ideal relationship between botanical constituents and major mill Table 1: fractions (Evers & Bechtel, 1988).

a) Bran

The hairs of brush, or epicarp hairs of the wheat kernel can be seen in Figure 1. During milling, the first (outermost bran) fraction contains the highest concentration of epicarp hairs (Gan et al, 1992). Gan et al (1989) found that these epicarp hairs appear to play a prominent role in disrupting the gluten protein matrix.

The bran consists of the outermost layer of cells of the wheat grain. These cells have thick walls made mainly of cellulose and hemicellulose. The bran layers account for 14-15% of the entire kernel (Wheat Board, s.a.) and consists of the pericarp, the seed coat and nucellar epidermis, as well as the aleurone layer (Hoseney, 1994).

The pericarp surrounds the entire seed and is composed of several layers. The outer pericarp is called the beeswing. The innermost portion of the outer pericarp consists of remnants of thin-walled cells; because of their lack of continuous cellular structure, they from a natural plane of cleavage. When they are disrupted, the beeswing is released (Hoseney, 1994). The inner pericarp is composed of intermediate cells, cross cells and tube cells (Evers & Bechtel, 1988).

The seed coat is firmly joined to the tube cells on the outside and the nucellar epidermis on the inside (Hoseney, 1994). The seed coat does not cover the entire surface of the seed, because it stops short on either side of the crease. However, the space in the crease is occupied by a structure called the pigment strand which runs along the chalazal region just inside the conducting tissue (Evers & Bechtel, 1988). The seed coat consists of three layers: a thick outer cuticle, a layer that contains pigment (for coloured wheats) and a thin inner cuticle. The thickness of the seed coat varies from 5 to 8 μ m. The nucellar epidermis, or hyaline layer, is about 7 um thick and closely united to both the seed coat and the aleurone layer (Hoseney, 1994).

The aleurone layer, which is generally one cell thick, completely surrounds the kernel, covering both the starchy endosperm and the germ. From a botanical point of view, the aleurone layer is the outermost layer of the endosperm. During milling, however, it is removed along with the nucellar epidermis, seed coat and pericarp to form part of the bran. The aleurone cells are heavy-walled, essentially cube-shaped and free of starch. The average thickness of the cells is about 50 μ m. The cell walls are 3-4 μ m thick, and

have been reported to be largely cellulosic in composition (Hoseney, 1994). The cell walls of the aleurone layer are somewhat thicker than those of the starchy endosperm. They consist of two distinct layers: an inner, relatively thin layer (0.3-0.6 µm) that remains essentially intact during germination, and an outer layer that has a striated or lamellated appearance and is degraded during germination (Fincher & Stone, 1986).

b) Germ

The germ of wheat comprises 2.5-3.5% of the kernel. The germ is composed of two major parts, the embryonic axis (rudimentary root and shoot) and the scutellum, which functions as the storage organ (Hoseney, 1994). A layer of crushed cells (fibrous region) separates the scutellar epithelium from the starchy endosperm. These crushed cells are extremely important, for it is at this junction that the embryo is separated from the rest of the grain during milling. Crushed cells are formed from starchy endosperm cells that fail to develop normally (Evers & Bechtel, 1988).

c) Endosperm

The starchy endosperm consists of three types of cells: peripheral, prismatic and central. The cells vary in size, shape and location within the kernel. The peripheral cells are the first row of cells inside the aleurone layer and are usually small, equal in diameter in all direction or slightly elongated toward the center of the kernel. Several rows of elongated prismatic cells are found inside the peripheral cells. They extend inward to about the center of the cheeks. The central cells are inside the prismatic cells. They are more irregular in size and shape than the other cells (Hoseney, 1994).

The contents and cell walls of the endosperm make up flour. The cells are packed with starch granules embedded in a protein matrix. The protein is mostly gluten, the storage proteins of wheat. In maturing wheat, the gluten is synthesised in protein bodies. However, as the grain matures, the protein bodies are compressed together into a matrix that appears claylike, and the protein bodies are no longer discernible. The starch granules occur as large, lenticular granules of up to 40 µm across the flattened side and as small spherical granules (2-8 µm in diameter) (Hoseney, 1994).

2.1.2 Chemical composition

The chemical composition of whole grain, endosperm, bran and germ, as reviewed by Bushuk and Scanlon (1993), is given in Table 2.

Composition of whole grain, endosperm, bran and germ (Bushuk & Table 2: **Scanlon, 1993)**

^aFigures in parentheses are expressed as percent of whole grain

a) Bran

Much of the available information concerning the composition of the bran layers refers to them only as a whole. The reported composition depends on the type of wheat milled, the way in which the bran was separated, the mill stream from which the bran has been taken, as well as the method used to determine the composition (Oakenfull & Topping, 1987; Pomeranz, 1988). To emphasise this point, it is of interest that a procedure described for separating the aleurone and pericarp-seed coat of wheat bran produced fractions that have widely different effects in dietary trials with animals (Cheng, Trimble, Illman, Stone & Topping, 1987). The proportion of fat, starch and protein present depends to a large degree on the amount of germ and endosperm extracted with the bran. Bran is therefore a variable commodity (Oakenfull & Topping, 1987).

Commercial bran has a very low gluten protein content. When bran replaces flour in a brown bread formulation, it dilutes the functional gluten protein (Pomeranz et al, 1977). Czuchajowska and Pomeranz (1993) found that when high protein flour was replaced by dietary fibre (10%), dough rise was impaired as a result of dilution of functional protein.

It was possible to compensate, to various degrees, for the volume decrease when the dough was supplemented with high-quality gluten.

The mineral content of bran is between 4.8 and 6.7%, the biggest contributors being potassium (1.2-1.9%) and phosphorus (0.9-1.55%). Lipid constituents (determined by ether extraction) of mill bran make up 3.1-4.8% of the total composition. Niacin is present in the range of 140-214 ppm, while tocopherols range from 25-87 ppm, pantothenic acid 39.9 ppm, riboflavin 1.5-4.8 ppm and thiamin 4.5-6.4 ppm (Pomeranz, 1988).

Bran is a good source of dietary fibre (Theander, Westerlund & Åman, 1993). As reviewed by Delcour and Eerlingen (1996), there are a number of definitions of dietary fibre: 1) the sum of plant polysaccharides and lignin that are resistant to hydrolysis by the digestive enzymes of man; 2) the sum of non-starch polysaccharides (NSPS) and lignin; and 3) non-starch polysaccharides. Furthermore, resistant starch can also be classified as dietary fibre. Lee and Prosky (1996) proposed the expansion of the definition of dietary fibre to include resistant oligosaccharides (oligosaccharides that are resistant to hydrolysis by human alimentary enzymes), in addition to currently included non-starch polysaccharides, resistant starch and lignin. The dietary fibre content and composition of wheat bran is given in Table 3.

^aCalculated as percentage of dry product

A number of enzymes are present in the bran, including phosphomonoesterase, diamine oxidase, lipoxidase, dehydrogenases, hypertensinase, glycosidases, catalase and peroxidase. Catalase activity is four to eight times higher in bran than in flour and peroxidase five to 13 times higher in bran (Pomeranz, 1988). O'Connor, Perry and Harwood (1992) studied the location and source of wheat lipase using an adapted copper soap method and a radioactive assay. The majority of wheat lipase activity was located in the bran component of the grain (75-80%). Lipase catalyses hydrolysis of triglycerides to produce diglycerides and free fatty acids; and monoglycerides to give glycerol and free fatty acids (Kent & Evers, 1994). Tait and Galliard (1988) investigated the effect of changes in lipid composition during storage on the baking quality. There appeared to be a close association between lipase activity and loss of baking performance during storage of wholemeal, with high lipase activity being associated with poor baking performance.

b) Germ

The protein content of mill germ has been reviewed by Pomeranz (1988) and is reported to be in the range of 17.7-35%. Germ protein types, as percentages of the total, are albumin, 30.2%; globulin, 18.9%; prolamin, 14%; glutelin, 0.3-0.37%; and insoluble types, 30.2%.

The lipid content of mill germ, as reviewed by Pomeranz (1988) is given in Table 4. The influence of the method of extraction on the results has been demonstrated.

^aMean values in parentheses

Vitamin E is one of the major vitamins present in wheat germ (Pomeranz, 1988). Values (as total tocopherol) range from 256-500 ppm, averaging 332 ppm. Of the total 49-59% is α -tocopherol, 28-29% β -tocopherol, and 1.5-9% ϵ -tocopherol. Other vitamins present include thiamine, niacin, riboflavin, folic acid and biotin. Germ contains approximately 17% sugars. Other carbohydrates, e.g. starch, come from contaminating endosperm and cellulose and hemicellulose from contaminating bran.

As would be expected from its embryonic nature, the germ contains innumerable Lipoxygenase catalyses the enzymes, including lipoxygenase (Pomeranz, 1988). peroxidation of polyunsaturated free fatty acids to form free radicals and peroxides, which in turn, oxidise glutathione (Lai, Hoseney and Davis, 1989c). Lipoxygenase has a number of effects on wheat flour doughs. It is an effective bleaching agent; a coupled oxidation destroys the yellow pigments in wheat flour. The enzyme also increases the

mixing stability of wheat flour doughs and has been reported to alter rheology to produce a strong dough (Hoseney, 1994). Frazier, Brimblecombe, Daniels and Eggit (1977) investigated the improving effect of lipoxygenase on bread doughs and found that the presence of fat was necessary for the improvement.

c) Endosperm

The starchy endosperm varies in composition from its outer portion, just beneath the aleurone layer, to its centre: There is a decrease in both protein and ash content from the outer portion to near the central portion of the endosperm (Pomeranz, 1988). The aleurone cells have 2% of cellulose, 2% glucomannan, 60% arabinoxylan and 30% ßglucan (Bacic & Stone, 1981ab).

Carbohydrates of the rest of the endosperm consist of starch, sugars, oligosaccharides Starch, the principle constituent of the endosperm, has been and hemicellulose. estimated to constitute 64-74% (14% mb) of milled endosperm with a protein content of 8,3-13% (Pomeranz, 1988).

Among the cereal flours, only wheat flour has the ability to form a strong, cohesive dough that retains gas and produces a light baked product. Wheat proteins, and more specifically the gluten proteins, are believed to be primarily responsible for that uniqueness of wheat. As reviewed by Hoseney (1994), the gluten complex is composed of two main groups of proteins: gliadin (a prolamin) which is responsible for the dough's cohesiveness, and glutenin (a glutelin) which gives dough its property of resistance to Gliadins have an average molecular weight of about 40 000, while the extension. glutenin proteins vary in molecular weight from about 100 000 to several million, averaging about 3 million. The molecular weights of the glutenin subunits vary from less than 16 000 to about 133 000. The HMW subunits of glutenin are important in dough formation. If they are absent, a very weak dough is formed. As reviewed by Schofield & Chen (1995), the HMW subunits cross-link the individual polypeptide chains by the disulphide bonds of the amino acid cysteine. These disulphide bonds play an important role in the formation of a stable dough. In addition, about 35% of the total amino acids of It is believed that hydrophobic gluten proteins have hydrophobic side chains. interactions between gluten proteins also have an influence in stabilising the gluten structure and in the rheological and baking properties of flour (Hoseney, 1994).

The protein content of wheat flour depends greatly on the cultivar type, but Pomeranz (1988) reported it to range from 10.8-16.6%. The total lipid content of milled endosperm is in the range of 0.75-2.16%, averaging 1.08% .

Some of the enzymes found in the starchy endosperm of wheat have been reviewed by Pomeranz (1988) and Hoseney (1994). They include esterase, lipase, α -amylase, β amylase, protease (proteinase and peptidase) and phytase. The α -1,4 glucosidic bonds are broken on a nearly random basis by α -amylase. β -amylase attacks every second α -1,4 glucosidic bond from the non-reducing ends of the polymers. The amylase activity of wheat, barley and rye appears to be much higher than that found for the other cereal grains. Both proteinases and peptidases are found in mature, sound cereals; however, their levels of activity are relatively low. They hydrolyse internal peptide bonds and peptide chains from the end respectively. Phytase is an esterase that hydrolyses phytic acid. Phytic acid is inositol hexaphosphoric acid, and the enzyme converts it to inositol and free orthophosphate (Kent & Evers, 1994). According to Hoseney (1994), about 70-75% of the phosphorus in cereals occurs as phytic acid, which is thought to chelate divalent ions and keep them from being absorbed in the intestinal tract. Thus, the enzyme's activity appears to be important, as it converts a detrimental entity into inositol (a vitamin) and nutrients. Studies have shown that at least part of the phytic acid in wheat flour is hydrolysed during fermentation. The limiting factor appears to be the solubility of the substrate.

MILLING AND BREADBAKING PRACTICES 2.2

$2.2.1$ **Milling practices**

The objective of milling is to make cereals more palatable and thus more desirable as food. Milling generally involves the removal of bran (i.e. the pericarp, seed coat, nucellar epidermis and aleurone layer). The germ, which has relatively high oil content, is also removed in order to prevent rancidity during storage. In addition to making the product more palatable and increasing its ability to store longer, milling often involves some type of constraint with regard to particle size. Cereals can be dry- or wet milled. Dry milling,

which is employed to make flour for breadmaking, attempts to separate the anatomical parts of the grain as cleanly as possible (Hoseney, 1994).

The flour milling process as applied to wheat can be divided into six main parts: 1) Receiving and storing wheat, 2) Cleaning the grain, 3) Tempering or conditioning, 4) Production of flour and by-products, 5) Treatment of flour with additives (not the case in South Africa), and 6) Packaging, storing and shipping the products (Matz, 1989).

Wheat is received and stored in silos. Silos consist of unloading facilities, scales, storage bins, conveying systems, preliminary wheat cleaning equipment, and associated equipment such as exhaust systems, wheat dryers and wheat turning and blending facilities. Incoming wheat is weighed, sampled and immediately analysed for foreign material (such as other seeds, sand, straw, stones, insects and damaged kernels), moisture and protein content, as well as α -amylase activity. The wheat is weighed as received, dumped through a grate to remove coarse foreign material, passed over a magnet, and passed through a preliminary cleaner ("receiving separator") on its way to the storage bin where it is stored according to class, grade and protein content (Bass, 1988), or some "in house" segregation criteria.

Although large variations are found in the type and number of cleaning steps that different mills employ, certain basic steps are necessary. Early in the system, a magnetic separator removes any tramp metal that would be undesirable in the product and to protect machines from damage and reduce sparks that might trigger an explosion. Also early in the cleaning system, a milling separator removes foreign material (e.g. maize, soybeans, mustard seed, rapeseed) that differ in size from the grain being cleaned. Aspiration cleaning is used to remove chaff, dust and shrivelled kernels (Bass, 1988).

From the milling separator, the wheat passes through a destoner which removes materials similar to wheat in shape and size but differ in specific gravity (e.g. mudballs, small stones, glass and nonferrous material). The wheat from the destoner may still contain impurities either slightly longer or shorter than the wheat grains but of similar cross section (e.g. oats, barley, wild buckwheat seeds). Either disc separators or cylinder separators are used to remove these type of impurities (Bass, 1988). Another

important piece of cleaning equipment is the scourer. The grain is scoured against itself or a perforated metal screen to remove adhering dirt. The adhering material loosened by this treatment is removed by aspiration. This machine is useful to remove not only mud, but also smut or rust (Hoseney, 1994).

After cleaning, the wheat is conditioned before milling. Conditioning (or tempering) is the addition of water to wheat through spray nozzles (Bass, 1988) up to a maximum moisture content of 17% (Bushuk & Scanlon, 1993). Soft wheat is usually tempered to 15.0-15.5% moisture, while hard winter or spring wheats are conditioned to 16.5% and durum wheat to even higher moisture levels (Hoseney, 1994). The time given for the water to penetrate the grain also varies with the grain hardness. Soft wheat requires a much shorter temper time than does hard wheat: 4-6 hours as opposed to 10-36 hours. Correct tempering of wheat is essential to ensure maximum milling efficiency and optimum performance in the final product (Bass, 1988). Wheat is tempered to achieve the following objectives: 1) to toughen the bran and thus make it resist being broken into small pieces during milling; 2) to soften the endosperm and make it easier to grind (Hoseney, 1994); 3) to facilitate the physical separation of endosperm from bran; 4) to ensure that all materials leaving the grinding rolls are in optimum condition for sifting; and 5) to ensure that grinding produces the optimum level of damaged starch consistent with the hardness of the wheat and the end use of the flour (Bass, 1988).

The cleaned, conditioned wheat can now be milled. The objective of milling is to break open the grain, scrape off as much endosperm from the bran skin as possible, and gradually grind (reduce) the practically pure endosperm into flour (Bass, 1988). The number of parts of flour by weight produced per 100 parts of wheat milled is known as the flour yield, or percentage extraction rate (Kent & Evers, 1994). The wheat grain contains about 82% of white starchy endosperm which is required for white flour, but it is never possible to separate it exactly from the 18% bran, aleurone and germ. The extraction rate for bread flour in typical South African mills is 76% (Mr. N. Dumas, Mill Manager, Ruto Mills - Personal communication). Colour and ash curves are used to determine the maximum extraction rate that can be used. This is based on the principle that bran has a darker colour and higher mineral content than starchy endosperm. Milling of wheat is carried out on a large number of size reduction (grinding) and separation machines (Bushuk & Scanlon, 1993). For simplification the milling machines

can be divided into three systems: the break rolls and their associated sifters, the scratch system, and the reduction rolls and their sifters (Kent & Evers, 1994). A flow diagram of wheat milling in a typical South African mill is illustrated in Figure 2.

Fig. 2: Flow diagram of wheat milling in a typical South African mill

Break system

The objectives of the break system are to break open the wheat grain and remove starchy endosperm from the grain in pieces as large as possible. The break system consists of four or five breaks. Each break consists of a pair of corrugated rolls rotating in opposite directions. At the nip, where the two rolls approach each other, the two surfaces are going in the same direction. The rolls are generally run at different speeds, with the fast roll going about two and a half times the speed of the slow roll. Thus, in addition to the crushing action as a large particle passes the narrow gap between the two rolls, there is a shearing action because of the speed differential (Hoseney, 1994).

A thin curtain of particles is fed into the nip between the rolls. In the first break the particles are whole grains. The flutes shear open the grain, often along the crease, and unroll the bran coats so that each consists of an irregular, relatively thick layer of endosperm closely pressed to a thin sheet of bran. A small amount of endosperm is detached from the bran coats, mostly in the form of chunks (semolina); small fragments of bran are also broken off, but little flour is made. The first break grind thus consists of The largest are the bran coats, still thickly coated with a mixture of particles. endosperm; the intermediate-sized particles are either semolina, middlings and dunst, or bran snaps; the smallest are flour (break flour). The types of particles are separated from one another according to size by scalping, dusting and grading. The overtails of the wire scalping sieves become the feedstock to the second break. The finer stocks are graded and conveyed to be purified in preparation for the appropriate further treatment on reduction mills, except for the flour, which is a finished product and is retained for blending with other machine flours (Kent & Evers, 1994).

In each grinding (break) stage the bran becomes progressively cleaner until, following the last grind, it can yield no more endosperm through further grinding (Kent & Evers, 1994). It may become a finished by-product, or it may be subjected to treatment in bran finishers to remove final traces of endosperm. For reducing the size of bran flakes, cutting rolls, grinders or bran cutters may be used.

Part of the roller floor and plansifters of a South African mill are shown in Figures 3 and 4 respectively.

LITERATURE REVIEW

Fig. 3: Part of a roller floor in a South African mill

Fig. 4: Plansifters in a South African mill

Scratch system

The scratch system separates particles of bran and endosperm which are still stuck together after passing through the break system. These particles are too small to have overtailed to the next break, but are not clean enough to go to the reduction system. The scratch system consists of two to four grinding stages. The feed to the scratch system contains large particles of semolina and pieces of bran with attached endosperm, but particles are much smaller and cubical than break feeds. The objective are to scrape endosperm from the bran without undue production of fine stock, and to reduce oversize semolina. The grind from the scratch rolls is scalped, dressed, graded and purified in much the same way as that from the breaks, and fractions dispatched respectively to to subsequent grinding stages, to offals, to flour (as end product) or to the reduction system (Kent & Evers, 1994).

Reduction system

The reduction system is primarily concerned with the gradual reduction of the pieces of endosperm into particles of flour size by grinding on successive smooth roll stands (Bushuk & Scanlon, 1993). It consists of 8-16 grinding stages, interspersed with siftings for removal of the reduction flour made by each preceding grind, and a coarse (tailings) fraction made from some grinds. The feeds to the first two rolls, inevitable contain a few particles of bran. The feeds also contain particles of germ. By carefully controlling the roll pressure, these can be flattened without fragmentation, and subsequently sifted off as coarse "cuts" from the grinds (Kent & Evers, 1994).

In South African mills, all the bran fractions collected from the different mill streams are collected in one stream and then sieved to produce three sizes: Digestive bran (largest), Select bran, and Pollard (smallest) (N. Dumas - Personal communication). The epicarp hairs are also collected in a separate mill stream. Pollard is regarded as an offal product and is not analysed routinely for quality. The properties of the Digestive and Select bran are given in Table 5.

Properties of Digestive and Select Bran produced at Ruto Mills, Table 5: Pretoria, South Africa

In South Africa, brown flours are produced by adding Select bran (12%) back to white flour (N. Dumas - Personal communication). In other countries, bran is not separated from the white flour, and different terms are used to describe different flours (Kent & Evers, 1994). In the U.K., wholemeal flour, by definition, must contain all the products of milling of cleaned wheat, i.e. 100% extraction rate. In the U.S.A. the 100% product is known as wholewheat flour. Brown flours can be of extraction rates between 85% and 98%. For milling brown flour, the white flour process may be modified in various ways:

Releases are increased throughout the break system, by narrowing the roll gap, and by using scalping covers of slightly more open mesh.

Operation of the purifiers is altered by adjusting the air valves, altering the sieve clothing and reflowing the cut-offs so that less stock is rejected to wheatfeed, and more goes to the scratch and reduction systems.

The scratch system is extended by the use of additional grinding stages.

The extraction of flour from the reduction system is increased by more selective grinding, by changing some of the smooth grinding rolls to fluted rolls, and by employing additional grinding stages to regrind offally stock that would be rejected as unremunerative in white flour milling.

Additional flour may be obtained, at the sacrifice of good colour, by bringing the wheat onto the first break rolls at 1-1.5% lower moisture content than the optimum for white flour milling.

2.2.2 Breadbaking practices

Bread can be made by many different procedures. All of these processes have a single, common aim, namely to convert wheat flour into a palatable and aerated food product. In achieving this conversion there are a number of common steps which are used.

The mixing of flour, water, yeast, salt and other specified ingredients.

The development of a gluten structure in the dough through the application of energy during mixing.

The incorporation of air bubbles within the dough during mixing.

"Ripening" or "maturing" of the dough where the gluten structure is further developed.

The creation or modification of particular flavour compounds in the dough.

The subdivision of the dough into unit pieces

A preliminary modification of the shape of the divided dough pieces.

A short delay in processing to further modify the physical and rheological properties of the dough pieces.

The shaping of the dough pieces to achieve their required configurations.

The fermentation and expansion of the shaped dough pieces during "proof".

Further expansion of the dough pieces and fixation of the final bread structure during baking (Cauvain, 1998b).

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a) Ingredients

Allthough the word "bread" covers a considerable variety of products, they share the same four basic ingredients:

- 1. Wheat flour, which contains unique proteins which, when hydrated with water, form a viscoelastic protein structure called gluten. Under the influence of mixing and further processing, gluten is capable of forming a bubble structure in the dough which can be inflated by carbon dioxide gas produced by fermentation. Flour also contains starch which is partially hydrated with the dough water. During baking the dough protein coagulates and the hydrated starch partially gelatinises to form the structure of the bread (Brown, 1993). Bran, which is added back to white flour to produce brown bread, causes the dough to absorb more water and depresses the loaf volume of the final baked product (Pomeranz, 1977).
- 2. Water, to hydrate the flour proteins, to be partially absorbed by the flour starch and to form a water phase in the dough in which soluble solids such as sugars, salt and soluble proteins are dissolved and in which the yeast cells are dispersed (Brown, 1993). In South African bakeries, water addition for white bread averages 61% (based on flour weight) and for brown bread 64% (Mr. D.G. Carroll, Technical Services Manager, Albany Bakery - Personal communication).
- Yeast, to aerate the dough by the production of carbon dioxide gas by fermentation. $3₁$ The carbon dioxide goes into solution in the dough/water phase, which, when saturated with carbon dioxide, releases it into gas cells in the dough, which have been formed by mixing and processing. Yeast fermentation also contributes greatly to the flavour of bread. In addition it produces reducing sugars which react with the dough proteins to produce the characteristic brown colour during the baking process (Brown, 1993). In South Africa yeast is added at 1.5-3.0% (based on flour) levels (D.G. Carroll - Personal communication).
- 4. Salt, to flavour the bread, both by its own characteristic flavour and by highlighting other flavour products in the bread. Salt also has a retarding effect on fermentation (Brown, 1993), and strengthens the dough, presumably by shielding charges on the dough proteins (Hoseney, 1994). Levels of salt used in South African bakeries range from 1.8-2.2% (based on flour), averaging 2.0% (D.G. Carroll - Personal communication).

While these are the only four essential ingredients required, other optional ingredients may be added to improve loaf quality or to change specific bread characteristics (Brown, 1993):

- 1. Fats (shortening) change the eating characteristics of bread, giving it a shorter, softer bite and at the same time a modest enhancement of the soft-eating shelf life (Williams & Pullen, 1998). Fats also gives bread with increased loaf volume compared to bread made without shortening (Hoseney, 1994). Hydrogenated palm oil with a slip point between 40 and 45°C, is the fat most commonly used in South African bakeries. Levels used range bewteen 0 and 1% (based on flour) (D.G. Carroll - Personal communication).
- 2. Sugar is added to bread formulas for two reasons. It is a source of fermentable carbohydrate for the yeast, and it provides a sweet taste to the bread (Hoseney, 1994). South African bakeries use brown sugar at 0.5-1% (based on flour) level of addition (D.G. Carroll - Personal communication).
- 3. Soya flour has two beneficial functions, both being due to a particular enzyme This enzyme system, lipoxygenase, system present in the uncooked bean. catalyses the oxidation of unsaturated fats by atmospheric oxygen to lipid peroxidases via a number of intermediate oxidation compounds. Some of these intermediate compounds transfer oxygen from the atmosphere surrounding the dough to the yellow-coloured carotinoid pigment of flour. The oxidised products derived from these pigments are colourless and therefore the first functional effect of soya flour is as a bleaching agent. The second effect, as a mild oxidising agent, improves bubble formation and dough structure (Williams & Pullen, 1998). The average usage of soya flour in South Africa is 0.75% (based on flour) (D.G Carroll -Personal communication).
- 4. Milk or milk solids can be added to improve both nutritional value and crust colour (Brown, 1993).
- 5. Oxidants such as ascorbic acid improve dough strength and result in bread with better loaf volume and texture (Hoseney, 1994). The role of the oxidant is to first oxidise the water-soluble protein -S-H groups in the system. This would benefit the dough structure by preventing them from preferentially reacting with the -S-H groups of the glutenin molecules exposed during the development period. The removal of the water-soluble protein -S-H group from the system causes a shift of the balance of the reactions of glutenin molecules towards the formation of interglutenin -S-S-

bonds rather than glutenin-soluble protein -S-S- bonds, thus producing a more elastic structure (Williams & Pullen, 1998). Ascorbic acid is the most commonly used oxidant in South African bakeries. It is used at levels of 70-80 ppm for white bread and 100 ppm for brown bread. The use of azodicarbonamide is also possible, but because this is a fast acting oxidant, which could damage the dough during any delay that might be experienced on the production floor, it is not a popular choice (D.G. Carroll - Personal communication).

- 6. Emulsifiers, e.g. monoglycerides are added to increase the softness shelf life of bread. Other surfactants, e.g. sodium stearoyl lactylate (SSL), are used as dough strengtheners to allow the dough to withstand the mechanical abuse of processing lines (Hoseney, 1994). When, for example, diacetylated tartaric acid esters of mono- and diglycerides of fatty acids (DATA esters) are incorporated into bread doughs, they bond rapidly and totally to the hydrated gluten strands. The resultant gluten network is not only stronger, but is more extensible and has a more resilient character. This produces a dough which has a gas bubble network with small-sized, strong and extensible gas cell walls, and a baked bread with higher volume and whiter crumb (Williams & Pullen, 1998). The most effective emulsifiers are DATA esters, and in South Africa, they are used at 0.1-0.2% (based on flour) levels. The most commonly used emulsifier in South Africa is SSL, which is used at a 0.5% level of addition. Glycerol monostearate (GMS) (0.1-0.5%) is also used (D.G. Carroll -Personal communication).
- 7. α -Amylases catalyse the breaking of hydrated starch molecules, both amylose and amylopectin, into short-chain unbranched molecules known as dextrins. In. combination with β -amylase, an enzyme which attacks the ends of the amylose and amylopectin chains breaking off individual maltose sugar molecules, and given sufficient time and the right conditions, they are capable of converting starch almost totally to maltose. It is the level of α -amylase which is the rate-determining enzyme in the system. Most wheat flours contain adequate levels of β -amylase, but usually only a low level of natural α -amylase and therefore it has become common practice to make adjustments to the amylase levels through the addition of suitable materials. These include α -amylase from fungal, cereal or bacterial sources. The use of fungal α -amylase reduces the risk of the formation of an excessive level of dextrins in the bread during the later stages of the baking process as compared to

cereal α -amylase because of its lower heat inactivation temperature. Bacterial α amylases are very heat stable and will continue to produce dextrins throughout the entire baking cycle and even during cooling. The resulting bread is very sticky and it may even become impossible to slice the loaf (Williams and Pullen, 1998). The South African baking industry make use of so called enzyme "cocktails" which is a It may include α -amylase, pentosanases, preparation of different enzymes. xylanase and hemicellulase. Depending on the activity of the enzymes, these cocktails are used at levels between 50 and 200 ppm (D.G. Carroll - Personal communication). Pentosanases have been reported to increase gluten coagulation, and xylanase have been shown to increase gluten strength (Qi Si, 1997). Lipase can aso be used to increase loaf volume, crumb structure and crumb softness.

8. Preservatives are added to inhibit the growth of moulds and thermophilic bacteria. Propionates (e.g. calcium- and sodium propionate) provide the most effective protection against mould growth (Williams & Pullen, 1998). In South Africa the most commonly used preservatives are calcium propionate, sodium diacetate and calcium acetate. The legal limit for these preservatives are 0.3% (based on flour) (D.G. Carroll - Personal communication).

b) Breadmaking processes

The production of bread is a complicated series of operations which must be carried out to produce the particular bread product required. The operations involved can be listed in basic terms as follows:

- 1. Selection of flour and other ingredients for the product to be produced.
- 2. Mixing of the ingredients into a dough.
- 3. Maturing of the dough so that it is capable of producing the bread quality required. Maturing may be achieved during mixing of the dough by mechanical or chemical means, or it may be brought about by the fermentation of the dough in bulk for a set period of time.
- 4. Dividing the dough into pieces of required size, molding it to the shape required, placing it in pans and transferring it to the final prover.
- 5. Proving of the dough in a temperature and humidity controlled area until it reaches the proved volume or height required.
- 6. Baking, depanning and cooling of the loaves (Brown, 1993).

Dough development

If flour, yeast, water and salt are mixed into a dough and processed immediately, the resultant bread will be of poor volume and with an uneven, dense crumb cell structure. This is because the dough protein has not been developed or matured correctly to allow it to retain the gas produced by the yeast and expand. A dough development process is any method of mixing and handling of the dough which gives it the following properties when it is ready for dividing and processing into the required dough shapes:

- 1. The protein structure of the dough should be in a condition which will withstand the stresses of dividing and molding and be capable of being formed into the final shape without tearing.
- 2. The dough after molding should contain a network of gas cells which have been formed during mixing and subsequent processing. These gas cells will be inflated by carbon dioxide gas during final proof and the early stages of baking.
- 3. The dough protein structure surrounding the gas cells should be capable of retaining the gas produced by the yeast during final proof and the early stages of baking and expand evenly without rupturing.
- 4. The dough must contain active yeast and sufficient substrate for the yeast to continue producing carbon dioxide during final proof and the early stages of baking (Brown, 1993).

The methods by which dough development is achieved in the bakery have been reviewed by Kulp (1988), Hoseney (1994) and Cauvain (1998b). These methods may be fitted into five broad processing groups:

- 1. Straight dough bulk fermentation
- 2. Sponge and dough
- 3. Liquid ferment
- 4. Rapid processing
- 5. No-time method (continuous processing or mechanical dough development).

Chorleywood Bread Process (CBP)

The system employed in most South African bakeries is a mechanical dough development system of which the CBP is most commonly used by larger bakeries (Kent & Evers, 1994). In mechanical dough development, there is no fermentation period in bulk, and dough development is largely, if not entirely, achieved in the mixing machine.

The changes brought about by bulk fermentation periods are achieved in the mixer through the addition of improvers, extra water and a significant planned level of mechanical energy. According to Cauvain (1998b), the essential features of the CBP are:

Mixing and dough development in a single operation lasting between 2 and 5 minutes at a fixed energy output.

The addition of an oxidising improver.

The inclusion of a high melting point fat, emulsifier or fat and emulsifier combination.

The addition of extra water to adjust dough consistency to be comparable with those attained in bulk fermentation.

The addition of extra yeast to maintain final proof times comparable with those obtained in bulk fermentation.

Rapid development of the dough occurs in the mixer rather than through a prolonged resting period. The protein structure in the dough is modified to improve its ability to stretch and retain gas from yeast fermentation in the prover. The advantages gained by changing from bulk fermentation to the CBP include:

A reduction in processing time.

Space savings from the elimination of bowl of dough at different stages of bulk fermentation.

Improved process control and reduced wastage in the event of plant breakdowns.

More consistent product quality

Financial savings from higher dough yield through addition of extra water and retention of flour solids which are normally fermented away.

Disadvantages include:

Faster working of the dough is required because of the higher dough temperatures used.

In some views, a reduction of bread crumb flavour because of shorter processing times (Cauvain, 1998b).

As the level of energy per kilogram of dough in the mixer increases, so bread volume increases, and with this comes a reduction in cell size and increased uniformity. Optimum energy levels are 11 Watt-hour/kg dough. This high energy input is capable of mechanically breaking the disulphide bonds holding the original protein configurations together since such processes are known to occur in the mechanical modification of other molecules. The effect of mechanical energy might therefore be likened to the effects of natural or chemical reduction and will increase the sites available for oxidation. The input of energy during mixing causes a considerable temperature rise to occur. Final dough temperatures are higher than those with other breadmaking processes and fall in the region of 27-32°C. This gives a dough which is more "relaxed", in other words has less resistance to deformation during molding (Cauvain, 1998b).

A given bread volume can be achieved with a lower flour protein content in the CBP than with bulk fermentation. A standard sandwich loaf can be made with a flour protein content of about 10.5% (14% moisture base). However, not all bread using the CBP benefits from such a low protein content. The quality characteristics demanded in the bread will largely dictate the flour specification and it is common for higher protein contents (12%) to be used for other bread varieties e.g. brown bread (Cauvain, 1998b). This, however, is not common practice in South African bakeries. Brown bread in South Africa is baked with flour specified to have a protein content of no less than 11% protein. In addition, brown bread recipies are formulated with higher levels of oxidants, enzymes and emulsifiers in order to help the gluten carry the bran (D.G. Carroll - Personal communication).

In contrast to the situation in bulk-fermented doughs the cell structure in the final bread does not become finer as a result of processing CBP doughs. In the case of CBP doughs, the final bread crumb cell structure is almost exclusively based on an expanded The creation of bubble version of that created during the initial mixing process. structures in mechanically developed doughs depends on the occlusion and subdivision of air during mixing. The number, sizes and regularity of the gas bubbles depend in part on the mixing action and energy inputs (Cauvain, 1998b).

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No-time doughs with spiral mixers

In many bakeries, especially the smaller ones, the spiral mixer has taken over as the main type of mixer being used. Spiral mixers have higher work input levels compared with traditional low-speed mixers, with accompanying reductions of mixing times to achieve maximum dough development. Although mainly used for no-time doughs some bakers will use short periods of bulk fermentation (20-30 minutes) to assist with dough development after mixing. Some spiral mixers impart sufficient energy to raise dough temperatures above that expected from the ingredients. Final dough temperatures vary widely and practical examples may be found from 21°C to 27°C. For many bakers the advantage of using lower dough temperatures lies in restricting yeast activity which comes with the usually higher levels of added yeast. A counter to this advantage is the reduction in chemical and enzymic activity which will occur at lower dough temperatures with a subsequent reduction in overall dough development (Cauvain, 1998b).

THE INFLUENCE OF BRAN ON DOUGH AND BREAD BAKING 2.3 **PROPERTIES**

2.3.1 Water absorption

On average, water absorption increases linearly with the addition of wheat bran. Pomeranz et al (1977) found that water absorption increased linearly from 64.2% in the control to 68.3% in the sample with 15% bran (Fig. 5).

Fig. 5: Bake water absorption (%) of doughs from wheat flours in which 3, 5, 7, 10 or 15% was replaced by wheat bran (B) (Pomeranz et al, 1977)

Other work showed similar trends (Pomeranz, Shogen & Finney, 1976; Shogren, Pomeranz & Finney, 1981; Lai et al, 1989a; Lai, Hoseney and Davis, 1989b; Young, 1998). The effect of replacing wheat flour with bran on water absorption as found by Pomeranz et al (1976) is shown in Table 6.

Even higher water absorption levels were reported in this study. When the water absorption of the added fibre were computed, it was found that it generally increased as the level of added bran increased within the range of 3 to 15%. The computed increase was 80 to 102% (Pomeranz, et al, 1976). It is possible that because the bran binds a

relatively large amount of water, the gluten is not properly hydrated and developed at normal absorption levels. The use of an inappropriate absorption level therefore results in a reduction in loaf volume (Lai et al, 1989a)

2.3.2 Mixing properties

In general, an increase in bran level is accompanied by an increase in mixing time (Pomeranz et al, 1976; Pomeranz et al, 1977; Shogren et al, 1981; Cheetham, 1997; Young, 1998). Cheetham (1997) found that dough development of mature brown meal is about 1.5 minutes longer than for mature white flour. Mixograms and average mixing times of doughs with different brans as found by Pomeranz et al (1977) are given in Bran did not increase mixing time at 15% Figure 6 and Table 7 respectively. replacement level (with the exception of coarse bran-medium). The mixing time of the control flour was 3 7/8 minutes. Replacing wheat flour with 15% bran had no consistent effect on bake mixing time.

Fig. 6: Mixograms of control flour (A) and control flour in which 15% was replaced by finely ground fine bran (B) (Pomeranz et al, 1977)

Mixing time of flours with different replacements of bran (Pomeranz Table 7: et al, 1977)

2.3.3 Gassing power

To determine whether loaf volume was depressed more than expected by impaired gas formation or gas retention, Pomeranz et al (1977) determined gassing power at 1 hour intervals for 5 hours on 10 and 8.5 g flour doughs and on a dough prepared with 8.5 g flour plus finely ground fine bran. They detected no significant differences in the gassing power the first three hours, but the recorded pressures after 4 and 5 hours are given in Table 8.

Gas pressure of doughs made with straight flour and with the Table 8: addition of bran (Pomeranz et al, 1977)

Bran did not depress gassing power; on the contrary, it increased it considerably. Consequently, loaf-volume-depressing effects of bran seem to result from reduced gas retention rather than reduced gas formation.

To produce a loaf of bread with a light and even crumb structure, the dough must be able to retain gases produced by yeast fermentation as discrete cells for a sufficiently

long period (Bloksma, 1990). The foam structure of a fermenting dough is a dispersion of discrete gas cells in a continuous starch-protein matrix. A possible explanation for a loss of gas retention is that the matrix ruptures during baking (because of a sharp increase in dough viscosity), resulting in the interconnection of adjacent gas cells, converting the foam structure of dough into an open sponge and enabling direct escape of gas (Fig. 7) (Gan et al, 1995).

Fig. 7: The structural transformation of dough as explained by the conventional starch-protein matrix hypothesis (Gan et al, 1995)

The liquid film hypothesis is a revised possible reason for gas loss (Gan et al, 1995). Soon after mixing, the dough consists of discrete gas cells lined with liquid films and embedded in a continuous starch-protein matrix. The matrix fails to enclose the gas cells completely at advanced stages of fermentation, leaving areas that contain only a thin liquid lamella. Baking increases the rate of expansion until the lamellar film is incapable of meeting the demand for new surface area generation, thus converting the foam structure of dough into an open sponge. The loss of gas retention is caused, therefore, by the rupture of the liquid film, not that of the starch-protein matrix (Fig. 8).

Early stages of fermentation

Advanced stages of fermentation t_{Ω} early stages of baking

End of oven spring or baking

Fig. 8: A revised model of dough expansion (Gan et al, 1995)

Czuchajowska and Pomeranz (1993) found that it was possible to compensate, to various degrees, for a volume decrease as a result of poor gas retention, by supplementing dough (both brown and white) with high-quality gluten. Other authors (Gan et al, 1989) also found that the gas-cell walls of gluten-supplemented wholemeal bread appear to be thicker, smoother and more continuous than those from the unfortified flour. They proposed that the gluten supplement may strengthen the gas-cell walls, thereby counteracting the adverse effects of the non-endosperm components.

2.3.4 Loaf volume

When brown bread is produced, a proportion of the flour is replaced by bran (nonendosperm components). The resulting bread has a dense crumb structure and a low specific volume. This cannot be explained solely in terms of the dilution effect of the non-endosperm components on the gluten-forming proteins (Lai et al, 1989b; Rogers & Hoseney, 1982; Pomeranz, 1977). Pomeranz et al (1977) compared the effects of adding bran on loaf volume with the theoretical effect (Fig. 9). The theoretical line was calculated from loaf volume decreases expected from dilution of gluten proteins. Replacing up to 5% of the wheat flour with fibre materials reduced volume by the

expected theoretical amount. At 10% replacements, reductions in loaf volume were The deviation from the theoretical line increased almost greater than expected. exponentially with increase in amount of fibre material.

Fig. 9: Effects on loaf volume decrease of replacing 3, 5, 7, 10 and 15% wheat flour by celluloses (C), wheat bran (B) or oat hulls (H), compared with the theoretical loaf volume decrease (heavy straight line) (Pomeranz et al, 1977)

The precise reason for the decrease in loaf volume, caused by the addition of bran, is not known (Bloksma & Bushuk, 1988). Research suggests that the reduction in loaf volume can be attributed to two possible main effects, namely chemical or physical or a combination of the two.

Chemical effect of bran on loaf volume

In commercial practice, flour is normally stored for short periods only. Loss of baking performance and consequent reduction in loaf volume would not be expected during this When flour is stored for longer periods, however, it shows progressive time. deterioration in baking performance. The actual mechanism of deterioration in baking quality remains unproven, although it is known that the variation in baking performance

during storage is dependent on such factors as moisture content, storage temperature and storage atmosphere (Warwick et al, 1979).

Most of the published information on the effects of storage on the baking properties of wheat concerns white flour, which consists almost entirely of the starchy endosperm. There is little information concerning wholemeal flour although it is widely accepted that wholemeal deteriorates rapidly on storage compared with white flour, and it is considered that this difference is due to the greater amounts of lipid-rich germ and bran tissue in wholemeal (Barnes & Lowy, 1986). Indeed Galliard (1986a) provided evidence that the decreased shelf-life of wholemeal flour, as compared with white flour, is due to the presence of lipid metabolising enzymes present in the bran and germ fractions of the wholemeal. They showed that the oxygen consumption values of aqueous suspensions of wheat wholemeal were substantially higher than those of white flours. The actual values depended upon storage history. The O₂ consumption value of the materials stored for 2-4 weeks at 20°C and 65% R.H., were many-fold higher than those of the same materials from freshly milled grain. The O₂ consumption of mixtures of finelyground (<0.5 mm) bran and germ increased on storage more rapidly than that of bran or The O_2 uptake was due primarily to the oxidation of germ stored separately. unesterified, polyunsaturated fatty acids, catalysed by lipoxygenase that is concentrated in the germ fraction. The increased $O₂$ demand of the stored materials was due to higher levels of polyunsaturated fatty acids, released during storage, by hydrolysis of triacylglycerols, catalysed by lipase that is concentrated in the bran fraction. According to O'Connor et al (1992), 75-80% of wheat lipase activity is located in the bran component of the grain. This work was done after these authors had established that very little lipase activity was found in the soluble fraction of the flour, providing evidence Galliard (1986a) that lipase is membrane-bound (O'Connor & Harwood, 1992). proposed that competition for oxygen between lipid oxidation and ascorbic acid oxidation in wholemeal dough might affect the improving action of the ascorbic acid and ultimately influence baking performance and loaf volume.

Further studies by Galliard (1986b) showed that the incipient rancidity of wholemeal flour, measured as the rate of oxygen uptake by aqueous suspensions, increased linearly with storage at 20°C and was correlated closely with increases in unesterified fatty acids. Fine milling of the bran, germ and blends of the two increased the rate of

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deterioration and enhanced the synergistic effect between bran and germ on both O₂ uptake and fatty acid increases. Lipid analysis of bran-germ blends supported his conclusions that, during storage, there is a relatively slow (over several weeks) release of fatty acids, catalysed by a lipolytic enzyme in the bran component of wholemeal, but that lipid oxidation occurs rapidly (within minutes) when excess water is added, facilitating lipoxygenase-catalysed peroxidation of polyunsaturated fatty acids.

The theories obtained in the above investigations led to the study of the effect of changes in lipid composition during wholemeal storage on the baking quality (Tait & Galliard, 1988). A simple and relatively rapid method for measuring the lipase activity of wholemeal flour was developed. It involved the measurement of the increase in fatty acid content observed during incubation of unhydrated wholemeal at elevated temperatures. The lipase activities of the different samples of finely-milled wholemeal showed a high correlation with their known rates of deterioration. There appeared to be a close association between lipase activity and loss of baking performance during storage of wholemeal, with high lipase activity being associated with poor baking performance. Storage of wholemeal at 20°C for 23 weeks had a deleterious effect on baking quality (as measured by loaf volume and crumb texture). Interchanging the lipids between freshly-milled and stored wholemeals showed that the extracted lipid fraction from the stored wholemeal could account totally for the decreases in baking quality observed when using stored wholemeal flour. Other authors have also found that wheat fatty acids reduced loaf volumes (Bell et al, 1979). In addition, Warwick and Shearer (1982) found that gluten formation is inhibited by free fatty acids.

Galliard and Gallagher (1988) looked at the effect of bran particle size on deterioration rate (measured as O_2 uptake capacity) baking performance. They found that the rate of deterioration increased as the particle size decreased (Fig. 10).

Fig. 10: The effect of bran particle size on the rate of deterioration (measured as O₂ uptake capacity) of wheat bran during storage at 20 \degree C. \Box , Bran as received (53% were 1-2 mm); \triangle , bran < 0.5 mm; \angle , bran = 0.5-1 mm; \triangle , bran = 1-2 mm; i, bran > 2 mm (Galliard & Gallagher, 1988)

Fine milling of 20 and -20°C-stored bran immediately before blending and baking had no effect on the baking performance when compared with blends containing untreated bran. However, when the bran was finely milled on receipt and then stored at 20 or -20°C for one month before blending with flour, the resultant loaf volumes were lower than the equivalent values with untreated bran or with bran milled immediately before use.

Nelles et al (1998) also found that a wet heat treatment of bran, which practically inactivated lipase and lipoxygenase activities, resulted in higher loaf volumes. From all these studies it can be concluded that the reduction in baking performance may be due mainly to the presence of lipid metabolising enzymes present in bran and germ fractions. Hydrolysis of triacylglycerols, by lipase present in the bran, leads to the accumulation of predominantly polyunsaturated fatty acids which are oxidised by lipoxygenase (present in the germ) when the flour and bran are subsequently hydrated (Tait & Galliard, 1988). This results in a poorer baking quality and a decrease in loaf volume.

Another chemical constituent which may also be responsible for a reduction in loaf volume, is the tripeptide glutathione (y-glutamylcysteinylglycine; GSH) (Sullivan et al, 1936; Hullet & Stern, 1941; Schofield & Chen, 1995; Chen & Schofield, 1996) which is present in substantial amounts the germ (Bloksma & Bushuk, 1988). The role of protein sulphydryl (SH) and disulphide (SS) groups in breadmaking is well established. The SS bonds are important because they form cross-links between polypeptide chains and because, by reacting with SH groups, they can interchange (Fig. 11) (Pomeranz, 1988).

Fig. 11: Thiol-disulphide interchange reactions in the protein network of dough (Bloksma & Bushuk, 1988)

Numerous low molecular weight SH compounds are present in flour and they could play important roles in determining dough properties. At high concentration they may cause a net reduction of protein SS bonds through two consecutive SH/SS interchange reactions, themselves becoming oxidised disulphides (Fig. 12). At lower concentrations, a single SH/SS interchange reaction may cleave a protein disulphide bond. The products are a mixed disulphide, comprising the protein and the low molecular weight SH compound, and a free protein SH group (Fig. 12(a)) (Schofield & Chen, 1995).

Fig. 12: Schematic representation of protein disuplhide bond cleavage by two consecutive SH-SS interchange reactions (Schofield & Chen, 1995)

Of particular interest in the context of the polymeric structure of glutenin and the redox reactions that might affect it, has been the possible role of the GSH (Schofield & Chen, 1995). GSH may cause a reduction in the cohesive molecular forces within the dough by permanent cleavage, or by catalysing disulphide interchange (Ewart, 1985). Oxidised glutathione (GSSG) has been reported to affect dough in the same direction as does the reduced form (GSH), although to a lesser extent. This can be explained by a series of interchange reactions:

> $GSSG + XSH \rightarrow GSSX + GSH$ PrSSPr + GSH → PrSSG + PrSH $GSSX + PrSH \rightarrow PrSSG + XSH$

Where Pr = protein, GSH = reduced glutathione, GSSG = oxidised glutathione, $PrSSG$ = protein-glutathione mixed disulphides and XSH = an arbitrary thiol compound. In this series of interchange reactions, rheoligically ineffective mixed disuplhides are formed at the expense of effective cross-links between protein chains (Bloksma & Bushuk, 1988). This may ultimately lead to a reduction in baking performance and loaf volume. Indeed, Chen and Schofield (1996) found that during short-term storage of white wheat flour, GSH levels decreased with a concomitant increase in flour breadmaking performance during the first 10 days of storage. Thereafter, GSH levels remained constant up to 40 days of storage, as did flour breadmaking performance. Nelles et al (1998) also found

that a wet oxidation treatment of bran, which lowered the levels of potentially oxidisable substances such as GSH, resulted in higher loaf volumes.

Physical effect of bran on loaf volume

Gan et al (1989) studied the crumb microstructure of wholemeal bread using SEM. This technique revealed that the wholemeal crumb, which contrasts markedly with that of white bread, is coarse and consists of thick gas-cell walls and a discontinuous gluten-It is characterised by the presence of non-endosperm protein matrix (Fig. 13). components of the wheat caryposis, including bran, epicarp hairs and particles rich in The non-endosperm components, notably the epicarp hairs, aleurone-pericarp. appeared to play a prominent role in disrupting the gluten-protein matrix. Epicarp hairs are seen to be either closely aligned with the surface of the gas-cell wall and or protruding at various angles from the gluten-protein matrix. In some cases, a single hair appears to puncture the surface of the gas-cell wall leaving a small fraction of the protein matrix at the tip of the hair (Fig. 14). Removing the epicarp hairs by pearling significantly improved the baking performance of the resulting flour. The flour from pearled wheat increased loaf volume with 8.5% when compared with wholemeal bread. Pearlings, in which most of the epicarp hairs present, detrimentally affected the baking performance when added back to flour milled from pearled wheat (Gan et al, 1989).

Fig. 13: Scanning electron micrographs of white bread showing (a) thin gas-cell walls; (b) small pores (SP) in the gas-cell wall; and whole meal bread showing (c) epicarp hairs (EH); (d) epicarp hairs (EH), aleurone layer (AL) and bran particle (B); (e) disruption of the gluten-protein matrix by a large particle of bran. Bar = $1000 \mu m$ (with four squares), = $100 \mu m$ (with three squares) (Gan et al, 1989)

Fig. 14: Scanning electron micrographs of wholemeal bread showing (a) epicarp hairs and the starch-protein matrix; and (b) a single hair that has punctured the starch-protein matrix and a fraction of protein at the tip of the hair (Gan et al, 1989)

Gan et al (1992) took this investigation further by studying the effect of the outer bran layers of wheat on the breadmaking quality (loaf volume) of flour by the abrasion (pearling) of wheat grains prior to milling. When the outer layers of six wheat grists were removed by pearling, three of the grists produced loaves with significantly higher volumes than the corresponding controls (Fig. 15). For the three grists giving an improvement in breadmaking quality, the average increase in volume was about 7.4%.

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Fig. 15: Volumes of loaves produced from six wheat grists, pre- and post-pearling. *** differences significant at P < 0.001 level; * difference significant at P < 0.05 level. \Box - Controls; \blacksquare - Bread produced from the flours of pearled grists (Gan et al, 1992)

Bran materials collected from a sequential pearling process affected baking performance detrimentally when added to a wholemal control of a unpearled grist and to a composite flour (white flour plus 12.5% bran). The most marked depression in loaf volume was observed when the outermost bran fraction containing the highest concentration of epicarp hairs was incorporated into the breadmaking recipe. Heat treatment of this fraction did not diminish its adverse effect on loaf volume, suggesting that heat sensitive components (e.g. enzymes) are not implicated. Also the differences in original unesterified free fatty acid levels between the white flour, the wholemeal and the flour from pearled wheat grain were found to be small, and were unlikely to account for the observed differences in baking performance. Microscopical examination of the wholemeal dough suggested that non-endosperm components induce serious structural distortion of gas cells and may contribute to the resultant crumb morphology, which is an important element of crumb texture (Gan et al, 1992).

A few authors have looked at the effect of bran particle size on loaf volume depression. Some found that finely ground bran produced higher loaf volumes than coarse bran (Pomeranz et al, 1977; Moder, Finney, Bruinsma, Ponte & Bolte, 1984; Lai et al, 1989b). Özboy and Köksel (1997), however, found that coarse bran showed unexpected strengthening effects on dough rheology and had a lessened deleterious effect on loaf volume. Brans used in these different studies had different proportions of chemical components such as crude fibre, ash and protein. Factors which could influence bran composition include cultivar (Moder et al, 1984; Pomeranz, 1988; Özboy and Köksel, 1997), the mill steam from which the bran has been taken (N Dumas, Ruto Mills -Personal communication, Pomeranz, 1988; Bass, 1988), as well as the milling regime (Oakenfull & Topping, 1987). Thus bran is not a substance of uniform composition and it is possible that the differences in composition could account for the contradicting data on the effects of particle size on loaf volume.

THE INFLUENCE OF BRAN ON BAKED BREAD QUALITIES 2.4

2.4.1 Sensory qualities

Pomeranz et al (1977) found that untrained taste panels consistently indicated that at all replacement levels a detectable change from added bran was not objectionable. The panels also found that bran darkened the crumb and crust and shifted the red colour component in the crust towards grey-red. Moder et al (1984) found that bread made with finely ground red bran had a darker colour than bread made with a coarse red bran.

2.4.2 Bread crumb structure

SEM and light microscopy of white and brown bread done by Pomeranz et al (1977) revealed a major difference between the crumb structure of the control (white) and breads containing fibre (brown). The control bread had a fine crumb structure composed of thin sheets and filaments. Such a structure was essentially absent in the brancontaining breads. To produce a loaf of bread with a light and even crumb texture, the dough must be able to retain the gases produced by yeast fermentation as discrete gas cells for a sufficiently long period (Bloksma, 1990). Gan et al (1995) proposed that loss

of gas retention during baking is caused by the rupture of the liquid lamellar film which lines gas cells in the dough structure.

Moder et al (1984) also found that bread produced from a finely ground bran showed a finer and more uniform crumb grain structure, when compared to bread produced from a coarse bran.

METHODS TO IMPROVE LOAF VOLUME OF BROWN BREAD 2.5

Quite a few methods have been proposed to increase the loaf volume of brown and whole wheat bread. Shogren et al (1981) found that the deleterious effects of up to 15 parts of wheat bran per 85 parts of wheat flour could largely be counteracted with the addition of vital gluten (together with surfactants). Gan et al (1989) also supplemented wholemeal flour with gluten and found an increase in loaf volume. Shogren et al (1981) found that surfactants also improved loaf volume. Similar results were found by Lai et al $(1989a)$.

Lai et al (1989a) also reported that the addition of certain salts (such as phosphates) improved loaf volume. Additionally they found that a reconstituted whole wheat bread with a loaf volume equal to the control was obtained by allowing endogenous lipoxygenase (soaking the non-flour milling fractions) or added lipoxygenase to oxidise the glutathione from the germ and using optimum water absorption.

Moder et al (1984) compared coarse brans and shorts (mixture of germ, aleurone and pericarp layers) in breadmaking to those that were finely ground. Finely ground bran and shorts produced somewhat higher loaf volumes than that for coarse bran and shorts.

Nelles et al (1998) studied three different bran treatments: hydration, wet heat and wet oxidation. They found that all treatments improved brown bread quality significantly, resulting in larger, softer loafs. All treatments resulted in an increase in water absorption of brown bread dough and a decrease in potentially oxidisable substances in bran.

2.6 **SUMMARY**

In South Africa brown bread is produced by addition of bran to white flour. This causes a reduction in loaf volume and bread quality. Although published information on this subject is limited, there is a clear indication that both chemical and physical factors could be responsible for this phenomenon. The negative effect of bran on bread texture and loaf volume can not be fully accounted for on the basis of dilution of the gluten proteins (Pomeranz et al, 1977). Active constituents of bran and germ interact with gluten proteins and other flour constituents to prevent the development of anticipated rheological properties and bread qualities. The key chemical constituents of bran and germ appear to be the lipid and lipolytic enzymes (Galliard, 1986a). Apart from the chemical constituents, bran also exerts a physical effect which adversely affects bread quality (Gan et al, 1992).

Modification of rheological and subsequent quality properties of doughs can result by several mechanisms: inhibition of gluten formation by free fatty acids (Warwick & Shearer, 1982); lipid hydrolysis, by lipase present in the bran, which produces polyunsaturated fatty acids that are oxidised by lipoxygenase (present in the germ) (Tait & Galliard, 1988); modification of the gluten proteins resulting from the reduction of disulplhide bonds by glutathione (Schofield & Chen, 1995; Chen & Schofield, 1996); physical effect of epicarp hairs and bran particles on gluten structure (Gan et al, 1989; Gan et al 1992); as well as the physical effect of different bran types and sizes on loaf volume (Pomeranz, 1977; Moder et al, 1984; Lai et al, 1989b; Özboy & Köksel, 1997).

It is proposed that brown bread quality could be improved if the interplay between numerous chemical and physical effects of bran could be elucidated.

CHAPTER 3: MATERIALS AND METHODS

3.1 **RAW MATERIALS**

3.1.1 Bran

To investigate the effect of bran origin, ten samples of wheat (50 kg) of widely different origins were obtained from Ruto Mills (Pretoria, South Africa) and the South African Wheat Board. Their origin and protein content (N \times 5.7) on a dry weight basis (in brackets) were as follows: 1-UK (10.9%) (soft winter wheat), 2-Germany (12.7%) (soft winter wheat), 3-Canada (14.7%) (hard spring wheat), 4-USA (15.3%) (hard winter wheat), 5- Western Cape (12.8%) (hard winter wheat), 6-Free State (18.1%) (hard winter wheat), 7-Western Cape (12.6%) (hard winter wheat), 8-Free State (14.8%) (hard winter wheat), 9-Free State (16.9%) (hard winter wheat) and 10-Mpumalanga (13.5%) (hard winter wheat). The wheat samples were milled using a Buhler Miag roller mill and the bran was collected in each case. A bran fraction of >0.85 mm - <1.61 mm (approximately the same as that generally used in the South African baking industry) was obtained by sieving. The bran samples were packed in high density polyethylene bags and stored frozen at -20°C.

To investigate the effect of bran type, bran particle size and effect of heat treatment, three different bran fractions, defined by the South African milling industry in terms of sieve size, were collected from different mill streams at Ruto Mills, Pretoria:

- Pollard (specified size < 0.75 mm)
- Select (specified size > 0.75 mm, < 1.8 mm)
- Digestive (specified size > 1.8 mm) \bullet

The bran was packed in high density polyethylene bags, and stored at -20°C.

The protein, starch, moisture, ash, fibre and fat contents of the bran were determined.

3.1.1.1 Protein

Protein was determined by the Kjeldahl AACC method 46-12 (American Association of Cereal Chemists, 1983a) with slight modification. The method was based on the wet combustion of bran by heating it with concentrated sulphuric acid in the presence of a Kjeltab catalyst (potassium sulphate : copper sulphate : selenium at 100:6:1) to effect the reduction of organic nitrogen in the sample to ammonia, which was retained as ammonium sulphate. The digest, having been made alkaline, was distilled to release the ammonia which was trapped in a 4% boric acid solution, and then titrated with 0.1 M HCL, rather than H₂SO₄. The protein content was calculated as follows:

$$
\% \text{Protein} = \frac{T \times N \times 1.4007 \times 5.7}{m}
$$

Where $T =$ titration figure (ml) $m =$ mass sample (g) $N =$ normality of HCI

3.1.1.2 Starch

A modification of the Ewers polarimetric method for starch determination was used. It is based on the principle that starch, which is optically active, rotate polarised light to an extent proportional to its concentration (Pomeranz & Meloan, 1994).

Reagents 0.3084 M HCI 4% Tungstophosphoric acid (4 g/100 ml)

Method

Bran was milled fine enough to go through a 0.8 mm sieve. A 2.5 g sample was weighed into a 100 ml erlenmeyer flask. HCI (50 ml) was added and placed in a boiling water bath for exactly 15 minutes and stirred every 5 minutes. It was then cooled in cold water to approximately 20°C. The mixture was transferred quantitively to a 100 ml volumetric flask. Tungstophosphoric acid (10 ml) was added and made up to 100 ml with distilled water. A double filtration with Whatman No. 4 filterpaper was done until the

filtrate was more than 60 ml. The rotation angle on the polarimeter was read. The starch content was calculated as follows:

 $\% Starch = \frac{\alpha \times 10.94 \times 100}{100 - moisture\%}$

Where α = angle of rotation (degrees)

3.1.1.3 Moisture

The moisture was determined in a Brabender moisture oven (Duisburg, Germany). Approximately 10 g of sample was dried at 131°C for 1 hour, after which the weight difference was determined.

$3.1.1.4$ Ash

A magnesium acetate method, described by Pomeranz and Meloan, (1994), was used to determine ash content.

Reagents

Magnesium acetate (30 g) was dissolved in 150 ml distilled water and made up to 2 litres with 96% ethanol.

Method

Clean silica crucibles were heated in a muffle furnace at 700°C for 30 minutes and allowed to cool in a sealed desiccator containing fresh desiccant. The mass of a crucible was determined to 4 decimal places of a gram. Bran (ca. 5 g) was weighed accurately into the crucible and the mass noted. Magnesium acetate solution (5 ml) was pipetted onto the sample. The sample was ignited and left to burn out. The crucibles were placed in the muffle furnace for 3 hours after the temperature reached 700°C. After removing the crucibles from the oven, they were allowed to cool in a desiccator to The mass of each crucible containing ash was determined room temperature. immediately. A blank sample was also prepared. The % ash was calculated as follows:

MATERIALS AND METHODS

 $\%Ash = \frac{(Mass_{ash} - Blank) \times 100 \times 100}{Mass_{sample} \times (100 - moisture\%)}$

3.1.1.5 Fibre (acid detergent)

Fibre was determined with an acid detergent method.

Reagents

Acid detergent: N-cetyl-N,N,N-trimethyl ammonium bromide (100 g) was dissolved in ca. 3 litres distilled water in a 5 litre beaker. Concentrated sulphuric acid (204 ml) was added very slowly while stirring. The mixture was left overnight, transferred to a 5 litre volumetric flask and made up to volume with distilled water.

Method

Bran (ca. 1 g) was weighed in duplicate into 600 ml beakers. Samples were wetted with decahydronaphthalene (2 ml). Hot acid detergent (200 ml) was added to the sample. It was then boiled under reflux conditions for 1 hour. The crucibles were dried for 1 hour at 105°C, cooled in a desiccator and weighed after 30 minutes. After 1 hour, beakers with samples were removed from hotplates and filtered through crucibles using a water suction pump. The beakers were rinsed three times with ca. 50 ml hot distilled water and filtered through. Fibres were scraped from the sides of the beakers with a spatula, rinsed with acetone and filtered through. The fibre containing crucibles were placed in an oven at 105°C for 1 hour, cooled in a desiccator for 30 minutes and then weighed.

The % fibre was determined as follows:

 $\%Fibre = \frac{(M_{cf} + M_c) \times 10000}{M_s \times (100 - moisture\%)}$ Where $=$ mass of crucible and fibre M_{cf} $M_{\rm c}$ $=$ mass of crucible $=$ mass of sample $M_{\rm s}$

3.1.1.6 Fat

The AACC method 30-25 (American Association of Cereal Chemists, 1983b) was used to determine the crude fat content of the bran. Fat was extracted with petroleum ether in

Soxhlet extractors. After removing the ether from the samples by evaporation, the remaining fat was weighed. The fat content of the bran was determined as follows:

$$
\% Fat = \frac{M_f \times 100}{M_s}
$$

Where $=$ mass of fat (g) M_f $=$ mass of sample (g) $M_{\rm s}$

3.1.2 Base flour

For investigation into the effect of bran origin, a common base flour of 14.3% protein (N \times 5.7) (dry basis) was used.

For investigation into the effect of bran type, bran particle size and effect of heat treatment, a base flour from the same grist as the bran was collected from Ruto Mills (Pretoria). The protein content ($N \times 5.7$) of the base flour was 13.6% (dry basis).

The moisture content of the grain and flour was determined the same way as for the bran.

The flour was also stored at -20°C.

3.2 **METHODS**

3.2.1 Heat treatment

The bran samples were subjected to a heat treatment to eliminate heat-sensitive components that could be responsible for a decrease in loaf volume (e.g. enzymes) (Gan et. al, 1992). Preliminary heat treatments of the brans were carried out by autoclaving them in moisture-tight vessels at 121°C for 20 minutes, 30 minutes and 1hour. The heat-treated brans were compared with the untreated brans with the use of the following tests:

Lipase activity, to determine whether the heat sensitive components had been eliminated;

Colour (Hunterlab), moisture (oven-method) and size of the bran particles, to determine the effect of the heat treatment on the physical properties of the bran.

The brans used as *heat-treated* brans in further investigations were autoclaved in moisture-tight containers at 121°C for 1.5 hours. The same baking tests were performed using the heat-treated and the untreated brans.

3.2.2 Baking tests

3.2.2.1 Premixes

Formulation

The premix formulations for brown and white bread (control) are given in Tables 9 and 10 respectively.

Table 9: Premix formulation for brown bread

Premix formulation for white bread Table 10:

Preparation

The soya flour, ascorbic acid and amylase were mixed in a Hobart paddle mixer for 10-15 minutes. After addition of the Ca-propionate, it was mixed for another 10 minutes.

The mixture was transferred to a larger paddle mixer. Sugar, salt and fat were added allowing for a 15 minute mixing time between each addition.

The premix was stored in sealed bins at 12°C.

3.2.2.2 Bread formulation

For the investigation into bran type, bran particle size and effect of heat treatment, three levels of bran addition were used (9, 12, and 15% flour weight basis) and for the investigation into the effect of bran origin, a single level (15%) was used. The formulation of the brown and white bread (control) is given in Table 11.

Formulation of white and brown bread with 9, 12, and 15% bran Table 11: addition

^aSalt (2.0%), fat (1.0%), sugar, (0.5%), soya flour (0.5%), preservative (0.15%), ascorbic acid (0.0075%) and fungal α -amylase (0.005% for white bread and 0.008% for brown bread)

*b***Based on flour**

Eased on flour and bran mix

3.2.2.3 Baking procedure

A mechanical dough development baking process was used. The flour and the bran were first mixed together in a Chopin 10 litre mixer for 5 minutes, and the yeast was dissolved in part of the water $(\pm 17^{\circ}C)$. The flour plus bran and premix were put into the dough mixer (Morton Machine Coy, Ltd. Wishaw, Scotland, Ref no. 17822/M, Model NO/O.DUP), and then the water and yeast solution were added. The dough was mixed at high speed (11.0 Wh/kg). Two 900 g pieces of dough were allowed to rest for 10 minutes and were then formed in a dough former (Mono Universal No 71/09835 D. Ayres Jones & Co, Ltd. Swansea, Great Britain). The loaves were placed in greased, open pans, before being proved in a proof oven for 70 minutes at 40°C, 95% RH. The loaves were then baked for 30 minutes at 230°C in a rotary test baking oven (Simon Rotary Test Baking oven, Henry Simon, Ltd. Stockport, Great Britain No 36265). After cooling, loaf volumes (by rape seed displacement) and heights were measured.

The baking tests were repeated six times for the investigation into bran type, bran particle size and effect of heat treatment, and twice for the investigation into bran origin. A white bread control was baked with every batch.

3.2.3 Image analysis

Brans were subjected to IA using a Tracor Northern 8502 Image Analyzer (Tokyo, Japan). Samples were placed on black cardboard. A video camera, positioned over the samples, was directly connected to the main computer of the image analyzer. It was attempted to put as many particles as possible in a single layer in the visible field of view of the camera. The image analyzer then calculated the following parameters of the bran particles: Number of particles in the field, area, external perimeter, roughness, minimum projection, maximum projection, mean projection, standard deviation projection, width and aspect ratio.

3.2.4 Scanning electron microscopy (SEM)

A stub was fitted with double-sided tape and covered with bran in a single layer. Samples were then gold coated and studied using a Hitachi S-450 electron microscope (20 times magnification).

3.2.5 Lipase

The method of Sullivan and Allison Howe (1933) was used. Lipase present in the bran hydrolysed triacetin (added substrate) under standardised conditions into free fatty acids which were determined titrimetrically with standard alkali.

3.2.6 Total reducing substances (TRS)

The AACC method 10-01 for total reducing substances (American Association of Cereal Chemists, 1983c) was used with slight modification. Bran (5g instead of 2 g) was extracted with 50 ml (instead of 25 ml) 10% (w/v) trichloroacetic acid. After centrifugation, 20 ml clear supernatant and 3 ml standardised 0.005 M iodine solution was titrated against 0.005 M sodium thiosulphate, using 1 ml of a 2% starch solution as

indicator. Iodine that was not reduced by reducing substances present in the bran was determined titrimetrically.

3.2.7 CAT scans

To investigate bubble formation during proofing, bread loafs were CAT scanned by a Philips CAT scanner at the Johannesburg General Hospital. A white bread control, together with three brown breads (15% addition of three bran types) were studied. Doughs were developed as described. Four dough pieces (800 g) were placed in open glass loaf pans (235 mm X 130 mm X 70 mm) and taken to Johannesburg General Hospital where they were placed in a proofing oven at 40°C. The relative humidity was regulated by spraying the inside of the oven with water every 5 minutes. The first loaf was scanned after 0 minutes of proofing, the second after 20 minutes, the third after 40 minutes and the last loaf after 60 minutes. At each time interval, four slices were scanned (Fig. 16).

Fig. 16: CAT scan slices of bread

The loafs were scanned with the same settings as a lung:

Slice thickness: 1.5 mm Matrix size: 320

The CAT scans were then image analysed in order to determine bubble area, length, perimeter and roundness.

3.2.8 Sensory evaluation

3.2.8.1 Test objectives

The test objectives were to compare the acceptability of brown bread baked with different bran types and to determine preference by:

- Evaluation of visual appearance, both internally and externally \bullet
- Evaluation of the following sensory quality characteristics on a seven-point scale: \bullet Texture, moistness, graininess, chewiness, taste and overall acceptability

An example of the score sheet is given in Appendix A.

3.2.8.2 Panel

Use was made of five panelists who are experts in the field of bread baking. The panelists were familiarised with the score sheet, as well as with the terms used to describe the various characteristics of the product.

3.2.8.3 Test environment

An air-conditioned panel room with partitioned booths was used. The use of individual booths eliminated any distractions and prevented communication between panelists. The conditions in the testing area were controlled and panelists were supervised throughout the testing period. White light conditions prevailed in order to ensure that the product appearance was not influence by lighting, as proposed by Larmond (1977).

3.2.8.4 Test material

Six different types of brown bread were evaluated. The bran types that were used were as follows: pollard, heat-treated pollard, select, heat-treated select, digestive and heat-

treated digestive bran. All the breads were baked with a 12% level of bran addition to simulate bread baked in industry. The breads were cut into cubes and each panelist received a representative sample of each of the six breads.

3.2.9 Statistical analysis

Analysis of variance was carried out using the Statgraphics programme version 5.0 (Statistical Graphics Corporation, Wiley & Sons, New York). The Least Significant Difference test was used at the 95% significance level.